ICON-SCI

The 8th RMUTP International Conference

on Science, Technology and Innovation for Sustainable Development

Challenges Towards the Digital Society

22-23 June 2017, Thailand

Proceeding Book

http://iconsci.rmutp.ac.th/

Welcome Message

With great pleasure, the Rajamangala University of Technology Phra Nakhon (RMUTP) welcomes you to "The 8th RMUTP International Conference on Science, Technology and Innovation for Sustainable Development: Challenges towards the Digital Society 2017 (8th RMUTP ICON SCi-2017)", organized by RMUTP and held on the 22-23 June, 2017 at the Pullman Bangkok King Power hotel, Bangkok, Thailand. We also welcome participants from overseas to Thailand and look forward to giving you a taste of Thailand's culture.

Our conference provides an outstanding international forum to present and discuss progress in research, development, standards, and applications of the topics related to Science, Technology and Innovation for Sustainable Development.

The 8th RMUTP International Conference will offer high quality activities including research and poster sessions. We feel sure to provide you an engaging environment with an excellent opportunity to exchange new research results, major ideas and start fruitful collaborations. International visitors are also encouraged to experience the Thai culture and attractions around Bangkok. We take this opportunity to thank you for your participation, we hope you enjoy your time and take advantage of our conference. We look forward to seeing you.

Sincerely Yours,

Supatra Kosaiyakanont

Assoc. Prof. Supatra Kosaiyakanont
President of Rajamangala University of Technology Phra Nakhon
Conference Chair, The 8th RMUTP International Conference

ICON-SCI Committee

Advisory Committee

Assoc. Prof. Supatra Kosaiyakanont (RMUTP)

Organizing Committee

Fuangfah Mekkriengkrai (General Chair)
Jirasak Tharajak (RMUTP) (General Co-chair)
Darren Albutt (UK)
Jing Yang (UK)
Naphat Albutt (RMUTP)
Thanapong Sareein (RMUTP)
Paisan Kanthang (RMUTP) (Secretary)

Committee

Jakrapong Kaewkhao(CEGM)
Worawat Meevasana(SUT)
Metini Janyasupab (KMITL)
Nithi atthi (TMEC)
Narit Triamnak (SU)
Putinun Uawongsuwan (KMUTNB)
Theerachai Bongkarn (NU)
Narumon Seeponkai (NU)
Sithipong Mahathanabodee (KMUTNB)
Kham Khanthavivone (NUOL, Laos)
Dwayne Cartmell (OSU, USA)
Kantawong Vuttiyorakulchai (ETH Zurich, SW)
Monamorn Precharattana (MU)
Natthapong Wongdamnern (RMUTS)
Suthisa Leasen (RMUTS)
Sumana Kladsomboon (MU)
Tirawut Worrakitpoonpon (RMUTS)
Natthaphon Raengthon (CU)
Suejit Pechprasarn (Polyu, HK)
Nattha Pensupa (NU)
Titiwat Sungkaworn (uni-wuerzburg, GM)
Yingchun FU (ZU, China)

Organizers

Main Organizer:

Rajamangala University of Technology Phra Nakhon (RMUTP)

Co-Organizers:

Centre for Financial Econometrics, Faculty of Business and Law, Deakin University, Australia

Institute for Innovative Learning, Mahidol University (IIL/MU)

Royal Thai Army

The International Business Machines Corporation (IBM)

General Information

Venue:

Pullman Bangkok King Power Hotel
8-2 Rangnam Road Thanon Phayathai Ratchathewi, Bangkok, 10400
Tel: +66 2 6809999
Fax: +66 2 6809998
e-mail: rsvn@pullmanbangkokkingpower.com
http://www. pullmanbangkokkingpower.com

Registration Desk:
Place:
Pullman Bangkok King Power hotel
Hours:

Thursday June 22, 2017	08:00 am – 08:30 pm
Friday June 23, 2017	09:00 am – 05:00 pm

Information Desk:
Place:
Pullman Bangkok King Power Hotel
Hours:

Thursday June 22, 2017	08:00 am – 08:30 pm
Friday June 23, 2017	09:00 am – 05:00 pm

Presentation Instructions:

Oral presentations are required to be made by PowerPoint, which should be controlled by the speaker. The oral presentation will be 15 min plus 5 min questions. All speakers are required to load and check the files before the presentation. A PC-compatible computer and a LCD projector will be provided.

Poster presentations are to be mounted at the allocated area. The content of the poster should cover titles, objectives, methodology, results discussion and conclusion. The poster board size should not exceed 90 cm width x 120 cm height.

ICON-SCI Awards:

1. ICON-SCI Best Oral Presentation Awards
2. ICON-SCI Best Poster Presentation Awards

Table of Contents

Sustainability Science

Screening of oleaginous yeast for oil production from sugarcane leaves

Ratchana Pranimit[1,a], Somboon Tanasupawat[2,b] and Ancharida Savarajara[1,c *]

[1]Department of Microbiology, Faculty of Science, Chulalongkorn university, Thailand

[2]Department of Biochemistry and Microbiology, Faculty of Pharmaceutical Sciences, Chulalongkorn University, Thailand

Keywords: oleaginous yeast, *Yarrowia*, 26S rDNA gene sequence, xylose

Abstract. Glucose and xylose are major sugar component found in hydrolysate of sugarcane leaves pretreated with diluted sulfuric acid and further hydrolysed by cellulase. Oleaginous yeast which able to utilize both glucose and xylose was screened from 81 soil samples collected from Ranong province, Thailand. Obtained 106 isolated yeasts were tested for their ability to accumulate lipid and assimilate xylose. Nile red staining revealed large intracellular lipid globule in 24 isolates and only 16 isolates could assimilate xylose. Identification of the 16 isolates base on their 26s rDNA sequence (D1/D2) indicated that they were belong to 4 genus and 7 species. Among the 16 yeast strains isolated, 5 strains were oleaginous yeasts. *Yarrowia* sp. NG17 which identified as new species had the highest lipid content (27.93%, w/w dry cell wt).

Introduction

Biodiesel, a renewable energy, which is environmental friendly due to net carbon dioxide and sulfur emission are zero [1].

Originally, it is made from plant oil and waste oil by transesterification with short chain alcohol [2]. The supply of waste oil is not high enough for a high demand of the biodiesel, while plant oil is on food vs. energy confrontation [1]. Microorganisms including bacteria, yeast, fungi and algae accumulate high intracellular lipid in form of triacylglycerol (TAG) as an oil droplet when grown under nitrogen limited condition. Microalgae accumulates high lipid level (50-70% w/w, dry cell wt) but its limitation are large area requirement for cultivation, climate and seasonality dependent and water treatment system required [1,9]. Filamentous fungi takes longer time to grow [10,11]. Bacteria accumulates unsuitable lipid form (i.e. polyhydroxyalkanoates) and generates lipid in the outer membrane which make it difficult to extract [10]. Therefore, yeast is the promising single cell oil producer for biodiesel production. Yeast which accumulates lipid more than 20% w/w (dry cell wt) is defined as oleaginous yeast. *Rhodosporidium* sp., *Rhodotorula* sp. and *Lipomyces* sp. accumulated intracellular lipid over 65% (w/w) [3].

Thailand is the world second largest exporter of sugarcane. There are around 11 million rais of plantation area which produce sugarcane leaves (250 kg/ton/rai) after harvesting [6,7]. The abandance sugarcane leaves are removed by burning. This causes serious air pollution. To reduce the pollution caused by sugarcane leaves and also create value to the sugarcane leaves, Jutakanoke et al. (2012) used the sugarcane leaves as fermentable sugar source for fuel ethanol production by treated with dilute sulfuric acid and further hydrolysed by cellulase [8]. Sugar analysis of the sugarcane leaves hydrolysate founded xylose and glucose at 9.0 g/l and 6.24 g/l, respectively [8].

In this study, we isolated and screened for oleaginous yeast which assimilated xylose. Glucose/xylose mixed sugar in the sugarcane leaves hydrolysate will be used as carbon source to produce yeast oil by the isolated yeast.

Material and methods

Soil samples. Eighty one soil sample were collected form Ranong province, Thailand.

Yeast isolation. Yeast was isolated from the samples by enrichment culture method by inoculating one gram of the sample into 10 ml of YM broth (1% glucose, 0.3% yeast extract, 0.3% malt extract, 0.5% peptone (w/v), pH 5.5) containing 0.025% sodium propionate and 0.0001% chloramphenicol in 15×160 mm test tube and incubated at room temperature for 72 h. Purification was performed by streak plate method using YM agar.

Pre-screening of high-lipid accumulating yeast. All of the isolated yeasts were grown in nitrogen-limited medium (5% glucose, 0.01% yeast extract, 0.1% $(NH_4)_2SO_4$, 0.005% $MgSO_4 \cdot 7H_20$, 0.001% NaCl, 0.001% $CaCl_2$, (w/v), pH 5.5) at 30ºC, 200 rpm for 6 days. High lipid accumulating yeast was screened by staining the obtained culture (10 µl) with 100 µl of Nile red solution (Chem-Impex Co. Ltd, USA) and examined for size and yellow-gold intensity of intracellular oil droplet under 1000X magnification fluorescence microscope (excitation and emission at 470-490 and 520 nm, respectively).

Xylose assimilation determination. The selected high lipid accumulating isolate was streaked on xylose agar medium (1% xylose, 0.67% yeast nitrogen base w/o amino acid, 2% agar (w/v), pH 5.5) and incubated at 30ºC for 2 days. Resultant colony classified as xylose assimilating isolate was further identified and quantitative analyzed for intracellular lipid content.

Yeast identification. Oleaginous yeast isolated was identified based on its 26s rDNA sequence (D1/D2 domain) using NL1 and NL4 primers as described by Kunthiphun et al. (2016) [5].

Quantitative analysis of intracellular lipid content. The isolated xylose assimilating yeast was analyzed for intracellular lipid content by inoculating 5 ml of culture grow in YM broth at 30ºC, 200 rpm (24 h) into 45 ml of YM broth in 250 ml flask and incubated at the same condition for 48 h. Cell pellet collected by centrifugation (4ºC, 8,000 rpm, 10 min) and washed twice with sterile distilled water was then transfered into 50 ml of nitrogen-limit medium in 250 ml flask and incubated at 30ºC, 200 rpm for 6 days. Cells collected by centrifugation were lyophilized and determined for biomass and intracellular lipid content by modified method of Folch [4]. In brief, the lyophilized cells were suspended in chloroform-methanol mixture and sonicated. Liquid phase obtained was mixed with 0.73% NaCl, separated by centrifugation (4ºC, 200 rpm, 10 min) and dried at room temperature.

Results

Isolation and pre-screening for high lipid accumulating yeast. One hundred and six yeasts were isolated. Nile red staining indicated that 24 isolates accumulated high lipid (Fig. 1).

Xylose assimilating yeast isolated. From the 24 high lipid accumulating yeast isolates, 16 isolates could assimilate xylose.

Yeast identification. The 16 xylose assimilating yeasts isolated were identified as *Candida tropicalis* (8 strains), *Candida orthoplosis, Cyberlindnera subsufficients, Cryptococcus humicola, Cryptococcus laurentii, Rhodotorula mucilaginosa, Trichosporon mycotoxinirovans* (2 strains) and *Yarrowia* sp. All strains had 26s rDNA (D1/D2) sequence (99%) similarity to their type strains except *Yarrowia* NG17 (94% similarity). This indicated that the *Yarrowia* NG17 was new species.

Quantitative analysis of intracellular lipid content. Analysis of intracellular lipid content of the 16 xylose assimilating strains revealed that 5 strains were oleaginous yeasts. Lipid content of *C. humicola* NG2, *C. subsufficiens* NG8.2, *R. mucilaginosa* 11-2.3, *T. mycotoxinivorans* 11-12.3 and *Yarrowia* sp. NG17 were 22.74, 20.12, 25.30, 20.78 and 27.93%, w/w dry cell weight, respectively (Table 1).

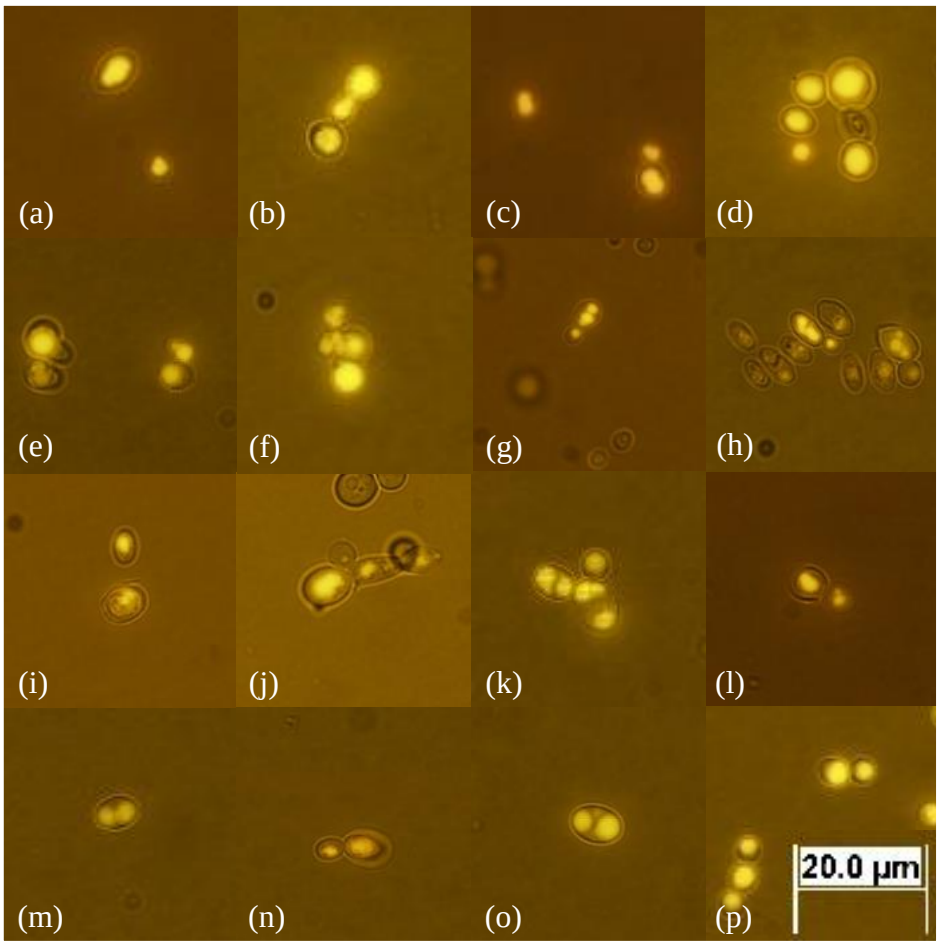


Fig. 1. Nile red staining of the 16 xylose-assimilating yeasts. Strain *Cryptococcus humicola* NG2 (a), *Cryptococcus laurentii* P1 (b), *Cyberlindnera subsufficiens* NG8.2 (c), *Rhodotorula mucilaginosa* 11-2.3 (d), *Trichosporon mycotoxinivorans* (e), *T. mycotoxinivorans* 11-12.3 (f), *Yarrowia* sp. NG17 (g), *Candida tropicalis* 11.20w.4 (h), *C. tropicalis* R1 (i), *C. tropicalis* 1-8 (j), *C. tropicalis* 11-4.1 (k), *C. tropicalis* 11-8.2 (l), *C. tropicalis* 11-17.3 (m), *C. tropicalis* 11-4w.2 (n), *C. tropicalis* 11-20w.2 (o) and *Candida orthopsilosis* 11-11.3 (p).

Table 1. Lipid production of the 16 xylose assimilating yeasts isolated

Strains	**Biomass (g/L)**	**Lipid content (%)**	**Lipid yield (g/L)**
C. humicola NG2	14.70	22.74	3.34
C. laurentii P1	12.80	18.51	2.37
C. subsufficiens NG8.2	6.30	20.12	1.27
R. mucilaginosa 11-2.3	14.20	25.30	3.59
T. mycotoxinivorans NG4.1	16.73	14.98	2.51
T. mycotoxinivorans 11-12.3	16.27	20.78	3.38
Yarrowia sp. NG17	18.13	27.93	5.06
C. tropicalis 11.20w.4	6.69	3.55	0.24
C. tropicalis R1	6.70	7.80	0.52
C. tropicalis 1-8	7.36	9.12	0.34
C. tropicalis 11-4.1	6.69	7.20	0.41
C. tropicalis 11-8.2	10.02	4.46	0.45
C. tropicalis 11-17.3	6.68	4.77	0.32
C. tropicalis 11-4w.2	6.68	5.49	0.37
C. tropicalis 11.20w.2	6.72	6.47	0.43
C. orthopsilosis 11-11.3	9.26	5.16	0.48

Discussion

Oleaginicity of *Trichosporon mycotoxinivorans* NG4.1 and 11-12.3 were different. The oleaginicity of yeast was strain specific and also there was variation of lipid production capability among strains of the same species[1]. The difference of strain oleaginicity might be useful for studying of yeast oil accumulation mechanisms [1,15]. *Trichosporon mycotoxinivorans* had been reported as respiratory pathogen in patients with cystic fibrosis[12]. Therefore, it should not be used as an oil producer for industry.

Genera *Cyberlindnera* and *Yarrowia* are known as oleaginous yeasts [16]. At present, there are 2 species of *Cyberlindnera* reported as oleaginous yeast, *C. saturnus* and *C. jadinii*. Their lipid content were 22% and 25% w/w (dry cell weight), respectively. *Cyberlindnera subsufficiens* was first reported as oleaginous yeast in this study. *Yarrowia lypolytica* has been used as model for studying on yeast oil production because of the oil produced contains linoleic acid as a major component. Higher content of linoleic acid oil indicates high quality of biodiesel product [16]. *Y. lipolytica* Po1g had lipid content (58.5% w/w, dry cell weight) when cultured in sugarcane bagasse hydrolysate that contained 13.59 g/l xylose, 3.98 g/l glucose and 2.78 g/l arabinose [13].

Cryptococcus humicola and *Rhodotorula mucilaginosa* had lipid content at 35.48% and 32.74% w/w (dry cell weight), respectively when cultured in 12% glucose solution without any nitrogen supplementation. *C. humicola* accumulated 40% w/w (dry cell weight) lipid content when cultured in alkaline pretreated corn stover hydrolysate (63.2 g/L glucose and 28.9 g/L xylose) [17].The lipid content of *Cryptococcus humicola* NG2 and *Rhodotorula mucilaginosa* 11-2.3 (in this study) were 22.74% amd 25.30% w/w (dry cell weight), respectively which were lower than previous report

In lipid production medium, *C. humicola* NG2 and *R. mucilaginosa* 11-2.3 might use glucose for growth through energytic pathway before shifting to lipid synthetic pathway after nitrogen was used up.

Conclusion

Five oleaginous yeasts which assimilated xylose, *Cryptococcus humicola* NG2, *Cyberlindnera subsufficiens* NG8.2, *Rhodotorula mucilaginosa* 11-2.3, *Trichosporon mycotoxinivorans* 11-12.3 and *Yarrowia* sp. NG17 were isolated. Their lipid content were 22.74, 20.12, 25.30, 20.78 and 27.93 % w/w (dry cell weight), respectively when cultured in lipid production medium. *Cyberlindnera subsufficiens* was first reported as oleaginous yeast.

Acknowledgements

This research was financial supported by the Graduate School, Chulalongkorn University to commemorate the 72nd anniversary of his Majesty King Bhumibala Aduladej and the National Research University Project, Office of Higher Education Commission (WCU-58-016-EN).

References

[1] I.R. Sitepu, L.A. Garay, R. Sestric, D. Levin, D.E. Block, J.B. German and K.L. Boundy-Mills, Oleaginous yeasts for biodiesel: Current and future trend in biology and production, Biotechnol. Adv. 32 (2014) 1336-1360.

[2] S. Kitcha and B. Cheirsilp, Screening oleaginous yeast and optimization for lipid production using crude glycerol as a carbon source, Energy Procedia. 9 (2011) 274-282.

[3] J. Lin, H. Shen, H. Tan, X. Zhao, S. Wu, C. Hu and Z.K. Zhao, Lipid production by *Lipomyces starkeyi* cells in glucose solution without auxiliary nutrients, J. Biotechnol. 152 (2011) 184-188.

[4] M. Axelsson and F. Gentli, A single step method for rapid extraction of total lipids from green microalgae, Plos One. 9 (2014) 1-6.

[5] S. Kunthiphun, R. Endoh, M. Takashima, M. Ohkuma, S. Tanasupawat and A. Akaracharanya, *Trichosporon heliocopridis* sp. nov., a ureasenegative basidiomycetous yeast associated with dung beetles (*Heliocopris bucephalus* Fabricius), Int. J. Syst. Evol. Microbiol. 66 (2016) 1180-1186.

[6] Information on http://www.ocsb.go.th

[7] B.K. Gullett, A. Touati, J. Huwe and H. Hakk, PCDD and PCDF emission from simulated sugarcane field burning, Environ. Sci. Technol. 40 (2006) 6228-6234.

[8] R. Jutakanoke, N. Leepipatpiboon, V. Tolieng, V. Kitpreechavanich, T. Srinorakutara and A. Akaracharanya, Sugarcane leaves: pretreatment and ethanol fermentation by *Saccharomyces cerevisiae*, Biomass Bioenergy. 39 (2012) 283-289.

[9] Y. Chisti, Biodiesel from microalgae, Biotechnol. Adv. 25 (2007) 294-306.

[10] X. Meng, J. Yang, X. Xu, L. Zhang, Q. Nie and M. Xian, Biodiesel production from oleaginous microorganism, Renew. Energy. 34 (2009) 1-5.

[11] A. Tanimura, M. Takashima, T. Sugita, R. Endoh, M. Kikukawa, S. Yamagushi, E. Sakuradani, J. Ogawa, M. Ohkuma and J. Shima, *Cryptococcus terricola* is a promising oleaginous yeast for biodiesel production from starch through consolidated bioprocessing, Sci. Rep. 4 (2014) 1-6.

[12] P.W. Hickey, D.A. Sutton, A.W. Fothergill, M.G. Rinaldi, B.L. Wickes, H.J. Schmidt and T.J. Walsh, *Trichosporon mycotoxinivorans*, a novel respiratory pathogen in patients with cystic fibrosis, J. Clin. Microbiol. 47 (2009) 3091-3097.

[13] Y.A. Tsigie, C.Y. Wang, C.T. Truong and Y.H. Ju, Lipid production from *Yarrowia lipolytica* Po1g grown in sugarcane bagasse hydrolysate, Bioresour. Technol. 102 (2011) 9216-9222.

[14] J.M. Ageitos, J.A. Vallejo, P. Veiga-Crespo and T.G. Villa, Oily yeasts as oleaginous cell factories, Appl. Microbiol. Biotechnol. 90 (2011) 1219-1227.

[15] I.R. Sitepu, R. Sestric, L. Ignatia, D. Levin, J.B. German, L.A. Gillies, L.A.G. Almada and K.L. Boundy-Mills, Manipulation of culture conditions alters lipid content and fatty acid profiles of a wide variety of known and new oleaginous yeasts species, Bioresour. Technol. 144 (2013) 360-369.

[16] A. Beopoulos, J. Cescut, R. Haddouche, J.L. Uribelarrea, C. Molina-Jouve and J.M. Nicaud, *Yarrowia lipolytica* as a model for bio-oil production, Prog. Lipid. Res. 48 (2009) 375-387.

[17] I.R. Sitepu, M. Jin, J.E. Fernandez, L. da Costa Sousa, V. Balan and K.L. Boundy-Mills, Identification of oleaginous yeast strains able to accumulate high intracellular lipids when cultivated in alkaline pretreated corn stover, Appl. Microbiol. Biotechnol. 98 (2014) 7645-7657.

FACTORS AFFECTING CONDOMINIUM RESIDENTS' PARTICIPATION IN WASTE MANAGEMENT IN BANGKOK

Nattapassorn Saksrisuwan[1,a*], Kampanat Vijitsrikamol[1,b], Sujinna Karnasuta[2,c]
[1]Faculty of Economics, Kasetsart University, Chatuchak, Bangkok, Thailand 10900
[2]Faculty of Environment, Kasetsart University, Chatuchak, Bangkok, Thailand 10900

Keywords: Waste Management, High-rise residential buildings, Condominium, Economic incentives, Binary Logit Model

Abstract

This study was carried out to obtain some crucial information in 1) socio-demographic background of residents who lived in condominiums in Bangkok, Thailand, 2) their attitude towards waste management in their residential building; and 3) factors that may affect residents' participation in waste management. This study differs from other studies; where they focus on low-rise low-density housing while this paper considers high-rise high-density residential buildings. Random samples were drawn from 400 residents who live in condominiums organized by Lumpini Development Public Company Limited in Bangkok by random quota sampling and interview survey. Analytical tools employed the study are comprised of descriptive statistics and binary logit model for analyzing factors that may affect the residents' participation in waste management. The results obtained from the study revealed that the key factors affecting condominium residents' participation comprised of (1) community management and rules, (2) quality of life, and (3) attitude towards condominium waste management system respectively. These research findings are applicable and can assist policymakers and stakeholders, especially Pollution Control Department, Department of Environment and Real Estate Agencies in providing some guidelines in waste management activities in condominiums. Example policies include promoting waste separation through public awareness raising, creating economic incentives and non-monetary rewards for waste management activities.

Introduction

Thailand government has recently established waste management as an important national agenda that governmental municipalities and every citizen should be concerned especially waste management in cities like Bangkok, the capital of Thailand. Due to development and expansion of city's economic growth leading to a rapid increase population, most of residential areas in Bangkok has been fully occupied. As a result of limited horizontal land space in Bangkok, vertically high-rise residential development projects, such as, apartment buildings and condominiums can now be easily spotted in Bangkok. This, of course, leads to an increasing number of high-rise residential units and consequently leads to unforeseen effects in the society. One of them is, of course, an increase in waste amount.

Table 1 shows amount of solid waste collected and disposed in Bangkok during a 5 year-period from 2011 to 2015 [1]. It can be clearly seen from the table that the amount of waste has steadily increased every year.

Table 1 The Amount of solid waste collected and disposed in Bangkok 2011-2015 [1]

Year	Amount of solid waste collected and disposed in Bangkok	
	Tonnes/Year	Tonnes/day
2011	3,264,232	8,943
2012	3,558,020	9,748
2013	3,636,495	9,963
2014	3,628,100	9,940
2015	3,710,955	10,167

Fig 1 shows statistics relationship between the increasing high-rise residential buildings and continuously raise amount of solid waste collected and disposed in Bangkok since 1999 – 2015 [2,3]. It can be clearly seen that the amount of waste is very well collerated to the number of condominiums.

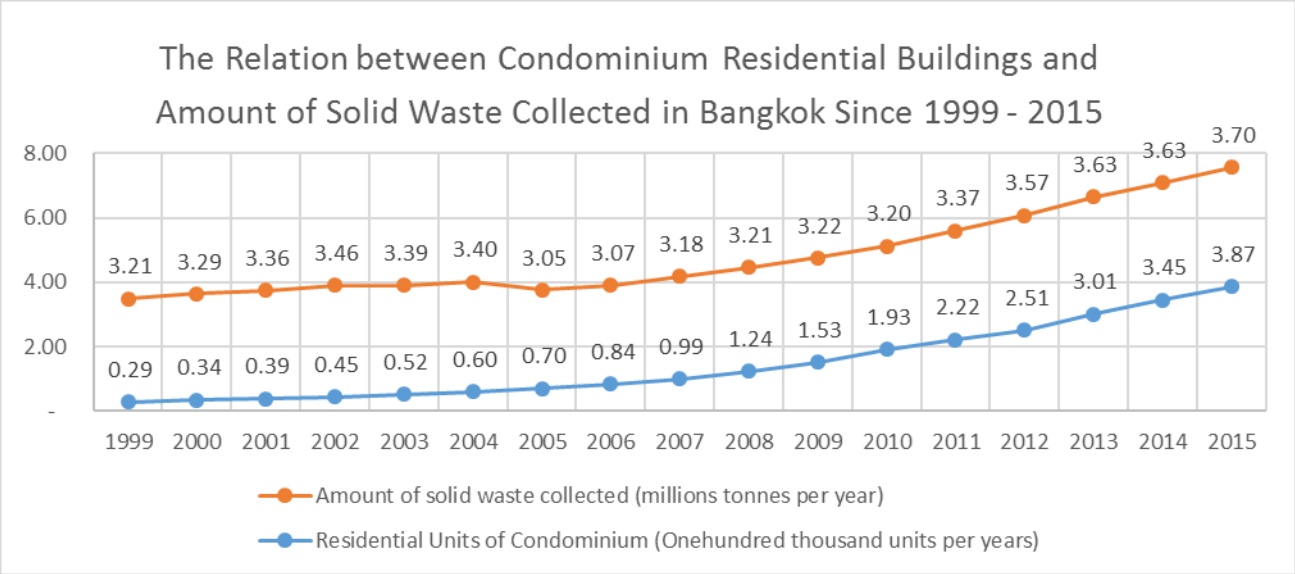


Figure 1: The Relation between Condominium Residential Building and Amount of Solid Waste Collected in Bangkok Since 1999 - 2015) [2,3].

Condominium Governance should be concerned in social responsibility and environment aspect to be green community along with community association for sustainable development. However, waste management in condominium should be participated by residents through activities organized principle of Community Based Management (CBM)[4]. This establishment was a driving force for relevant authorities, including the government, the private sector, or the public, to become more involved in effectively resolving the issue.

Consequently, this study is primarily interested to explore the socio-demographic, residential data waste management behavior, economics incentive environmental features including participation in community association. The purposes of this research were to study:

1. socio-demographic background of residents who lived in condominiums in Bangkok, Thailand
2. their attitude towards waste management in their residential building
3. factors that may affect residents' participation in waste management.

Materials and methods

This study was conducted in March 2017 in condominiums organized and managed by Lumpini Development Public Company Limited. The survey data was drawn by the interviewing 400 residents out of the total 203,500 residents in 118 community based on Bangkok area[5]. Condominium in Bangkok can be classified into 3 categories based on price per sq.m., location and public facility. The three catagories used here were High class, Middle class and Low class. Random data samplings were selected from 10 community with the best practice in waste management by quota random sampling process as shown in Fig 2.

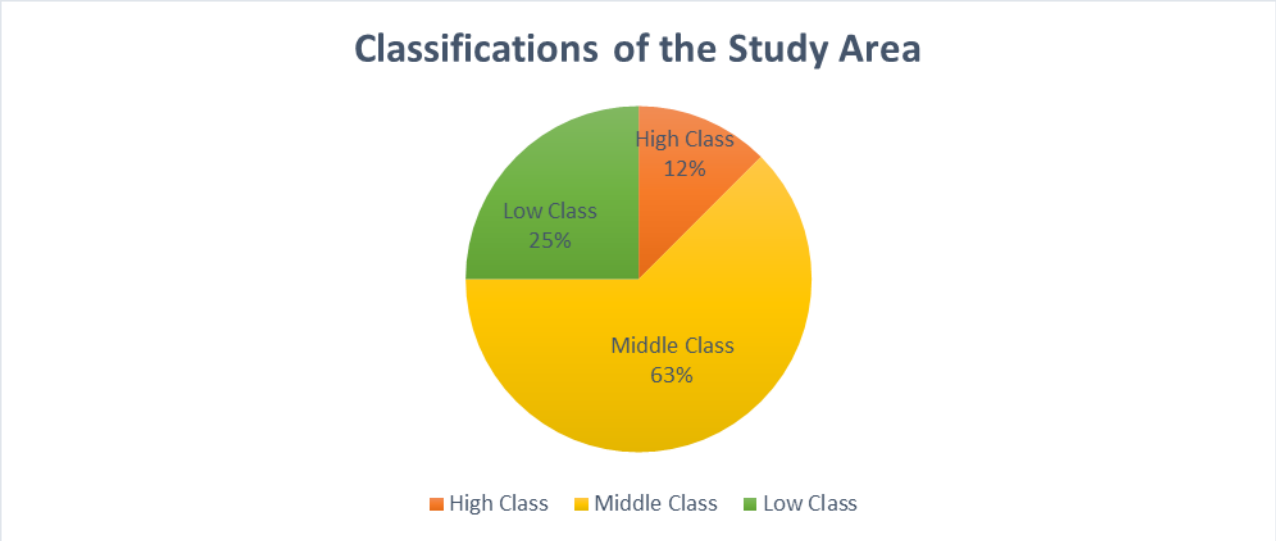


Figure 2 shows Classifications of the Study Area

Basic statistic analyses such as mean, maximum, minimum, and standard deviation were applied to portrait the socio-demographic background and condominium waste management activity. Furthermore, a binary logit model[6] was used to explore the factor affecting condominium resident's participation in waste management. With two choices of dependent variables ($i = 1, j = 0$) a binary logit model gives the choice probability for alternative variable for i, while the independent variables this analysis has a capacity to analyze a mix of all types of predictors (continuous, discrete, and dichotomous) . This model was therefore employed to statistically sign identify significant residents participation factors as shown in the model. The binary logit model can be structurally exhibited in the following mathematics form Eq. 1

$$\Pr_n(i = 1) = \frac{e^{U_{in}}}{1+e^{U_{in}}} = \frac{1}{1+e^{-U_{in}}} \quad (1)$$

Where $Pr_n(i)$ is the probability of participation in waste management i for condominium's resident n

$U_n(i)$ is the utility of participation in waste management i for condominium's resident n

i is waste participation with two choices ($i = 1$ and $j = 0$)

n is condominium's resident ($n = 1,\ldots,400$)

$\mathcal{E}_i$ is the natural logarithm value = 2.7183

Based on the binary logistic regression, random utility model was used to determine the probability that condominium residents would choose to participate in waste management activity. The random utility model[7] is defined as expressed by Eq. 2

$$U_{in} = \beta_0+\beta_1 GEN_{in}+ \beta_2 TOL_{in}+\beta_3 BEH_{in}+\beta_4 WMS_{in}+\beta_5 PROB_{in}+\beta_6 RB_{in}+\beta_7 ECI_i +\beta_8 NMI_{in}+\beta_9 QOL_{in}+\beta_{10} ENV_{in}+\beta_{12} PR_{in}+\beta_{13} LPN_{in}+\mathcal{E}_i \quad (2)$$

where GEN is Gender
TOL is During Time of Resident for living (in year)
BEH is Individual Waste Management Behavior
WMS is Attitude toward Waste Management System
PROB is Problem from Waste Management in area
RB is Recycling Behavior
ECI is Economics Incentive
NMR is Non - monetary Reward
QOL is Awareness of quality of life
ENV is Awareness in an environment
PR is Perception in Condominium News
LPN is Community Governance
$\mathcal{E}$ is Error
$\beta_0 - \beta_{12}$ are the coefficients of the independent variables

Marginal Effects[8] were used to measure changes in probability of participate or not participate in waste management activity due to given changes in the explanatory variables. Marginal effects of continuous variables were calculated at the means of the data. For dummy variable, a value of 0 was used if the mean was less than 0.5 and a value of 1 if the mean was greater than or equal to 0.5.

Results

The results of the study can be divided into 3 parts dealing with overall results described in details as follows:

Part 1 General Socio-demographic Background of the condominium residents

1. Residents' Background

The majority of residents are female (58%) under the average age of 38 years old with Bachelor Degree. The occupations are private company staff, government officer, and business owner, respectively. On the average monthly income is approximately 47,000 bath per person. Half of an interviewee has household size around 2-3 person and come from provincial domicile.

2. Residential Information

This research classify Bangkok condominiums into 3 categories based on price per sq.m., location and public facilities. Classifications are based on the maximum number of condominiums in a project under organized by L.P.N. Development Public Co., Ltd. consist of middle class, low class, and high class with 62.5%, 25.0%, and 12.5% respectively. Mostly residents 73.8% are an owner in their residence. 45.25% has average room size around 26-29 sq.m. An interview respondents have lived in their condominium approximate for 3-5 years with an average of 4 years. More than a half of residents who was interviewed has frequently lived in their habitat everyday.

.

Part 2 Waste Management in Condominium

Awareness of individual recycling behavior evaluated by the principle of 7Rs was comprised of Reduce, Reuse, Recycle, Reject, Repair, Refill, and Return. It was found that the prior issue residents' behavior were realize of separate hazardous waste, separate food waste and decided to buy refilled product respectively. The score level show that most of residents have moderate awareness in waste management. Mainly of resident agreed with current waste management system in condominium. Nevertheless, there was only 10% of residents disagreed with this management,

they complained about the foul-smelling problem, the spread of infectious diseases, and the uncleaned rubbish bins with around 60.11%, 39.89% and 33.88% respectively.

Part 3 Factor Affecting Residents' Participation in Waste Management Activity

From the results show in Table 3, the binary logit model was specified to represent the dichotomous participation in waste management activity, and a logit procedure was used to fit the model.

Marginal effects were calculated to measure the effects of changes in the explanatory variables on the probability of participation. Marginal effects suggest that residents has a good attitude and approved in community governance (LPN), Waste Management System in community (WMS), and Awareness of quality of life (QOL) were 18.55%, 18.02%, 12.83% respectively. They are more likely to participate in waste management activity than those who has worst attitude towards community management. This is obviously because most of residents in high-density would like to live in clean and hygine environment.

Moreover, the marginal effects of interviewees show that non-monetary rewards influence residents participation in waste management activity with percentage of 5.58%

In the contrary, a 1-year increase during time for living (TOL) resulted in a 2.07% decrease in the probability on participating in waste management activity because former residents already ever participated in this activity before.

Table 3 Marginal effect values indicating probability of participation in waste management activity under related factors (with 5% significant level)

Variable	Definition	Mean	Coefficient	Sig.	Marginal Effect
Explanatory Variables					
GEN	Gender	0.58	0.430	0.050**	0.1028
TOL	During Time of Resident for Living (in year)	3.98	-0.087	0.027**	-0.0207
BEH	Individual Waste Management Behavior	3.44	0.040	0.037**	0.0096
WMS	Waste Management System	0.89	0.733	0.034**	0.1802
RB	Recycling Behavior	0.39	0.408	0.090*	0.0960
NMR	Non-monetary rewards	4.01	0.234	0.092*	0.0558
QOL	Quality of life	4.37	0.539	0.035**	0.1283
LPN	Community Governance	0.84	0.757	0.017**	0.1855
C	Constant		-3.025		

Conclusion

With the assumption that residents would maximize expected utilities, a binary logit model was specified to represent the dichotomous participation decision in waste management activity. The probability of participate in waste management activity was assumed to depend on factors such as Community Governance, Attitude toward Waste Management System, Awareness to quality of life, Individual Separation Waste Behavior, and Non-monetary rewards respectively. The estimated model was then used to evaluate the response of an individual having mean characteristics.

Overall, the results indicate that information obtained from residential respondents are more influential in a resident's participate in waste management. In order to make it applicable and can assist policymakers and stakeholders especially Condominium Real Estate Agencies in promoting waste separation through public awareness raising creating economic incentives and non-monetary rewards for waste management activities.

Recommendation

This study aims to analyses factors affecting condominium resident's participation in waste management. waste management activities were depending on the findings of this study would be beneficial for local authority and city planner to integrate waste management activity planning in the high-rise residential buildings in Bangkok. In order to promote residents participation in waste management the related agencies such as Department of Pollution Control, Department of Environment Bangkok, Real Estate Agencies especially Condominium Community should focus on the following issues:

1. Recommendations from research

1.1 Municipal Solid Waste in Bangkok should consider economic incentive in their policies and support recycling subsidy for residential buildings community to resolve pollution problems, promote and maintain environmental quality in term of waste management systems in condominium community. Furthermore, they should increase the efficiency of waste management in high-rise residential buildings as specific law compliance and enforcement in community based on Community Based Management (CBM) to reduce the overview amount of solid waste in Bangkok.

1.2 Thai government should applied the economics aspects pricing system example advance disposal fee with high effective collected and disposed. Subsidies for resource conservation, resource recovery.

1.3 Real Estate Agencies should design more attractive activity to encourage residents willingness to participate in waste management activity by promoting incentive schemes especially non-monetary rewards for example, foodstuffs, daily goods or cash coupons.

1.4 Real Estate Agencies should be organized proper waste management activity for all residents to participate in community association activity include continuously launch campaign. All creates activity has the goal of good environmental management livable communities by collaborating to reduce waste, and improve the environment clean, beautiful, livable, good welfare.

1.5 Widely and continuously launch campaign to promote as well as consciousness of the residents in community in waste management by the media should be varied to the target residents social media and viral public news board in residence.

2. Recommendations for further research

2.1 Determination of waste management in a high-rise residential buildings, from development of the prototype of waste management community (Best Practice) according to the Community Based Solid Waste Management (CBM) should prepare a manual of waste management activities in each method.

2.2 Study what type of reward has stronger motivating effect.

2.3 Could involve similar analysis with impact of participate in others activity.

Acknowledgement

The authors would like to greatly thank L.P.N. Development Public Co., Ltd. for financial support for this study.

References

[1] Department of Environment. Bangkok State of the Environment 2013-2014. Ministry of Natural Resources and Environment.

[2] Pollution Control Department. Thailand State of Pollution Report 2015. Ministry of Natural Resources and Environment.

[3] Department of Land Real Estate Information Center. Ministry of Interior. (2015)

[4] Ahsan T. Zaman AU Household Waste Management in High-Rise Residential Building in Dhaka, Bangladesh: Users'Perspective. Int J Waste Resources vol.4, no. 1, pp. 133 (2014)

[5] L.P.N. Development Public Co., Ltd. Annual Report 2016. (2016)

[6] Greene, W.H. Econometric Analysis, 3rd ed. Upper Saddle River, NJ: Prentice Hall, 1997.

[7] Ben-Akiva, M.E., and S.R. Lerman. Discrete Choice Analysis: Theory and application to Travel Demand. Cambridge, MA: MIT Press, (1985)

[8] Judge, G.G., W.E. Griffiths, R.C. Hill, and T.C.Lee. The Theory and Practice of Econometrics. New York: John Wiley & Sons Inc., (1980).

Treatment of the textile bleaching and dyeing industrial effluent by halophytes in lab-scale constructed wetland and hydroponic experimental units

Kiattipum Kongkeaw[1*], Chachatwal Singhakant[2*] and Supawadee Polprasert[3*]

Department of Environmental Health Sciences, Faculty of Public Health, Mahidol University. Bangkok 10400, Thailand

Abstract

Textile bleaching and dyeing is an important industry in Thailand. This industry generate high volume of wastewater with characteristics of high total dissolve solids (TDS) and undesirable color which usually remained in the factory effluent although that wastewater passed the wastewater treatment processes. Thus this study aimed to find an alternative method to minimize color, TDS, and also chemical oxygen demand (COD) in the factory effluent before release and cause problem to the receiving water bodies. Three types of halophyte, *Cyperus alternifolius, Phragmites australis, and Sesuvium portulacustum*, were experimented to treat the effluent sample with 61 days in lab-scale hydroponic and constructed wetland experimental units. The results showed that the constructed wetland experimental units with halophytes planting could treat the effluent better than the halophytes alone in hydroponic experimental units. Among three types of halophyte in the constructed wetland experimental units, *Cyperus alternifolius* showed the highest removal efficiencies of color (81.46%) and COD (89.29%) to treat the effluent. While TDS cannot be treated in these constructed wetland experimental units. However, this study can suggest that the constructed wetland with *Cyperus alternifolius* planting can be an alternative tertiary treatment process to treat the remaining color and COD in the factory effluent before discharge.

Keywords: halophyte, textile bleaching and dyeing industry, constructed wetland, hydroponic system

1. Introduction

At present, textile bleaching and dyeing industry is an important industry for the economic and development of Thailand from the value of exports, reducing the import of textile raw materials, and creating employment in country [1].This industry, the production is mainly chemical processes which use chemicals and dyes to modify the properties of cloth or fibers and make them colorful. The high volume of water is required to be media in these chemical processes, thus this industry generate high volume of wastewater which can cause the nearby water resource deteriorated and the consequent environmental problems [2]. A case of the released wastewater from a textile factory affecting the nearby agricultural area and fish pond had been reported in Bangkachao district, Samut Prakan province [3,4]. The problem of undesirable color and high total dissolve solids (TDS) in this wastewater can impact to the receiving stream and ecosystem if the wastewater was not treated properly. The TDS are difficult to be removed by conventional wastewater treatment processes and their high concentration can be toxic to the living organism. Besides, about 8 % of the used dyes in dyeing process still remained in the wastewater and the released effluent from the factory [5], their color affects to decrease the dissemination of daylight reducing the overall rate of photosynthesis of algae and other aquatic vegetation [6]. Lower concentrations of dissolved oxygen ultimately hamper the water quality causing a wide array of toxic effects to the aquatic ecosystems. Therefore this industrial wastewater is necessary to be treated properly before discharged to rivers, other water bodies, or municipal sewers.

Halophyte is a group of salt-tolerate plant species, these plants have salt excluded mechanisms which cause they survive in high salinity soil condition [7]. Several studies reported the use of halophytes, such as *Suaeda maritime, Bruguiera gymnorhiza* [8] and *Phramites australis* [9] to treat color, COD and TDS in industrial wastewaters. Thus the halophyte may appropriate to treat the textile bleaching and dyeing industrial wastewater.

This study aimed to determine the potential of halophytes for treatment of the effluent from the textile bleaching and dyeing factory in lab-scale experimental unit by comparing between hydroponic and constructed wetland systems. The parameters of color, COD and TDS were used to calculate the removal efficiencies for comparison.

2. Material and Methods

2.1 Preparation of the halophytes

The halophyte samples including *Cyperus alternifolius*, *Phragmites australis* and *Sesuvium portulacustum* were collected from The King's Royally Initiated Laem Phak Bia Environmental Research and Development Project. The plants were washed by tap water, cut for the 40 cm-stem length, cultivated in plastic bucket with 20% hoagland solution for 1 month before experiment.

2.2 Preparation of the experimental units

2.2.1 Lab-scale constructed wetland system

The experimental units of constructed wetland were prepared by using plastic bucket (29.5 cm diameter and 35.5 cm-height) containing substrate layers (consist of 10 cm of large gravel layer at the bottom, 5 cm of fine gravel layer at the middle, and 10 cm of coarse sand on the top), effluent sample (water level was controlled at 20 cm-height with 6 L of containing effluent sample), and 4 shoots of a halophyte species. A 30 cm of PVC pipe as monitoring pipe was installed at center of the bucket (Fig. 1).

2.2.2Lab-scale hydroponic system

The experimental units of hydroponic system were prepared by using plastic bucket (27 cm-diameter and 29 cm-height) containing 4 planting pots, effluent sample (water level was controlled at 15 cm-height with 6 L of containing effluent sample) and 4 shoots of a halophyte species (Fig. 2).

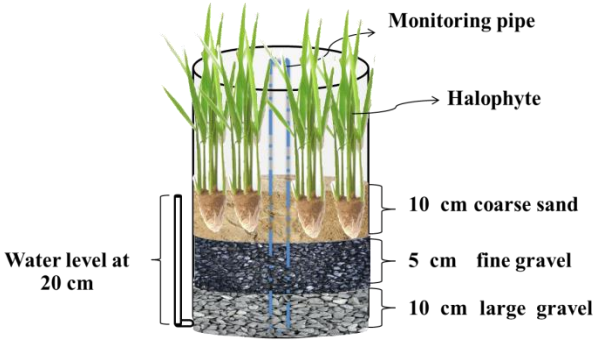


Fig.1 Lab-scale constructed wetland experimental unit with halophyte

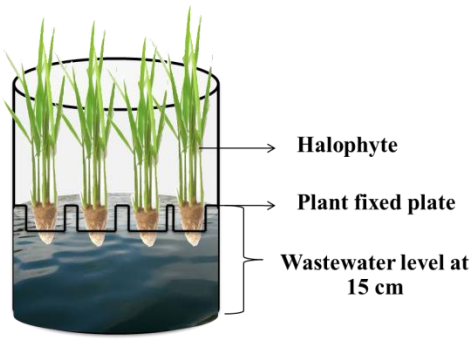


Fig. 2 Lab-scale hydroponic experimental unit with halophyte

2.3Textile bleaching and dyeing industrial effluent sample

The effluent samples were the treated wastewater in polishing pond, collected from a textile bleaching and dyeing factory in Samut Prakarn Province by grab sampling technique, kept in polyethylene gallons, and preserved at below 4 ˚C. The samples were leaved till room temperature before experiment. The characteristics of these effluent samples were appearance of indigo-red color with the color value of 28.3-41.08 S.U. (average of 30.19±3.15 S.U.), COD of 41.7-134.3 mg/L (average of 66.83±26.43 mg/L), TDS of 1700-6190 mg/L (average of 3719.92±1443.99 mg/L), and pH of 6.4 – 8.9 (average of 8.0 ± 0.7)

2.4Comparing the treatment efficiencies of textile bleaching and dyeing industrial effluent between constructed wetland and hydroponic systems with different halophytes

This experiment was conducted at Department of Environmental Health Sciences, Faculty of Public Health, Mahidol University. Total eight groups of experimental units were experimented to treat the effluent sample for 61 days with 3 replications (experimental units) per group. The detail of each group was as following;

Group 1: Constructed wetland with *Cyperus alternifolius* planting (CW_{CA})

Group 2: Constructed wetland with *Phragmites australis* planting (CW_{PA})

Group3: Constructed wetland with *Sesuvium portulacustum* planting (CW_{SM})

Group 4: Constructed wetland with no plant (CW_{NP})

Group 5: Hydroponic system with *Cyperus alternifolius* planting (HP_{CA})

Group 6: Hydroponic system with *Phragmites australis* planting (HP_{PA})

Group 7: Hydroponic system with *Sesuvium portulacustum* planting (HP_{SP})

Group 8: Hydroponic system with no plant (HP_{NP})

Each experimental unit was fed with textile industrial effluent sample every 3 days in the first month and every 7 days until the end of experiment to compensate and maintain the water level. The amounts of feeding effluent sample each experimental unit each time were recorded and used to calculate mass of COD and TDS input. The 150 ml of water samples before and after

treatment were collected from each experimental unit to analyze pH, color, COD, and TDS as the details in Table 1.

Table 1 Analytical method and sampling for wastewater analysis

Parameters	**Analytical methods**
pH	Electrometric
TDS	Electrometric
COD	Close reflux
Color	Space Unit [10]

2.5 Calculation for removal efficiency of color, COD, and TDS

Color (space unit (S.U)),water samples were centrifuged at 1000 rpm for 30 minutes and analyzed by UV-visible spectrophotometer for absorbance at wavelengths in range of 400-700 nm and calculated by using equation (1);

$$\text{Color (S.U.)} = A + B + C \quad (1)$$

where as: A = Area under absorbance curve between wavelength of 400 nm and 500 nm = $\left[\frac{absorbance\,at\,400\,nm + absorbance\,at\,500\,nm}{2}\right] x100$

B = Area under absorbance curve between wavelength of 500 nm and 600 nm = $\left[\frac{absorbance\,at\,500\,nm + absorbance\,at\,600\,nm}{2}\right] x100$

C = Area under absorbance curve between wavelength of 600 nm And 700 nm = $\left[\frac{absorbance\,at\,600\,nm + absorbance\,at\,700\,nm}{2}\right] x100$

-Percentage of color removal efficiency was calculated by using equation (2);

$$\text{Removal efficiency (\%)} = \frac{(C_{in} - C_{eff})}{C_{in}} \times 100 \quad (2)$$

where as;

C_{in} Initial color of effluent sample before treated in experimental unit (S.U.)

C_{eff} Final color of effluent sample after treated in experimental unit (S.U.)

-Removal efficiency of chemical oxygen demand and total dissolved solids was calculated by using equation (3);

$$\text{Removal efficiency (\%)} = \frac{(M_{in} - M_{eff})}{M_{in}} \times 100 \quad (3)$$

where as:

M_{in} = Total mass of COD or TDS in effluent sample before treated in experimental unit (mg/L)

M_{out} = Total mass of COD or TDS in effluent sample after treated in experimental unit (mg/L)

3 Results and discussion

3.1Comparison of the color removal between constructed wetland and hydroponic system with different halophytes

The result of color removal in term of remaining color in effluent sample in constructed wetland units and hydroponic system units were shown in Fig.3 and 4, respectively. These results indicated the high color removal efficiency of the constructed wetland units with halophytes planting. The color removal mechanisms in constructed wetland units included adsorption, accumulation, microorganism and subsequent enzyme mediated degradation [11,12]. While the color removal in hydroponic system units seemed to not occur, indicating, that the halophytes alone in hydroponic system units cannot treat color in the effluent effectively. The increasing color concentration in hydroponic system units was due to evapo-transpiration from the effluent sample and the added compensating effluent during experiment resulting in the increasing concentration.

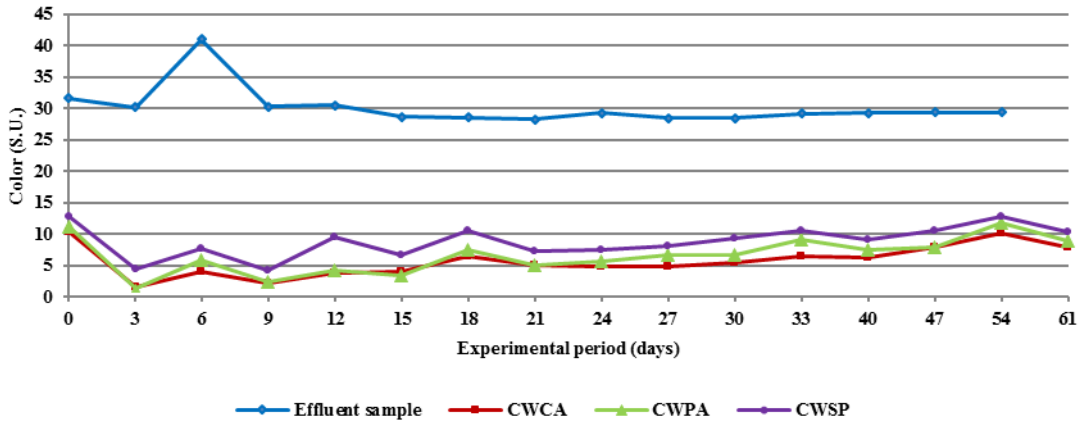


Fig. 3 Removal of color in constructed wetland with *C. alternifolius*, *P. australis* and *S. portulacustum*.

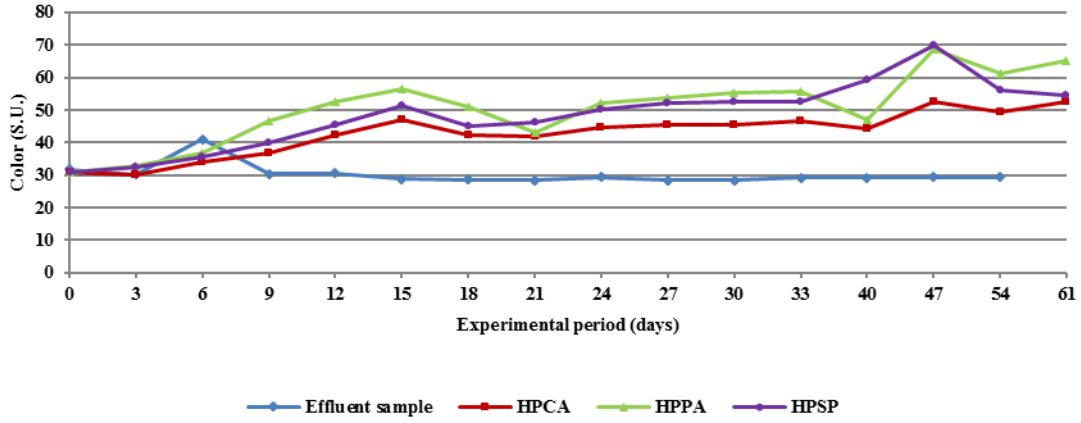


Fig. 4 Removal of color in hydroponic system with *C. alternifolius*, *P. australis* and *S. portulacustum*

Among the group of constructed wetland units, the CW_{CA} had the highest percentage of color removal efficiency (81.46%), following by CW_{PA} (78.64%) and CW_{SP} (70.86%), respectively. T. G. Bulc and A. Ojstrsek studied the use constructed wetland with *P. australis* planting for dry-rich textile wastewater treatment and found that it can reduce color at 90 % [13]. Another study, a vertical flow constructed wetland with *P. australis* was independently developed and employed for the treatment of effluent containing Direct Red 81 and showed color removal of 89% [9].

3.2Comparison of the COD removal between constructed wetland and hydroponic system with different halophytes

The results of COD removal in term of the remaining COD in effluent sample in constructed wetland units and hydroponic system units were shown in Fig. 5 and 6, respectively. These results indicated that the COD removal in constructed wetland units with halophytes planting higher was than the COD removal in hydroponic system units. The COD removal mechanisms in constructed wetland included sedimentation, filtration and degradation by aerobic and anaerobic microorganism [14,15]. However, halophyte alone also can treat COD by uptake and translocation from roots to the shoot parts [16].

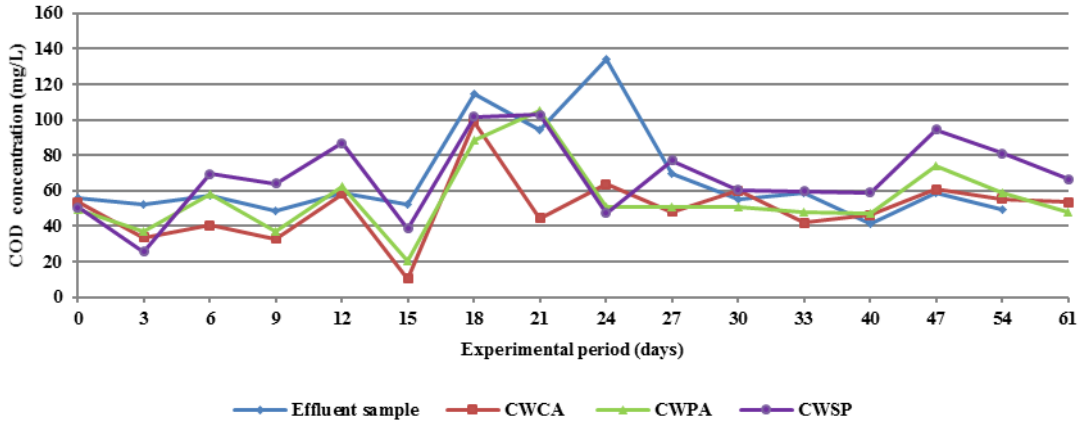


Fig. 5 Removal of COD in constructed wetland with *C. alternifolius*, *P. australis* and *S. portulacustum*

Among the group of constructed wetland units, the CW_{CA} had the highest percentage of COD removal efficiency (90.27 %) following by CW_{PA} (89.29%) and CW_{SP} (87.37%), respectively. A study of the textile effluents treatment in a vertical flow constructed wetland with *P. australis* plating reported the COD removal at 69% [17]. While T. G. Bulc and A. Ojstrsek reported the COD removal at 84% from the dye-rich textile wastewater in their constructed wetland with *P. australis* planting [13].

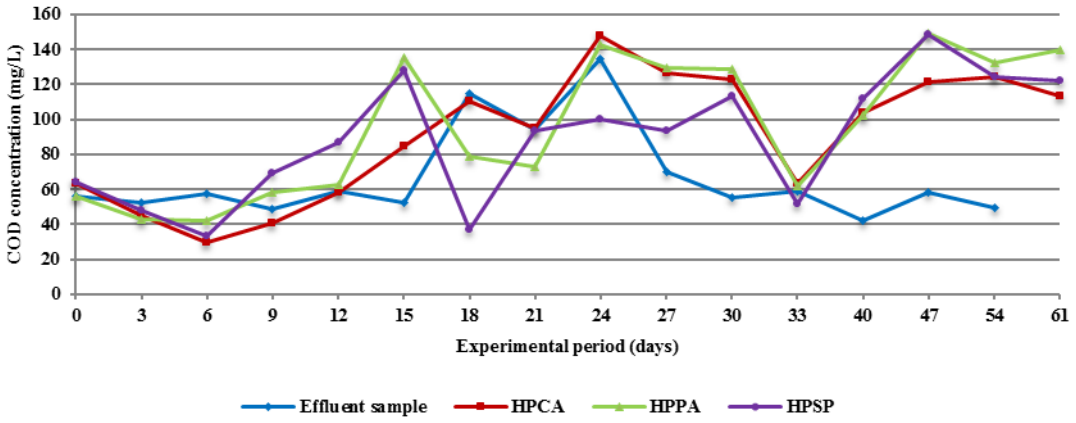


Fig. 6 Removal of COD in hydroponic system with *C. alternifolius*, *P. australis* and *S. portulacustum*

Among the group of hydroponic system units, the HP_{SP} had the highest percentage of COD removal (75.26%) following by HP_{CA} (75.19%) and HP_{PA} (73.81%), respectively. Rahul et al. reported that *Aster amellus Linn*. can remove COD from industrial effluent in the hydroponic system at 60% [18].

3.3Comparison of the TDS removal between constructed wetland and hydroponic system with different halophytes

The results of TDS removal in term of the remaining TDS in effluent sample in constructed wetland units and hydroponic units were shown in Fig. 7 and 8, respectively. The increasing TDS in all experimental units through the experiment was again due to the water evapo-transpiration and the added compensating effluent. These results indicated that both constructed wetland units and hydroponic system units cannot treat TDS in the effluent sample. Similar results Shereen N. Abed et al. studied the use of subsurface flow constructed wetland with *P. australis* planting for effluent treatment and found that TDS increased through the experiment [19]. Another study, the horizontal subsurface flow of constructed wetlands with *Arundo donax* planting were experimented to treat high salinity industrial wastewater and found that the concentrations of TDS were not different between the inlet and outlet [20].

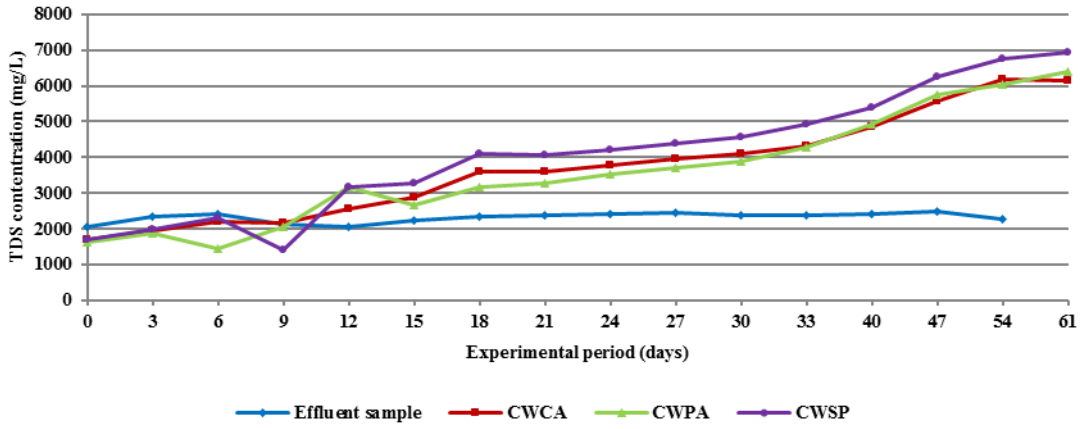


Fig. 7 Removal of TDS in constructed wetland with *C. alternifolius*, *P. australis* and *S. portulacustum*

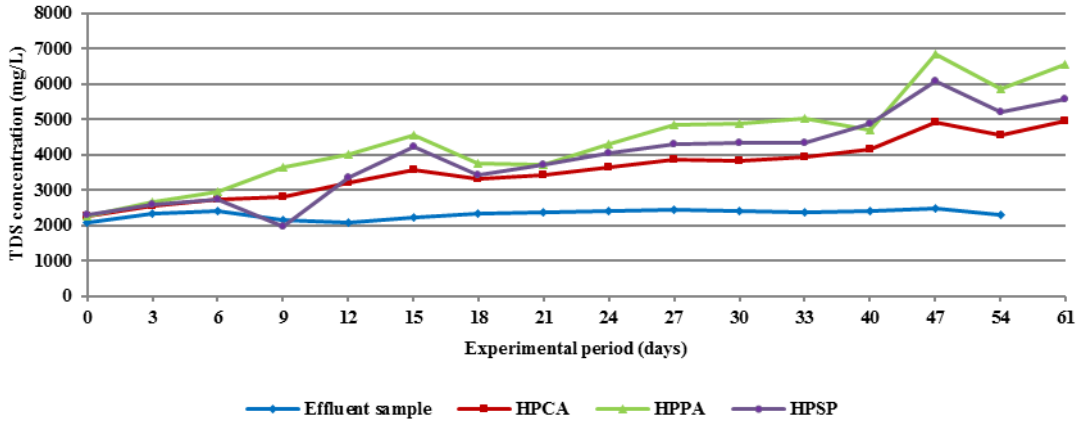


Fig. 8 Removal of TDS in hydroponic system with *C. alternifolius*, *P. australis* and *S. portulacustum*

4. CONCLUSION

The constructed wetland experimental units with halophytes planting could treat the textile bleaching and dyeing industry effluent better than the halophytes alone in hydroponic experimental units. Among three types of halophyte in the constructed wetland experimental units, *Cyperus alternifolius* showed the highest removal efficiencies of color (81.46%) and COD (89.29%) for the effluent treatment. While TDS cannot be treated in these constructed wetland experimental units. However, this study can suggest that the constructed wetland with *Cyperus alternifolius* planting

can be an alternative tertiary treatment process to minimize the remaining color and COD in the factory effluent before discharge.

ACKNOWLEDGMENTS

This research is supported by Department of Environmental Health Sciences, Faculty of Public Health, Mahidol University and The King's Royally Initiated Laem Phak Bia Environmental Research and Development Project.

REFERENCES

[1] R. Sawatdee. Thai Textile Statistic 2014 / 2015: Thailand textile institute; 2015.
[2] Study the problems and solutions to water problems in industrial [Internet]. 2007 [cited 15 november 2015]. Available from: http://www.tpa.or.th/writer/read_this_book_topic.php?bookID=370&pageid=2&read=true&count=true.
[3] Complaint textile bleaching and dyeing industry discharge wastewater was a problem agricultural area [Internet]. 2015 [cited 15 november 2015]. Available from: http://www.matichon.co.th/news_detail.php?newsid=1407908617.
[4] People Mon Canal hard trouble textile bleaching and dyeing industry release to canal [Internet]. 2015 [cited 15 November 2015]. Available from: http://www.thairath.co.th /content/499459.
[5] Kent R. Textile dyeing industry an environmental hazard. Nat sci. 2012;4:22–36.
[6] R.G. Saratale, G.D. Saratale, J.S. Chang, S.P. Govindwar. Bacterial decolorization and degradation of azo dyes: A review. Chemical Engineers. 2011;42:138-157.
[7] Salt tolerance plant and halophyte [Internet]. 2007 [cited 5 may 2015]. Available from: http://www.ldd.go.th/Lddwebsite/web_ord/Technical/pdf/P_Technical03001_3.pdf.
[8] S. Saiyood, A.S.Vangnai, D. Inthorn, P. Thiravetyan. Treatment of total dissolved solids from plastic Industrial Effluent by halophytic Plants. Water, Air and Soil Pollution. 2012;223:3-11.
[9] R. Ferreira J. Duarte, P. Vergine, C. Antunes, F. Freire, S. Martins-Dias. Phragmites sp. physiological changes in a constructed wetland treating an effluent contaminated with a diazo dye (DR81). Environmental Science and Pollution Research. 2014;21(14):9626–9643
[10] M. Tuntoolavest. Handbook of water quality analysis. 5, editor. Bangkok: Chulalongkorn University printing house; 2008.
[11] S. Noonpui, P. Thiravetyan. Treatment of reactive azo dye from textile wastewater by burhead (Echinodorus cordifolius L.) in constructed wetland : effect of molecular size. Environ Sci Health. 2011;46:37–41.
[12] R.V. Khandare, S.P. Govindwar. Phytoremediation of textile dyes and effluents: Current scenario and future prospects. Biotechnology Advances. 2015;33:1697–1714.
[13] T. G. Bulc, A. Ojstrsek. The use of constructed wetland for dye-rich textile wastewater treatment. Hazardous Materials. 2008;155:76-82.
[14] S.X. Zhua, H.L. Gea, Y. Gea, H. Q. Caoa, D. Liua, J. Changa, C.-B. Zhangb, B. J. Gua, S. X. Changc. Effects of plant diversity on nutrient retention and enzyme activities in a full-scale constructed wetland. Bioresource Technology. 2010;101(6):1686-1692.

[15] Y. Wua, A. Chungb, N.F.Y. Tama, N. Pi a, M.H. Wongb. Constructed mangrove wetland as secondary treatment system for municipal wastewater. ecological engineering. 2008;37:137-146.

[16] Govindwar, S., Kagalkar, A., 2010. Phytoremediation Technologies for the Removal of Textile Dyes: An Overview and Future Prospectus. Nova Science Publishers Inc., New York.

[17] L.C. Davies, C.C. Carias, J.M. Novais, S. Martins-Dias. Phytoremediation of textile effluents containing azo dye by using Phragmites australis in a vertical flow intermittent feeding constructed wetland. Ecological Engineering. 2005;25:594-605.

[18] R.V. Khandare , A. N. Kabra, D. P. Tamboli, S. P. Govindwar. The role of Aster amellus Linn. in the degradation of a sulfonated azo dye Remazol Red: A phytoremediation strategy. Chemosphere. 2011;82:1147-1154.

[19] S. N. Abed, N. Mahmoud. Effect of wastewater quality on the performance of constructed wetland in an arid region. The sis: Master degree of Water and Enviromental Engineering. Faculty of Graduate Studies, Birzeit-Palestine University; 2012.

[20] C.S.C. Calheiros, A. Teixeira, C. Pires, A.R. Franco, A.F. Duque, L.F.C. Crispim, S.C. Moura, P.M.L. Castro. Bacterial community dynamics in horizontal flow constructed wetlands with different plants for high salinity industrial wastewater polishing: Water research.2010;44:5032-5038.

Study of drying of kaffir lime leaves using conductive heat transfer

Chayutt Mingqaanchayut[1,a *], Arsit Boonyaprapasorn[2,b] and Weerachai Kaensup[1,c]

[1]Department of Mechanical Engineering, Faculty of Engineering, King Mongkut's University of Technology Thonburi, 126 Pracha-u-Tid Road, Bangmod, Tungkru, Bangkok 10140, Thailand.
[2]Department of Mechanical Engineering, Chulachomklao Royal Military Academy, Nakhon Nayok, 26000, Thailand.

Keywords: heat transfer, convective heat transfer, kaffir lime leaves

Abstract. Kaffir lime (Citrus hystrix D.C.) leaves are widely used as herbal aromatic and food ingredient. Drying process is required to preserve this product. In the present study, drying behavior of kaffir lime leaves was investigated in a conductive heat transfer system under different drying temperatures (100, 110, 120, 130, 140, 150, 160, 170, and 180 °C). The purpose of this study is to determine the optimal condition for kaffir lime leaves drying with heat conduction. The experimental device consists of two temperature-controlled hot aluminum plates and the pneumatic system. In the drying process, the hot plates were driven by the pneumatic system to move in the opposite directions to press a kaffir lime leave from both sides. This mechanism allows the heat to transfer to the leave. The experiment was conducted with variations in temperature and heating duration. Then, subsequent Citronella substances essential for the smell and the leave colors from each condition were measured and compared with those of the fresh leaves. It is evident that the proposed drying process preserves the quality of the dried leave in terms of the smell and color.

Introduction

Kaffir lime (Citrus hystrix D.C.) is a family of citrus fruit and primarily grown in South Asia and Southeast Asia such as Laos, Indonesia, Malaysia, Vietnam and Thailand [15]. Kaffir lime leaves are widely used as aromatic herb to add a distinctive aroma and flavor to food for example soups and curries. For the soupy menu, whole leaves or leaves torn into smaller pieces are added. The popular food especially hot and sour shrimp soup (Tom Yum Kung) have been used for cooking to cover the fishy smell or flavor spicy soup. Kaffir lime leaves can be used fresh or dried as a spice. However, the fresh leaves have very short shelf life approximately 3–4 days, causing a depreciation in market price [13]. So the kaffir lime leaves in the form of dried kaffir lime leaves can solved these problems. Moreover dried kaffir lime leaves are popular as a medical spice and also useful as a preserved food. It is packaged in plastic film and has been exported to many countries such as United States of America, United Kingdom and Australia [13]. Drying is the method for food preservation to inhibit microorganisms and bacteria growth. The kaffir lime leaves contain less moisture content, the growth of those bacteria can be inhibit and hence the food can be stored longer. Drying process is very importance in preserving food products. It is normally tray dried or hot air oven at high temperatures and long drying times to remove the water from the food material. It may cause serious damage in quality of the products in term of flavor, color and nutrients of the dried product [3]. Hence, many researches have been conducted on drying of kaffir lime leaves in various types of drying to overcome these problems. Pongsirikul and Poonlap, 2010 [2] studied the effect of drying methods on the quality of dried Kaffir lime leaves by using microwave vacuum drying, hot air solar drying, hot-air drying and tray drying. It was found that microwave drying

vacuum took minimal times and kept the quality of the color values, Citronella and final moisture content nearby. Among those are hot-air drying, low relative humidity air drying. Tapbuntom and Chinnasarn, 2007 [12] studied the drying kaffir lime leaves by comparison between the one step and two steps drying. Two steps drying at temperature 80 °C and 75 °C for one hour provided the moisture content as low as possible. Follow the drying machine-dried Kaffir lime leaves, heat pump was used in drying by many researches. Shiva et al., 2005 [10] proposed drying lime leaves with heat pump dryer. The best condition for drying was the temperature at 45 °C for 12 hours. Poomsa-ad, Deejing and Wiset, 2011 [5] also used heat pump for drying kaffir lime leaves. It was found that drying by using carbon dioxide as the convection media gave the optimal rate of moisture. The use of nitrogen gas into the medium convection resulted in minimal change color after drying. Tinjun, 2007 [8] studied the drying of kaffir lime leaves by various methods (freeze drying, vacuum microwave drying, oven drying and shade drying). Freeze drying used the long drying time whereas microwave drying in the vacuum condition took minimal time. And oven drying showed minimal color change.

Research drying for the leaves are mostly used the principle of heat transfer by convection, radiation, and how to use the microwave. However, the principle of heat transfer by conduction applied for drying kaffir lime leaves has not yet received much attention from other researchers. The advantages of heat transfer in thermal conductivity compared to convection provided high efficiency drying technique. The thermal conductivity is no need for an intermediary to transfer the heat from the heat source to the material to be dried. Thus the drying can be reduced power losses during the intermediate heat to moist material. Both devices reduced the heat to the drying material. Development of drying technique at high temperature and short drying time with low operating cost becomes the interested research topic in the food industry. The objective of this study was to investigate the drying characteristics of kaffir lime leaves using conductive heat transfer. Apart from that, the drying impact on the color of the final product, retention of Citronella essential oil, and moisture contents were also reported.

Materials and methods

Equipment

This research investigated drying of kaffir lime leaves on conductive heat transfer by creating hot plates for heating to the kaffir lime leaves during atmospheric pressure. Using the hot plate for heat transfer to the kaffir lime leaves, both front and rear side, directly. The conductive heat transfer is cylindrical in shape with heating area approximately 314.28 cm^2. Hot plate can be heated by the heater at power maximum 500 watts as shown in Fig.1 and 2. The hot plate on top and below can be moved to enclose by the pneumatic system's control in which heat transfer timer delay device

Material preparation

Fresh kaffir lime leaves used in this study were purchased from local market in Prachautid, Thrungkru, Thailand. Freshleaves contained about 69% w.b. moisture content. Prior to use, the leaves were washed, drained and selected the closely size and color for drying.

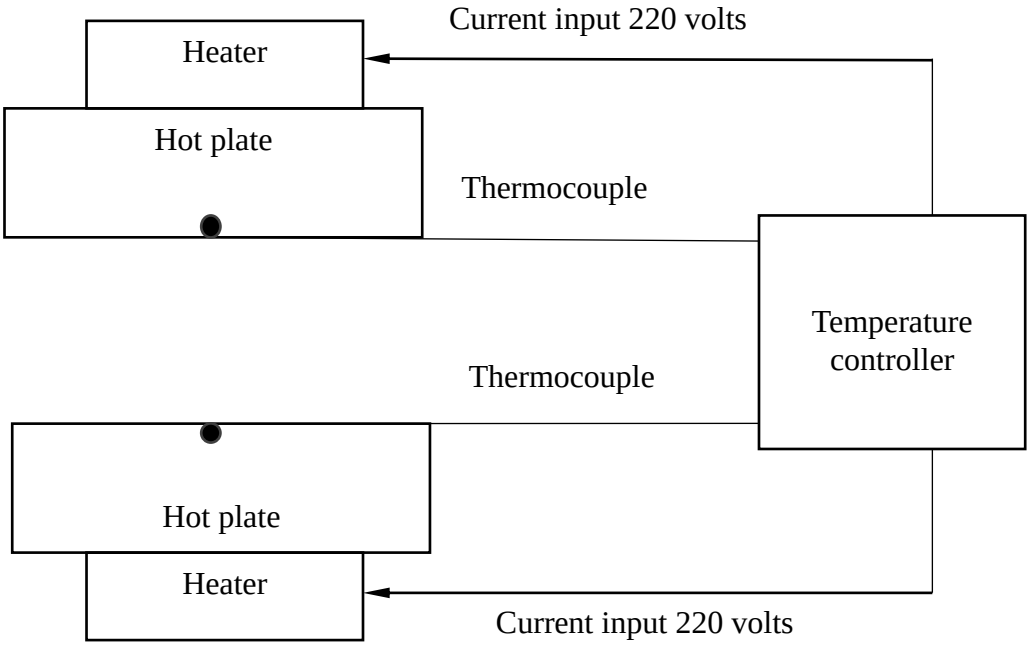


Fig. 1 Thermocouple temperature measurement hot plates surface

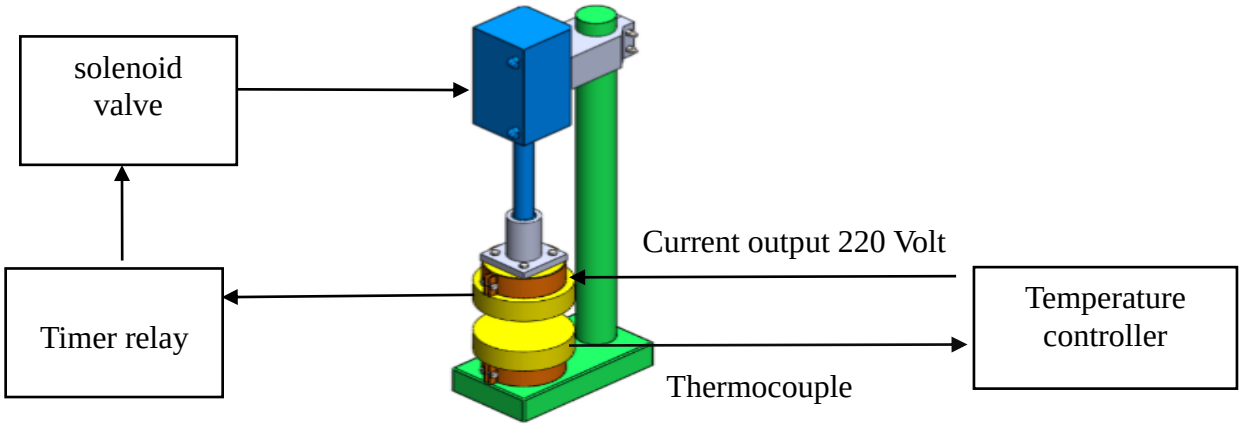


Fig. **2** The experimental set drying, thermal conductivity

Experimental method

The drying tempertures were varied at 100, 110, 120, 130, 140, 150, 160, 170, and 180 °C. In each drying temperature, the kaffir lime leaves were put on the bottom of hot plate and then opened the controller to strat the system. Both hot plates on top and below were moved to enclose for dring the kaffir lime leaves.

Determination of quality properties

Moisture content

Dried kaffir lime leaves were measured the moisture content by hot air drying. The sample was put in the moisture cup which was dried at 105 °C until the constant weight. Then the moisture cup was covered and weight at once. After that the sample was dried in the hot air oven at 105°C for 24

hours. After drying in an hot air oven, the cups were covered immediately and weighed. The moiture contents were calculated in % w.b. (wet basic) and d.b. (dry basic). The equations were shown as follow;

$$M_w = [(w\text{-}d)/w] \times 100 \quad (1)$$

and

$$M_d = [(w\text{-}d)/d] \times 100 \quad (2)$$

where w is the initial sample weight and d is the final sample weight.

Determination of the Citronella essential oil in dried kaffir lime leaves

The dried kaffir lime leaves from the optimal drying condition were selected to extract the Citronella by distillation method. 100 grams of dried kaffir lime leaves were extracted to get the essential oil. Then the oil was tested the amount of Citronella by using Flame ionization detector gas chromatography-type (GC-FID). The specification of column as Rtx - 5MS 30 meter x 0.25. The temperature of the column was started at 70 °C for 2 minutes, then add 4 °C per minute to 220 °C constant temperature for 5 minutes. The flow of helium gas as volume of 2.2 ml per minute. When the trial results, the results do not compare with compounds called Salmonella. (Citronella) standard to determine the number of flavoring agents.

Color value before and after drying

The color analysis of the fresh and dried (both front and rear leaves) kaffir lime leaves were determined by the Hunter Lab system in L, a and b color scales. Parameters L, a and b determine a three-dimensional color space, in which L represents brightness (on alightness–darkness scale), whereas positive and negative a values determine the redness and greenness and positive and negative b values determine yellowness and blueness, respectively.

Results and discussion

The effect of drying tempeatures on the moisture content

The moisture content profiles of dried kaffir lime leaves at different drying temperature were shown in Fig. 3. The results showed that the drying time decreased with the increase of the drying temperature. The rate of moisture loss was higher at high hot plate temperature. From the obsevation of the results in Fig. 3, the slopes of the graph in each temperature were presented the valules between 0 to 100 seconds. The moisture content was decreased until the final moisture content at 6.15 ± 0.31 (% wet basis) at all the hot surface drying temperature. The rate of reduction of moisture content showed the highest at the drying temperature between 150 to 180°C. The hot plate drying temperature at 140°C was selected as the optimal condition for drying the kaffir lime leaves. Since this condition provided the high rate of reduction of moisture content and less damaged of dried kaffir lime leaves. The kaffir lime leave was dried by heating exceed the boiling point of water which the moisture inside the kaffir lime leaves. Again, both kaffir lime leaves are characterized by thin, it caused heat transfer from the hot surface to the kaffir lime leaves quickly. Because the moisture in the kaffir lime leaves could spread at higher vapor pressure quickly and reduced the moisture content.

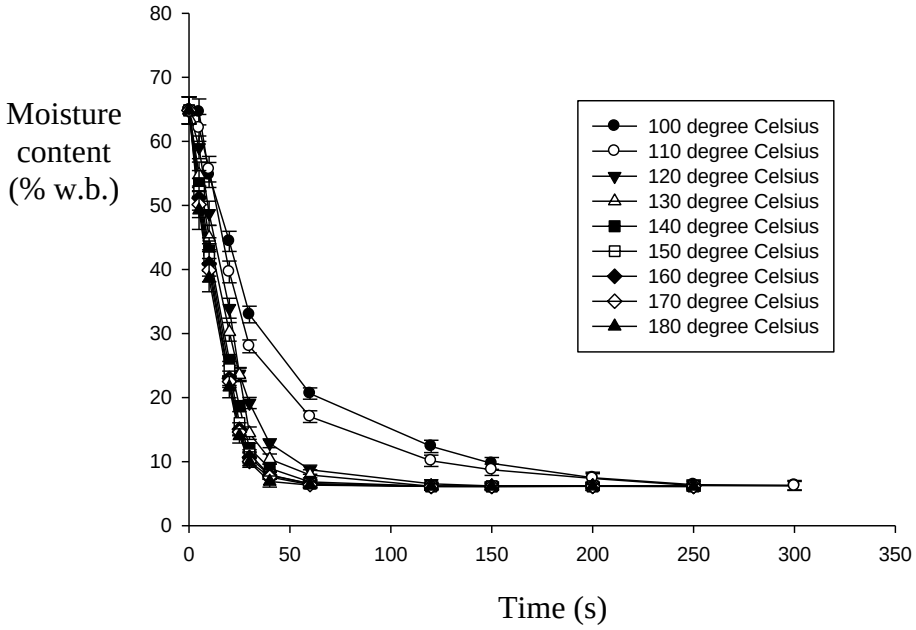


Fig 3. Moisture content profiles of kaffir lime leaves at different drying temperatures

Effect of drying on color parameters of dried kaffir lime leaves

One of the important quality attribute for food products is color. It is an indication for good quality of food and is generally associated with the acceptability of the food [1]. The consumer prefer the product that gave the quality close to the fresh leaves. So the measurement of color changes of kaffir lime leaves in both front and rear in term of lightness (L), greeness (a), yellowness (b), and total color difference (ΔE) . From result of drying times at 30 seconds, were proposed in Table 1.

Table 1 The color of kaffir lime leaves at front (a) and rear (b) under different drying temperatures

Hotplate temperature (° C)	L	a	b	ΔE
Fresh	31.78 ± 1.01	-7.87 ± 0.56	11.82 ± 0.31	-
100	33.57 ± 1.31	-7.97 ± 0.36	13.53 ± 0.25	2.48 ± 0.37
110	35.98 ± 1.03	-8.01 ± 0.43	14.59 ± 0.18	5.03 ± 0.18
120	36.39 ± 1.11	-8.35 ± 0.34	15.65 ± 0.26	6.01 ± 0.25
130	39.24 ±1.05	-8.63 ± 0.31	16.60 ± 0.21	8.8 9 ± 0.27
140	41.10 ± 0.97	-9.89 ± 0.23	17.11 ± 0.39	10.91 ± 0.34
150	41.12 ± 1.03	-10.39 ± 0.28	17.35 ± 0.91	11.14 ± 0.66
160	42.60 ± 1.65	-10.32 ± 0.20	17.41 ± 0.26	12.42 ± 0.74
170	43.61 ± 0.97	-10.56 ± 0.15	17.98 ± 0.32	13.61 ± 0.41
180	43.79 ± 0.85	-10.94 ± 0.39	18.23 ± 0.29	13.96 ± 0.23

(1a) The front of kaffir lime leaves

Hotplate temperature (° C)	L	a	b	ΔE
Fresh	50.01 ± 1.01	-11.21 ± 0.56	30.18 ± 0.31	-
100	51.31 ± 1.22	-11.71 ± 0.41	30.33 ± 0.65	1.40 ± 0.43
110	52.02 ± 1.03	-11.82 ± 0.33	30.63 ± 0.68	2.15 ± 0.44
120	52.71 ± 1.98	-12.10 ± 0.45	30.83 ± 0.66	2.92 ± 1.04
130	53.07 ± 1.06	-12.29 ± 0.46	31.08 ± 0.71	3.37 ± 0.42
140	53.42 ± 1.57	-12.50 ± 0.34	31.46 ± 0.55	3.86 ± 0.65
150	54.07 ± 1.66	-12.98 ± 0.56	31.79 ± 0.31	4.71 ± 0.65
160	55.35 ± 1.21	-13.03 ± 0.61	31.86 ± 0.45	5.89 ± 0.25
170	56.31 ± 1.43	-13.20 ± 0.21	32.13 ± 0.67	6.89 ± 0.53
180	56.74 ± 1.36	-13.33 ± 0.14	32.21 ± 0.21	7.34 ± 0.56

(1b) The rear of kaffir lime leaves

Table 1 showed the color parameters of fresh and dried samples at front (a) and rear (b). Lightness (L), greeness (a), yellowness (b), and total color difference (ΔE) value of the conductive heat transfer dried sample at 180°C were the highest change among the other drying temperatures. This may be due to the high drying temperature process. So the drying temperature at 140°C was optimal condition for drying kaffir lime leaves caused drying temperature over 140°C, the leaves were destoyed by increased vaper pressure . Moreover at this condition provided the the drying time within 30 seconds for drying kaffir lime leaves. The results agreed well with the effect of drying time on the color changes as shown in Table 2. The lower color degradation of conductive heat transfer dried kaffir lime leaves was due to the considerable reduction in drying time.

Table 2 The color of kaffir lime leaves at front (a) and rear (b) under different drying times

Time (seconds)	L	a	b	ΔE
Fresh	31.78 ± 1.01	-7.87 ± 0.56	11.82 ± 0.31	-
5	34.71 ± 1.20	-8.97 ± 0.34	14.81 ± 0.32	4.33 ± 0.29
10	39.69 ± 1.05	-9.60 ± 0.07	15.18 ± 0.17	8.77 ± 0.51
20	39.86 ± 1.16	-10.77 ± 0.05	15.57± 0.21	9.37 ± 0.54
30	42.16 ± 1.12	-10.90 ± 0.35	17.86 ± 0.19	12.39 ± 0.27
40	44.16 ± 0.95	-11.11 ± 015	19.49 ± 0.61	14.92 ± 0.51
50	49.03 ± 1.95	-11.10 ± 0.65	21.69 ± 0.53	20.13 ± 0.97

(2a) The front of kaffir lime leaves

Time (seconds)	L	a	b	ΔE
Fresh	50.01 ± 1.01	-11.21 ± 0.56	30.18 ± 0.31	-
5	51.80 ± 1.09	-11.86 ± 0.37	30.35 ± 0.21	1.91 ± 0.23
10	52.13 ± 1.08	-12.00 ± 0.17	30.46 ± 0.17	2.28 ± 0.42
20	52.56 ± 1.05	-12.50 ± 0.15	30.63 ± 0.16	2.89 ± 0.44
30	53.12 ± 1.12	-13.32 ± 0.25	30.86 ± 0.14	3.82 ± 0.37
40	55.21 ± 0.95	-13.82 ± 0.73	30.90 ± 0.33	5.86 ± 0.18
50	57.28 ± 1.63	-14.53 ± 0.43	31.10 ± 0.35	8.04 ± 0.63

(2b) The front of kaffir lime leaves

Dried kaffir lime leaves in both front and rear were changed when using the long drying time. The results similar with the effect of drying temperature on the color changes. At higher drying time, the color changes increased with the increased in drying times.

Effect of drying on the Citronella

The composition of essential oil of conductive heat transfer dried kaffir lime leaves was tested. Citronella is mainly as the composition of essential oil in kaffir lime leaves and appear. Citronellal is not only important commodities in the fragrance industry, but also potential as natural food preservatives having its antimicrobial activity. The amount of Citronella was measured by the Gas Chromatography method. The optimum drying temperature at 140°C for drying times at 5, 10, 20, 30, 40, and 50 were selected to test the Citronella content. The results as shown in Table 3.

Table 3 Citronella content (g dry weight 100 g) in the essential oil extracted from fresh and dried kaffir lime leaves.

Fresh kaffir lime leaves	Conductive heat transfer at 140°C at different drying times				
	5 s	10 s	20 s	30 s	60 s
185.47 ± 8.90	177.91 ± 1.51	180.18 ± 8.81	173.01 ± 8.83	184.29 ± 2.29	184.42 ± 3.05

From Table 3, it was shown that there were no significant differences in Citronella content essential oil between the fresh and dried kaffir lime leaves. This may be due to drying temperature was lower than the boiling point of Citronella (boiling at temperatures of 190 °C). It could be concluded that drying in a conductive heat transfer system can restore the important substances in the kaffir lime leaves.

Conclusions

The drying characteristics of kaffir lime leaves in a conductive heat transfer dryer were investigated at various drying temperatures. Drying time decreased with the increased in drying temperature. Vapor pressure at high drying temperature showed the highest because the vapor pressure in the leaf could not vent moisture out of time. And lead to the leaf tissue was damaged by drying at high temperature. At lower drying temperatures of 140 °C by using conductive heat transfer, dried kaffir lime leaves were brighten color when compared to the dried samples at more than 140 °C. The low drying temperatures also presented the least color degradation in the values of a and b. The results of color value showed that when the light - dark increased, the red-green color value reduced, the

yellow, and total color difference increased for both front and rear blades. Citronella is the main component in the essential oil and is the smell of the leaves. There were no significant loss in the Citronella content at the drying temperature at 140 °C for drying time of 5, 10, 20, 30, 40, and 50 seconds when comparing with the fresh leaves.

V. NOMENCLATURE

a	Greenness
b	Yellowness
° C	Degree Celsius
d	Dried kaffir lime leaves weight (g)
L	Lightness
M_d	Percentage of moisture from dry standards
M_w	Percentage of moisture from wet standards
w	Wet weight of kaffir lime leaves (g)

Reference

[1] Doymaz, I., Tugrul, N., Pala, M., 2006. Drying characteristics of dill and parsley leaves.J. Food Eng. 77, 559–565.

[2] I.Pongsirikul and P.Boonprasom ,2010, "Effect of drying per physical, chemical properties of dried Kaffir lime leaves," Re: GAO-1073, 41 years, science, vol 1, page 2553 Jan-Apr, special 548-551.

[3] Lin, T.M., Durance, T.D., Scaman, C.H., 1998. Characterization of vacuum microwave,air and freeze dried carrot slices. Food Res. Int. 31, 111–117.

[4] N.Faungfung ,2009, "Effect of drying methods, changes in physical, chemical, microbiological and sensory of Kaffir lime leaves during storage," Master of science thesis, food technology and food science branch, Faculty of science, University of Songkhla nakarin National Park, page 4-20.

[5] N.Poomsa-ad, K.Deejing, and L.Wiset ,2011, " Performance of heat pump dryer for kaffir lime leaves and quality of dried products under different temperatures and media," World academy of Science, Engineering and Technology, page 583-586.

[6] O. J. Loewer, T. C. Bridges, and R. A. Bucklin., 1994, "On-farm drying and storage systems", Joseph, Mich., ASAE.

[7] P.Sinchaisri, 2002, "Producing of herbs and spices," Department of agriculture, page 33

[8] P.Tinjun, 2007, Effect of Drying Methods on Free and Glycosidically-Bound Aroma Impact Compounds of Kaffir Lime Leaves (Citrus hystrix (DC.)),," Master of science thesis, food technology and food science branch, Faculty of science, Kasetsart university, page 4-20

[9] Sachin V.Jangam , Chung Lim Law and Arun S. Mujumdar, 2010, "Selection of dryer ," Drying of Foods, Vegetables and Fruits, Volum1, pp.65-67.

[10] Shiva et al, 2005, "Evaluation of herb drying performance using heat pump dryer," ME NETT ,19th

[11] Phoungchandang, S., Srinukroh, W., Leenanon, B., 2008. Kaffir lime leaf (Citrus hystrixD.C.) drying using tray and heat pump dehumidified drying. Drying Technol. 26,1602–1609.

[12] P.Tapbuntom, and S.Chinnasarn , 2007, "Effect of one step and two step dryings of Kaffir lime leaves and Lemon Grass Trunks," Agricultural sciences, 38 years, vol. 6. Nov - Dec, 2007, page 135-138.

[13] Raksakantong, P., Siriamornpun, S., Meeso, N., 2012. Effect of drying methods onvolatile compounds, fatty acids and antioxidant property of Thai kaffir lime(Citrus hystrix D.C.). Int. J. Food Sci. Technol. 47, 603–612.

[14] W.Srinuhroh et al, 2007, "Kaffir lime leaves drying by tray dryer and heat pump dehumidified dryer," Agricultural Sci. J.38: 5 (Suppl.)

[15] Tasirina, S.M., Puspasarib, I., Luna, A.W., Chaia, P.V. and Lee, W.T., 2013, Drying of kaffir lime leaves in a fluidized bed dryer with inertparticles: Kinetics and quality determination. Industrial Crops and Products 61, 193 – 201.

Algorithms for Measuring Persistence Lengths of Biological Structures: A Case Study of Cyanobacterial Filaments

Atitheb Chaiyasitdhi[1, a *], Panwong Kuntanawat[2, 3, b *]

[1]Biological Engineering Program, Faculty of Engineering,
King Mongkut's University of Technology Thonburi, Bangkok 10140, Thailand

[2]Division of Biotechnology, School of Bioresources and Technology,
King Mongkut's University of Technology Thonburi, 10140, Thailand

[3]Nanoscience & Nanotechnology Graduate Program, Faculty of Science,
King Mongkut's University of Technology Thonburi, 10140, Thailand

Keywords: Persistence length, Cyanobacteria, Algorithm

Abstract. Persistence length (L_p) is a measure used for inferring underlying mechanical properties, such as flexural rigidity (k_f), of rod-like structures under influences of thermodynamic thermal fluctuation (k_BT). As k_f is a product of Young's modulus or materials' elasticity (E) and the moment of inertia (I), investigation into the contribution of these factors to the diversity of morphological configurations can be made. L_p is acceptedly used to determine k_f of polymers, and biological filaments, such as actin fibers. However, L_p can also be applied to larger biological structures, such as cyanobacterial filaments due to their rod-like shape. In this study, we developed MATLAB scripts that can be used for measuring L_p of rod-like structures, particulary cyanobacterial filaments from their digital images by using image processing techniques. We also validated our algorithm and found that our measurements were consisten with L_p of cyanobacteria filaments previously reported in literatures.

Introduction

Mechanical properties of biological structures are important for understanding their implications in nature. For example, biological filaments, such as actin filaments requires to have certain flexibility to enable cellular movement and, at the same time, have enough strength to maintain morphology of cells [1]. Stiffness of stereocillia due to different in lengths enables frequency tuning ability of inner-hair cells [2]. Diatom chains with greater bending resistance were able to collect nutrients better than the more flexibles diatom chains [3].

Since numerous biological structures, such as actin filaments, microtubules, cilia, flagella, etc., are elongated shaped, mechanical properties related to bending, a major mode of deformation in elongated structures, are important. In terms of physics, flexural rigidity (k_f) is used for describing bending resistance. The k_f is determined by measuring persistence length (L_p) of biological structures [4-5].

The mechanical properties obtained from L_p are those related to bending or flexural rigidity (k_f), since the assumptions of L_p is based on the principles that elongated rod-like structures are likely to bend under the influence of thermal fluctuation (k_BT), according to the relation $L_p = k_f / k_BT = EI / k_BT$. Thus, the structures with higher degree of bending have lower persistence lengths and lower resistance to bending [6].

.

Cyanobacteria are multicellular organisms that some of them forms elongated structures or a filament of cells called *trichomes* [7]. Cyanobacteria are also important model organisms for studying evolution of bacterial shapes as they were one of the early organisms inherited on earth and they have various shapes from simple cells to filamentous and branched shapes. However, implications of k_f in cyanobacteria is less known than that of smaller biological structures like actin, microtubules, intermediate filaments, and other polymers. However, it has been reported that diatom chains with higher flexural rigidity may be able to collect nutrient better than more flexible diatom chains [3]. The study of k_f in cyanobacteria would pave a way to the insight of implications of mechanical properties in survivability of living organisms in the nature.

Therefore, the study of k_f of larger biological structures, such as cyanobacteria is important because it may help to improve our understanding of its benefit to the organisms. The study of Boal and Ng [8] suggested that L_p can also be applied to cyanobacterial filaments. The authors also showed that Lp obtained from fossils cyanobacteria can be used for determining their taxonomic categories and living relatives, if L_p of several groups of living cyanobacteria are known. However, algorithm or tools used for measuring L_p of biological structures from their digital images were not reported before. This can be an obstacle for other biologists who want to apply the concept of L_p to their research

. In this study, we develop MATLAB scripts for measuring L_p of cyanobacterial filaments (*Arthrospira platensis*) from digital images captured under a light microscope. In order to measure L_p of cyanobacterial filaments, image processing techniques are required to retrieve necessary information from digital images. We described the algorithms used to develop the MATLAB scripts from the step of image processing to data analysis so that the algorithm can developed and applied to other studies. Finally, we also validated the results made by our algorithms to results reported in literatures.

Materials and Methods

Algal strain, culture medium and growth condition. We obtained a helical *Arthrospira* strain C005-H from the Applied Algal Researh Laboratory, Chiang Mai Univeristy, Thailand and have maintained the strain as axenic cultures until we found the emergence of straight trichomes. Then, we isolated the straight *Arthrospira* trichomes and have maintained them as the strain C005-L under the same growth conditions as the C005-H. Both *Arthrospira* strains were cultured in the standard Zarrouk's medium [9] at 30 °C, with light intensity at 60 μmol photons $m^{-2}s^{-1}$, and continuous shaking at 120 rpm (SHO-2D, Wisd Laboratory Instruments, DAIHAN Scientific Co., Ltd., Korea). *Arthrospira* trichomes used in this study were collected from mid log-phases (OD_{560} = 1.288).

Persistence length as a relative indicator of flexural rigidity of elongated rod-like structures. According to the theory of entropic elasticity, a rod-like structure with intrinsic mechanical properties *EI*, where *E* is the elastic moduli and *I* is the moment of inertia of the cross-section, is likely to bend under the influence of thermal fluctuation (k_BT). Bending occurs from thermal fluctuation results in variation of local orientation along the body of the structure. For two rod-like structures exist in the same k_BT, the structure with lower k_f will have greater degree of bending and *vice versa*. A physical measure of variation of local orientations or bendiness is called persistence length or L_p. The L_p can be described as shown in the Eq. 1 as follows;

$$L_p = k_f / k_BT = EI / k_BT. \quad (1)$$

The L_p is widely used for measuring k_f of small biological structures that have an elongated rod-like shape, such as actin filaments, microtubules, intermediate filaments, and other polymers. However, Boal [3] also suggested that L_p can also be applied to larger biological structures, such as filamentous cyanobacteria. In this case, the thermodynamic term k_BT may represent influence of

fluctuating fluid environment rather than thermal fluctuation. However, the k_f of cyanobacterial filaments cannot be used as a quantitative measure as those from smaller biological structures due to small contribution of k_BT, the L_p of cyanobacteria from several genera can still be used comparatively to understand evolutionary relationship of those cyanobacteria based on their relative mechanical properties as shown in the study of Boal and Ng [6].

The L_p can be obtained from a 2D representation of an elongated rod-like structure by calculating a tangent correlation function $Ct(dS)$ of unit tangent vectors $< t(0) \cdot t(s) >$ along the body of the structure as a function of distance (dS). The correlation function shows that at greater distance, orientations along the body become more uncorrelated. However, a structure with higher L_p will have a lower rate of exponential decay ($-dS/L_p$) as shown in the Eq. 2 as follows;

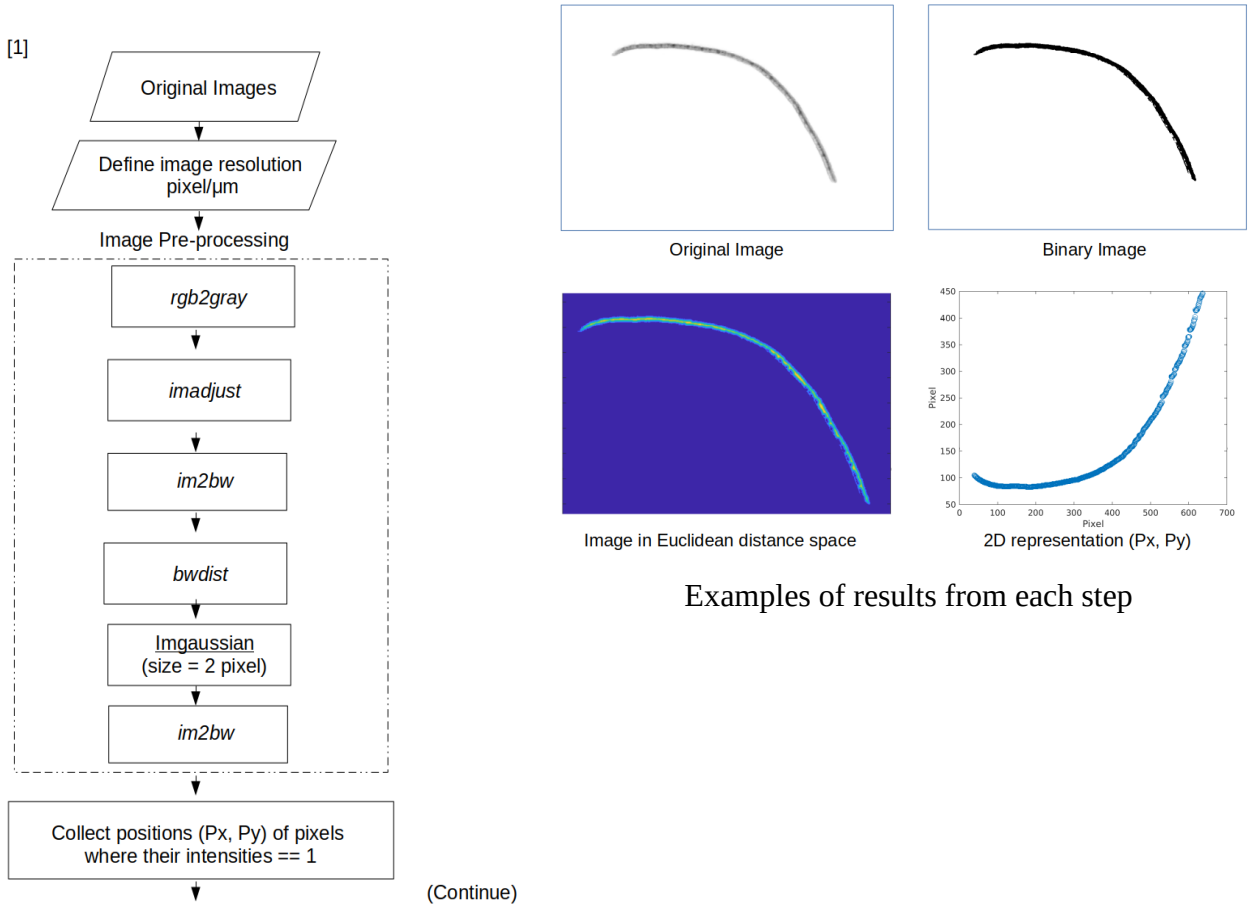
$$Ct(dS) = \exp(-dS / L_p). \quad (2)$$

In this study, we extracted 2D digital representations (P_x, P_y) of *Arthrospira* trichomes by using image processing package of MATLAB (Mathwork, USA) to retrieve information from digital images obtained under a light microscope. The algorithms developed in this study were implemented in MATLAB scripts based on the Eq. 1 and Eq. 2.

Algorithms for measuring persistence lengths of cyanobacterial filaments. To measure the L_p of an elongated rod-like structures, we developed MATLAB scripts that could retrieve necessary information for calculating L_p from digital images. In this study, the scripts were used to measure L_p of cyanobacteria *Arthrospira* from their digital images captured under a light microscope. The algorithm of the MALAB scripts developed in this study can be described as follows;

The algorithms were classified into 3 steps: 1) image pre-processing, 2) classification, and 3) calculation of L_p. The flow charts of Fig. 1, Fig. 2, and Fig. 3 describe the 3 steps of the algorithms respectively. Before image pre-processing, the user has to define sources of original digital images and the image resolution (pixel/µm). The image resolution will be later used for converting a pixel unit into a real unit. Italic characters inside a square box represent MATLAB functions with the same name. For example, *spline* is a MATLAB function called “spline” that is used to return a vector of interpolated values which can be used to smoothen curves and surfaces.

Image pre-processing. Prior to retrieving necessary information for calculating L_p, the original digital images of *Arthrospira* trichomes have to be pre-processed by using image processing techniques in order to improve quality of images for further analysis.



Examples of results from each step

Fig. 1 MATLAB algorithms for image pre-processing

First, the original images which usually had 3 channels of red (R), green (G), and blue (B), were converted into gray-scaled images, where intensity values vary between 0-255 by using *rgb2gray*. The contrast of images was increased by using *imadjust*, where 1% of the data is saturated at low and high intensities. The images were then further transformed into binary images, where intensity values are either 0 (black) or white (1) by using *im2bw*. These three steps were used to increase the contrast of *Arthrospira* trichomes from the background.

Second, the binary images of *Arthrospira* trichomes were transformed into a Euclidean distance space, which shows Euclidean distances of each pixel in the images, by using *bwdist*. The middle line of *Arthrospira* trichomes was defined in our algorithms as pixels where Euclidean distances were less than 2 pixels. We further applied 2D Gaussian filter to the images by using a custom-made function Imgaussian.

Finally, we used *im2bw* again to transform the images in a Euclidean distance space into binary images and collected positions representing *Arthrospira* trichomes (*Px*, *Py*) by choosing pixels where their intensity values equal to 1 (white).

Classification. After retrieving 2D representations of *Arthrospira* trichomes (*Px*, *Py*), the user classified the images into straight and helical structures. If the images are from straight *Arthrospira* trichomes (Fig. 2 – upper right), the positions (*Px, Py*) were directly used for L_p calculation. However, if the images are from helical *Arthrospira* trichomes (Fig. 2 – lower right), positions (*Px, Py*) were required to be transformed in to new positions (*Px_helix*, *Py_helix*) that represent the entire structure of trichomes.

To transform extracted positions in to positions representing the entire structure of trichomes, we used *spline* to smoothen curves drawn from the extracted positions. Finally, we used 2D Gaussian filters with a size of 60 pixels to smoothen helical curves into curves representing middle lines of helical *Arthrospira* trichomes by using a custom-made function Imgaussian. The positions obtained from this step were later used in L_p calculation.

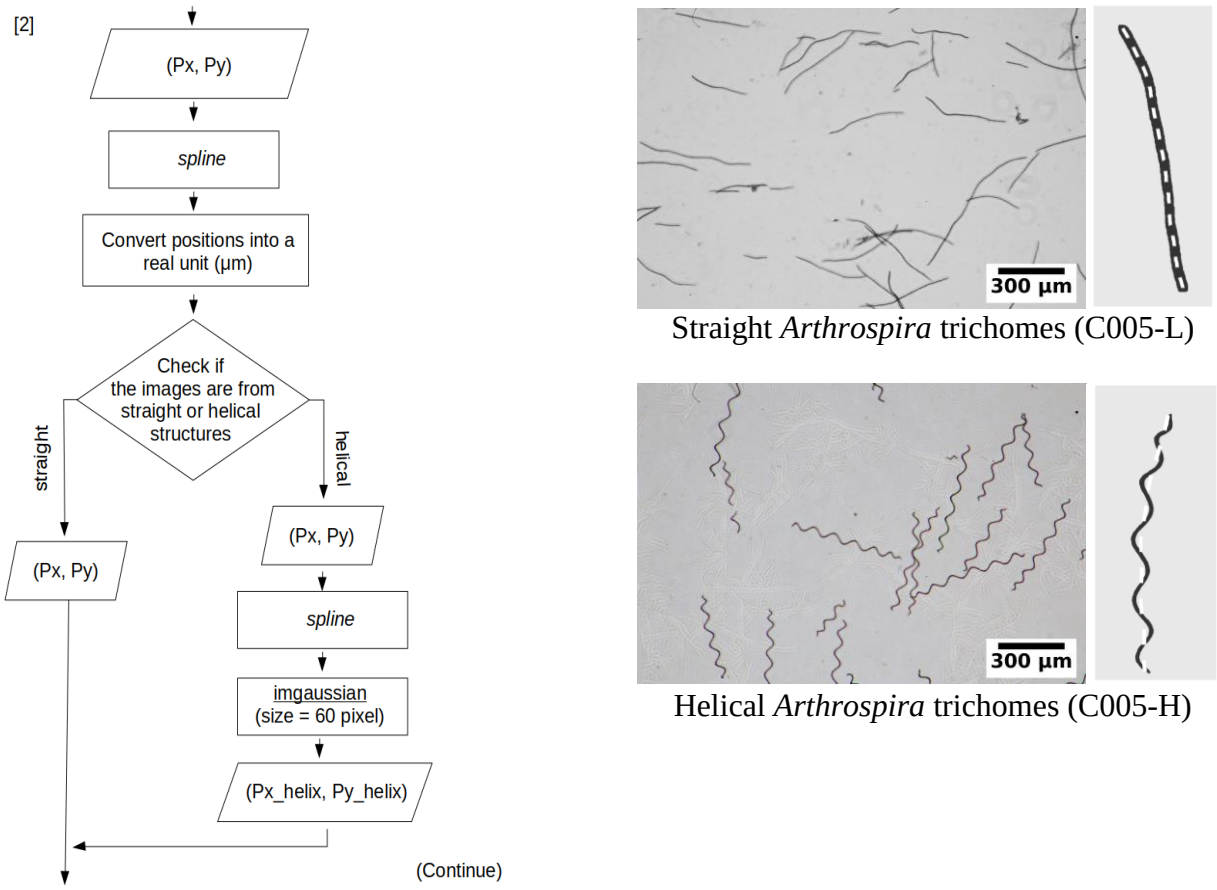


Fig. 2 MATLAB algorithms for image classification

Calculation of L_p. To calculate L_p of straight and helical *Arthrospira* trichomes, the positions (*Px, Py)* obtained from the previous step were used. According to the Eq. 2, a tangent correlation function as a function of distance (*dS*) had to be calculated. We initially calculated unit tangent vectors (r) at each position along the body of trichomes. Then, we calculated a correlation (*rr*) between adjacent unit tangent vectors with a distance of *dt.* Then, we calculated an average correlation at each *dt*, and iterated over increasing distance *dt.* Finally, the tangent correlation function was fitted with the Eq. 2 to yield L_p.

For other biological structures, k_f can be calculated by using the Eq. 1. However, in the case of *Arthrospira* trichomes, k_f obtained from this method can be used as a semi-quantitative measure for relative comparison.

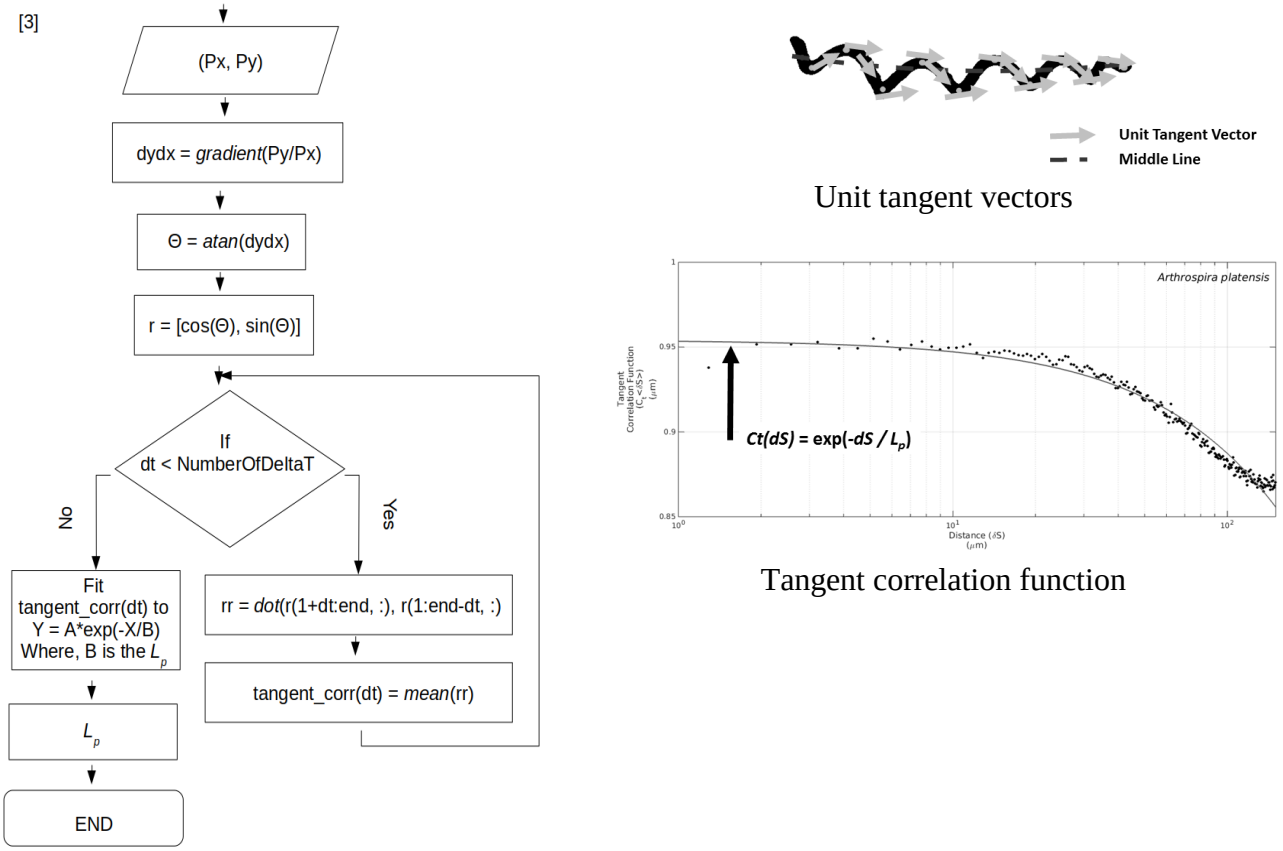


Fig. 3 MATLAB algorithms for calculation of L_p

Results and Discussion

Persistence lengths of straight and helical *Arthrospira* trichomes. In this study, we tested our algorithms with straight and helical *Arthrospira* trichomes. To calculate L_p of the trichomes, we implemented the algorithms described in the previous section into MATLAB scripts. The scripts required MATLAB's image processing package to work.

Arthrospira trichomes were collected and injected into a simple microfluidic channel as shown in the Fig. 4. Digital images of the trichomes were then collected under a light microscope. Finally, the MATLAB scripts yielded L_p of *Arthrospira* trichomes collected from a mid-log phase at each growth cycle. The results of L_p measurement made by algorithm are shown in the Fig. 5.

According to the results, we found that the median L_p from 2 growth cycles of straight and helical *Arthrospira* trichomes were [3.2 ± 1] x 10^3 µm (± *SD*) and [43.9 ± 22] x 10^3 µm (± *SD*), respectively. Since helical *Arthrospira* trichomes showed greater L_p than the straight trichomes, the results indicate that the helical trichomes have higher k_f and resist bending better than straight trichomes.

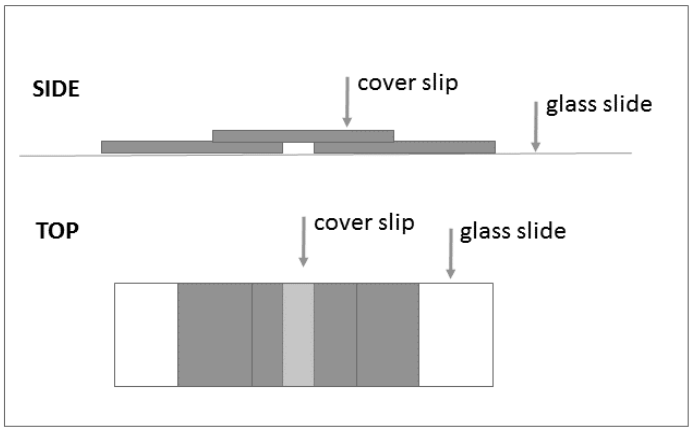


Fig. 4 Schematic of a simple microfluidic device used for injecting *Arthrospira* trichomes

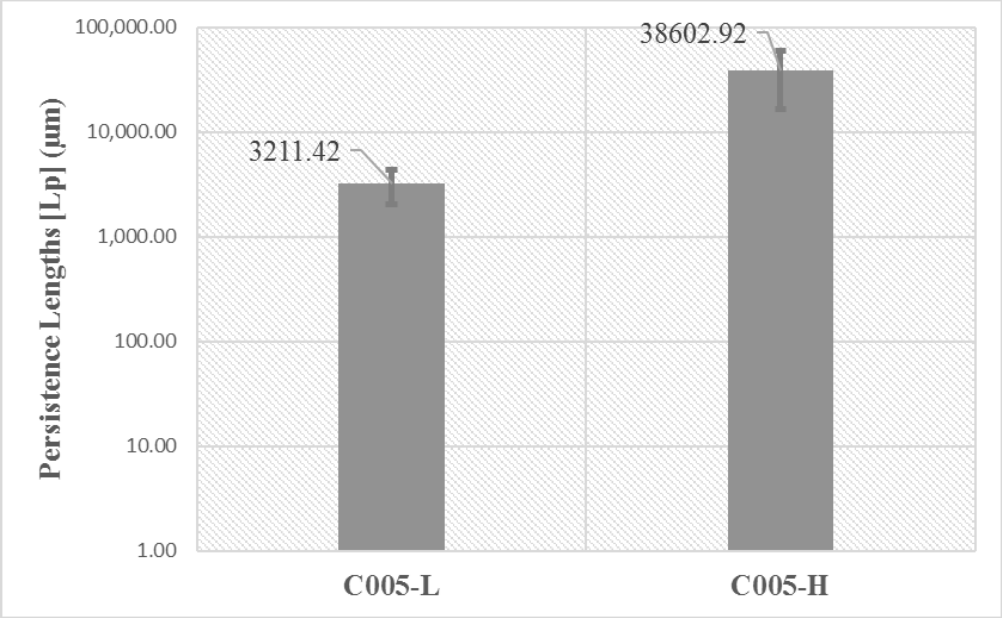


Fig. 5 Persistence lengths (L_p) of straight (C005-L) and helical (C005-H) *Arthrospira* trichomes

Comparison of L_p of *Arthrospira* trichomes with other cyanobacteria in literatures. After obtaining results of L_p from the previous step, we further compared our results to L_p of other cyanobacteria reported in literatures. Boal and Ng [6] reported L_p of both living cyanobacterial filaments and fossils. We plotted median of L_p of straight *Arthrospira* trichomes into the original plot reported in their study as shown in the Fig. 6. The plot shows the relationship between L_p and diameter (D) of cyanobacterial filaments. The results of helical trichomes were not used here as the diameter of helical trichomes corresponding to L_p obtained in this study were the diameter of helix instead of a diameter of a trichome because we determined L_p of the overall structure.

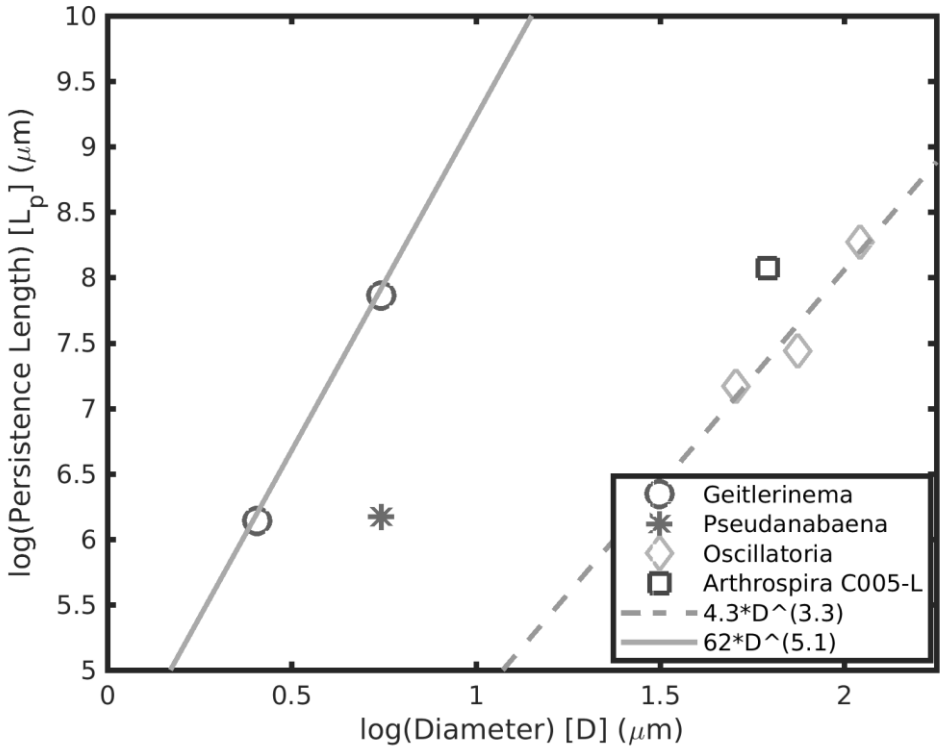


Fig. 6 The L_p of *Arthrospira* trichomes and cyanobacteria in literatures [6]

We found that L_p of straight *Arthrospira* trichomes were in the range of other *Oscillatoria* ($4.3 \cdot D^{3.3}$) while they were different from other genera, such as *Geitlerinema* ($62 \cdot D^{5.1}$), and *Pseudonabaena*. This results were consistent with the hypothesis of Boal and Ng [6] that cyanobacterial filaments from relative genera may have L_p that lies on the same hypothetical lines. The hypothetical line represents the relationship between L_p and D, since a rod-like structure with smaller diameter is likely to bend more than the structure with larger diameter.

Summary

In this study, we developed algorithms for measuring persistence lengths (L_p) of biological structures, and implemented the algorithms into MATLAB scripts. The scripts can be used for extracting L_p of biological structures from digital images. We tested our algorithms and scripts with straight and helical *Arthrospira* trichomes. The results obtained from our study were compared with other cyanobacterial trichomes in literatures. We found that the helical trichomes had higher L_p than the straight trichomes. This indicated that the helical trichomes can resist bending better than the straight trichomes. Benefits of having higher bending resistance may be the same with that reported in diatom chains that diatom chains with higher flexural rigidity (k_f) were able to collect nutrient better than that of more flexible diatom chains [5]. Moreover, we also found that L_p of straight *Arthrospira* trichomes were on the same hypothetical line $4.3 \cdot D^{3.3}$ as other *Oscillatoria*, and were distinct from other general, such as *Geitlerinema* ($62 \cdot D^{5.1}$), and *Pseudonabaena*. MATLAB scripts developed in this study can be downloaded from [10].

References

[1] G. M. Cooper and R. E. Hausman, The Cell: A Molecular Approach 2nd Edition. 2007.

[2] J. Kim, J.-H. Lee, J.-H. Ma, E. Chung, H. Choi, J. Bok, and J. Cheon, Magnetic Force Nanoprobe for Direct Observation of Audio Frequency Tonotopy of Hair Cells, Nano Lett., 16 (2016) 3885–3891.

[3] A. M. Young, L. Karp-Boss, P. A. Jumars, and E. N. Landis, Quantifying diatom aspirations: Mechanical properties of chain-forming species, Limnol. Oceanogr., 57 (2012) 1789–1801

[4] F. Gittes, B. Mickey, J. Nettleton, and J. Howard, Flexural rigidy of microtubules and actin-filaments measured from thermal fluctuation in shape, J. Cell Biol., 120 (1993). 923–934.

[5] A. M. Rosales, H. K. Murnen, S. R. Kline, R. N. Zuckermann, and R. A. Segalman, Determination of the persistence length of helical and non-helical polypeptoids in solution, Soft Matter, 8 (2012) 3673.

[6] D. Boal, Mechanics of the Cell, 2nd ed. Cambridge, UK: Cambridge University Press, 2012.

[7] A. Vonshak, Spirulina Platensis Arthrospira: Physiology, Cell-Biology and Biotechnology. 1997.

[8] D. Boal and R. Ng, Shape analysis of filamentous Precambrian microfossils and modern cyanobacteria, Paleobiology, 36 (2010) 555–572.

[9] Zarrouk, C. Contribution à l'étuded'unecyanophycée. Influence de Divers Facteurs Physiques et Chimiques Sur la Croissance et la Photosynthèse de Spirulina maxima. Ph.D. Thesis, Université De Paris, Paris. 1966.

[10] Information on https://www.mediafire.com/folder/k8bpwube4wrc4/PersistenceLength

Total phenols content and antioxidant activity and different solvent extracts of *Mokara* sp.

Napattaorn Buachoon

Keywords: *Mokara* sp., Total phenols content, DPPH, β-carotene, Antioxidant activity

Abstract. In this study, total phenols content, antioxidant activity and different solvent extracts of *Mokara* sp. The antioxidant activities of the extracts were determined by three different test methods, DPPH β-carotene and reducing power assays. In all methods, ethanol extract exhibited excellent activity potential than those of other extracts (chloroform, hexane, methanol, and ethyl acetate). As expected, the amount of total phenolics was very high in this extract. (56.91). Ethanol extract has been found to be rich in flavonoids. A positive correlation was observed between the antioxidant activity potential and total flavonoid levels of the extracts.

Introduction

Antioxidant compounds in food play an important role as a health protecting factor. Scientific vidence suggests that antioxidants reduce the risk for chronic diseases including cancer and heart disease. Primary sources of naturally occurring antioxidants are whole grains, fruits and vegetables. Plant sourced food antioxidants like vitamin C, vitamin E, carotenes, phenolic acids, phytate and phytoestrogens have been recognized as having the potential to reduce disease risk. Most of the antioxidant compounds in a typical diet are derived from plant sources and belong to various classes of compounds with a wide variety of physical and chemical properties. Some compounds, such as gallates, have strong antioxidant activity, while others, such as the mono-phenols are weak antioxidants.[1]

The main characteristic of an antioxidant is its ability to trap free radicals. Highly reactive free radicals and oxygen species are present in biological systems from a wide variety of sources. These free radicals may oxidize nucleic acids, proteins, lipids or DNA and can initiate degenerative disease. Antioxidant compounds like phenolic acids, polyphenols and flavonoids scavenge free radicals such as peroxide, hydroperoxide or lipid peroxyl and thus inhibit the oxidative mechanisms that lead to degenerative diseases.[2]

Various antioxidant activity methods have been used to monitor and compare the antioxidant activity of foods. In recent years, oxygen radical absorbance capacity assays and enhanced chemiluminescence assays have been used to evaluate antioxidant activity of foods, serum and other biological fluids[3] [4]. These methods require special equipment and technical skills for the analysis. The different types of methods published in the literature for the determinations of antioxidant activity of foods involve electron spin resonance (ESR) and chemiluminescence methods. These analytical methods measure the radical scavenging activity of antioxidants against free radicals like the 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical, the superoxide anion radical (O_2), the hydroxyl radical (OH), or the peroxyl radical (ROO). The various methods used to measure antioxidant activity of food products can give varying results depending on the specific free radical being used as a reactant. The various methods used to measure antioxidant activity of food products can give varying results depending on the specific free radical being used as a reactant. There are other methods which determine the resistance of lipid or lipid emulsions to oxidation in the presence of the antioxidant being tested. In the ORAC method, a sample is added to the peroxyl radical generator, 2,2'-azobis(2-amidinopropane)dihydrochloride (AAPH) and inhibition of the free radical action is measured[5] using the fluorescent compound, B-

phycoerythrin or R-phycoerythrin. Phenolic and polyphenolic compounds constitute the main class of natural antioxidants present in plants, foods, and beverages and are usually quantified employing Folin's reagent. Vinson et al. [6] have measured phenolics in fruits and vegetables colorimetrically using the Folin-Ciocalteu reagent and determined the fruit and vegetable's antioxidant capacity by inhibition of low density lipoprotein oxidation mediated by cupric ions.

The plant kingdom is a good source to produce a wide range of natural antioxidants. However, still there is not enough knowledge and data about the practical usefulness of most of them. Groups of secondary plant metabolites, antioxidant phenolics, and flavonoids are commonly found in various fruits, vegetables and herbs and they have been shown to provide a fruitful defence against oxidative stress from oxidizing agents and free radicals[7][2]. Most of the herbal infusions, commonly used as home medicines have antioxidative and pharmacological properties related to the presence of phenolic compounds, especially phenolic acids derivatives and flavonoids. Polyphenols, such as phenolic derivatives and flavonoids are also known for their ability to prevent fatty acids from oxidative decay, and provide an additional value to plants used as food ingredients, rich for example in rosmarinic acid[8].

The flowers of *Mokara* sp. is a orchid and hybrid of Ascocentrum, Arachnis and Vanda orchids and was first grown in 1969 in Singapore. *Mokara* sp. is among those orchid types which are very easy to grow and once these orchids art growing with care, they produce beautiful and exotic-looking flowers.

The aim of this work is to determine the total phenols content and antioxidant activity and different solvent extracts of *Mokara* sp by DPPH, β-carotene /linoleic acid and reducing power assays. Additionally, total flavonoid contents of ethanol, hexane, chloroform, methanol, and ethyl acetate extracts have been determined.

Materials and Methods

Chemicals

Standards of phenolic acids (gallic acid) of flavonoids (rutin hydrate) α,α-diphenyl-β-picrylhadrazyl(DPPH) and butylated were obtained from Sigma Chemicals(USA). The Folin-Ciocalteu's phenol reagent, and aluminium chloride($AlCl_3$) were from Fluka Chemie AG (Switzerland). Hydroxyanisole(BHA) was obtained from E. Merck (Germany). Solvents used for extraction were ethanol, hexane, chloroform, methanol, and ethyl acetate(HPLC grade)β-carotene linoleic acid Tween 40obtained from Merck(Germany). The deionised water was obtained from water distillation plants in our laboratory. All other chemicals were of analytical grade. UV spectra UV–Visible spectra measurements were done using a Spectrophotometer.

Plant material

Mokara sp. (Fig 1) was collected in August 2015 from Talad thai market. The collected plant material was air-dried in darkness at room temperature (20^{o}C). Dried plant parts were cut up and stored in tight-seal dark containers until needed.

Fig 1 *Mokara* sp.

Preparation of the extracts

Plant extracts were prepared according to a standard protocol. Prepared plant material (20 g) was transferred to dark-coloured flasks and mixed with 200 ml of solvents with different polarities (ethanol, hexane, chloroform, methanol, and ethyl acetate) respectively and stored at room temperature. After 24 h, infusions were filtered through Whatman No. 1 filter paper and residue was re-extracted with equal volume of solvents. After 48 h, the process was repeated. Combined supernatants were evaporated to dryness under vacuum at 40 °C using Rotary evaporator. The obtained extracts were kept in sterile sample tubes and stored in a refrigerator at 4 °C.

Determination of Total phenols content

The level of total phenols in the crude extracts was determined by using Folin–Ciocalteu reagent and external calibration with gallic acid. Briefly; 0.2 mL of extract solution and 0.2 mL of Folin–Ciocalteu reagent were added and the contents mixed thoroughly. After 4 min, 1 mL of 15% Na_2CO_3 was added, and then the mixture was allowed to stand for 2 h at normal temperature. The absorbance was measured at 760 nm using a spectrophotometer. The concentration of the total phenolics was calculated as mg of gallic acid equivalent by using an equation obtained from gallic acid calibration curve. The determination of total phenolic compounds in the fractions was carried out in triplicate and the results were averaged.

Determination of Total flavonoids

The level of total flavonoids in the examined plant extracts was determined using spectrophotometric method . The sample contained 1 ml of methanol solution of the extract in the concentration of 1 mg/ml and 1 ml of 2% $AlCl_3$ solution dissolved in methanol. The samples were incubated for an hour at room temperature. The absorbance was determined using spectrophotometer at λ_{max} = 415 nm. The samples were prepared in triplicate for each analysis and the mean value of absorbance was obtained. The same procedure was repeated for the standard solution of rutin and the calibration line was construed. Based on the measured absorbance, the concentration of flavonoids was read (mg/ml) on the calibration line; then, the content of flavonoids in extracts was expressed in terms of rutin equivalent (mg of RU/g of extract).

Antioxidant activity

Scavenging effect on 1,1-diphenyl-2-picrylhydrazyl

The hydrogen atoms or electrons donation ability of the corresponding extracts and some pure compounds were measured from the bleaching of purple colored methanol solution of DPPH. The effect of the extracts on DPPH radical was estimated according to 1 ml of various concentrations (0.2–1.0 mg ml^{-1}) of the extracts in methanol and water was added to a 1 ml of DPPH radical solution in methanol (final concentration of DPPH was 0.2 mM). The mixture was shaken vigorously and allowed standing for 30 min; the absorbance of the resulting solution was measured at 517 nm with a spectrophotometer. Inhibition of free radical DPPH in percent (I%) was calculated in following way:

$$I\% = (100 \times A_{Control} - A_{Sample})/A_{Control} \quad (1)$$

where $A_{Control}$ is the absorbance of the control reaction (containing all reagents except the test com pound), and A_{Sample} is the absorbance of the test compound. BHT and BHA were used as a control.

Total antioxidant activity by β-carotene–linoleic acid method

In this assay antioxidant capacity is determined by measuring the inhibition of the volatile organic compounds and the conjugated diene hydroperoxides arising from linoleic acid oxidation. A

stock solution of β-carotene–linoleic acid mixture was prepared as following: 0.5 mg β-carotene was dissolved in 1 ml of chloroform (HPLC grade) 25 μl linoleic acid and 200 mg Tween 40 was added. Chloroform was completely evaporated using a vacuum evaporator. Then 100 ml of oxygenated distilled water was added with vigorous shaking; 2.5 ml of this reaction mixture was dispersed to test tubes and 0.5 ml of various concentrations (0.4–2.0 mg ml^{-1}) of the extracts in solvents were added and the emulsion system was incubated for up to 2 h at 50°C. The same procedure was repeated with the positive control BHT, BHA and a blank. After this incubation period, absorbance of the mixtures was measured at 490 nm. Measurement of absorbance was continued until the color of β-carotene disappeared. The bleaching rate (R) of β-carotene was calculated according to

$$R = \ln)a/b)/t \quad (2)$$

where ln is the natural log, a the absorbance at time 0, b the absorbance at time t (30, 60, 90, 120 min). The antioxidant activity (AA) was calculated in terms of percent inhibition relative to the control using

$$AA = (R_{Control} - R_{Sample})/R_{Control}) \times 100 \quad (3)$$

Antioxidative activities of the extracts were compared with those of BHT and BHA at 0.4 mg ml^{-1} and blank consisting of only 0.5 ml methanol and water.

Reducing power

The reducing power was determined according to the method of Oyaizu [11]. Each extract (0.2–1.0 mg ml^{-1}) in methanol and water (2.5 ml) was mixed with 2.5 ml of 200 mM sodium phosphate buffer (pH 6.6) and 2.5 ml of 1% potassium ferricyanide and the mixture was incubated at 50°C for 20 min. Then, 2.5 ml of 10% trichloroacetic acid were added, and the mixture was centrifuged at 200 g (MSE Mistral 2000, London, UK) for 10 min. The upper layer (2.5 ml) was mixed with 2.5 ml of deionized water and 0.5 ml of 0.1% ferric chloride. Finally the absorbance was measured at 700 nm against a blank. BHT and BHA were used as a control.

Results and Discussion

Ethanol, hexane, chloroform, methanol, and ethyl acetate extracts were prepared to examine the total phenolic content, flavonoid concentration and antioxidant activity. The yield of extract obtained from 20 g of dry plant material was measured for each extract. The yields of Ethanol, hexane, chloroform, methanol, and ethyl acetate extracts of the leaves of *Mokara* sp. were 43.81%, 4.22%, 28.02%, 34.87%, and 29.18%, respectively(Table 1).

Table 1 The yields of solid residue after extraction and evaporation from 20 g dried plant parts.

Extracts	%yields
Ethanol	43.81
Hexane	4.22
Chloroform	28.02
Methanol	34.87
Ethyl acetate	29.18

Total phenols content

Among the protective actions in biological systems, phenolic compounds exhibit antioxidant activity. Thus, phenolics can be classified as free radical inhibitors (chain breaker), peroxide decomposers, metal inactivators or oxygen scavengers. Numerous studies have showed the

consumption of foods high in phenolics can reduce the risk of heart disease by slowing the progression of atherosclerosis due to their antioxidative properties [12].

The way of determination of the level of total phenolics is not based on absolute measurements of the amounts of phenolic compounds, but is in fact based on their chemical reducing capacity relative to gallic acid. It is very important to point out that; there is a positive relationship between antioxidant activity potential and amount of phenolic compounds of the crude extracts. From the phenol antioxidant index, a combined measure of the quality and quantity of antioxidants in vegetables has been obtained. In the present study the responses of the crude extracts in this assay may arise from the variety of phenolics found in five different extracts of the leaves of *Mokara* sp. Plants are the predominant sources of antioxidant, which act as free radical scavengers, making these foods essential to human health[13]. However, more than 80% of the total antioxidant activity in Plants comes from the ingredients other than antioxidant vitamins, indicating the presence of other potentially important antioxidants in these[14][15]. The phenolic derivatives compounds are the vital antioxidants which exhibit scavenging efficiency on the free radicals; reactive oxygen species are numerous and widely distributed in the plant kingdom[16].

Table 2 Total phenols content (as gallic acid equivalent) extracts of the leaves of *Mokara* sp.

Extracts	total phenolic(%w/w)
Ethanol	56.91±0.11
Methanol	35.92 ±0.12
Chloroform	11.27±0.23
Hexane	9.13±0.27
Ethyl acetate	8.95±0.11

Each value is the average of three analyses ± standard deviation.

As assumed, amount of the total phenolics was very high in ethanol extract (56.91%) methanol extract (35.92 %), chloroform extract (11.27%), hexane extract (9.13%) and ethyl acetate extract (8.95%). In recent studies, it has already been reported that the yield of extractable compounds was highest in ethanol extract from the leaves leaves of *Mokara* sp. in comparison with the solvents, such as hexane, chloroform, methanol, and ethyl acetate [17].

Total flavonoids

Flavonoids are class of secondary plant metabolites with significant antioxidant and chelating properties. Antioxidant activity of flavonoids depends on the structure and substitution pattern of hydroxyl groups [18].

The concentration of flavonoids in extracts of the species *Mokara* sp. was determined using spectrophotometric method with aluminum chloride. The content of flavonoids was expressed in terms of rutin equivalent, mg of RU/g of extract (Table 3). The concentration of flavonoids extracts from *Mokara* sp. ranged from 26.23 to 56.31 mg/g. Ethanol, hexane, chloroform, methanol, and ethyl acetate extracts contains the highest flavonoid concentration. The concentration of flavonoids in ethanol extract was 56.31 mg RU/g, which was very similar to the value of acetone extract concentration. The lowest flavonoid concentration was measured in chloroform and ethyl acetate extract.

Table 3 Concentrations of flavonoids in *Mokara* sp. extracts expressed in terms of rutin equivalent (mg of RU/g of extract)

Extract	mg of RU/g of extract
Ethanol	56.31 ±0.62
Methanol	51.27 ±0.32
Hexane	33.61±0.78
Chloroform	26.98±0.59
Ethyl acetate	26.23±0.52

Each value is the average of three analyses ± standard deviation.

Ethanol and methanol extracts from *Mokara* sp. have high concentration of total phenols (Table 2) and flavonoids (Table 3), which is in correlation with intense antioxidant activity of these extracts.

Antioxidant activity

Antioxidant activity of different solvent extracts from the leaves of *Mokara* sp. has been determined by three different test systems namely DPPH, b-carotene/linoleic acid and reducing power. In essence, the antioxidants react with the stable free radical i.e. 1,1-diphenyl-2-picrylhydrazyl (deep violet color) and convert it to 1,1-diphenyl-2-picrylhydrazine with discoloration. The degree of discoloration indicates the free radical scavenging potentials of the sample, antioxidant and it has been found that known antioxidant such as ascorbic acid, polyhydroxy aromatic compounds (hydroquinone, etc.) reduce and decolorize 1,1-diphenyl-2-picrylhydrazyl by their hydrogen donating ability. As can be seen from the Table 4, free radical cavenging effect of the samples exhibited a dose-dependent increase. The weakest radical scavenging activity was exhibited by Ethyl acetate and determined as 9.03%±0.47 (Table 4). It is extremely important to point out that, a strong correlation was observed between the radical scavenging capacity and polarity of the extracts. The ethanol extract which contains the most polar phytochemicals showed the strongest effect (43.89% ±0.46). This extract was followed by methanol (23.59% ±0.54). However, in the current study, none of the samples evaluated showed activity as strong as the synthetic antioxidants BHT (57.21% ±0.28) and BHA (59.98% ±0.46).

Table 4 Scavenging effect (%) on DPPH of different solvent extracts from the leaves of *Mokara* sp.

Extracts	Scavenging effect (%w/w)
Ethanol	43.89 ±0.46
Methanol	23.59 ±0.54
Hexane	15.90±0.32
Chloroform	11.29±0.31
Ethyl acetate	9.03±0.47
BHA	59.98 ±0.46
BHT	57.21 ±0.28

Each value is the average of three analyses ± standard deviation.

In β-carotene/linoleic acid model system, β-carotene undergoes rapid discoloration in the absence of an antioxidant. This is because of the coupled oxidation of β-carotene and linoleic acid, which generates free radicals. The linoleic acid free radical formed upon the abstraction of a hydrogen atom from one of its diallylic methylene group attacks the highly unsaturated β-carotene molecules. As a result, β-carotene is oxidized and broken down in part; subsequently the system loses its chromophore and characteristic orange color, which is monitored spectrophotometrically. In contrast to DPPH test system, the solvent extracts, as expected, the most active one was ethanol extract. Antioxidant activity of this sample was measured as 57.21% ±0.22. As can be seen from

the Table 5, this activity is almost equal to BHT at the same concentration value (67.35% ±0.02). One another way of defense mechanisms in preventing body against the hazardous effects of free radicals is reducing of these molecules by the antioxidant substances.

Table 5 Antioxidant activity (%) of different solvent extracts from the leaves of *Mokara* sp. by β-carotene-linoleate acid method.

Extracts	Antioxidant activity (%w/w)
Ethanol	57.21 ±0.22
Methanol	53.49 ±0.16
Hexane	19.90±0.34
Chloroform	18.29±0.32
Ethyl acetate	18.11±0.28
BHA	68.29 ±0.41
BHT	67.35 ±0.02

Each value is the average of three analyses ± standard deviation.

The antioxidant activity has been reported to be concomitant with the development of reducing capacity. Therefore, reducing capacity of a compound may serve as a significant indicator of its potential antioxidant activity [22]. Generally, the reducing properties are associated with the presence of compounds, which exert their action by breaking the free radical chain by donating a hydrogen atom or a single electron[3][23][4]. The electron donation group, especially hydroxyl group located at o-positions or p-positions of the compounds, makes the compound polar and therefore reducing power is increased [17]. The reducing power of the ethanol extract of *Mokara* sp. was found to steadily increase in direct proportion to the increasing concentration of the extract.

Table 6 Reducing power of different solvent extracts from the leaves of *Mokara* sp.

Extracts	Reducing power (%w/w)
Ethanol	38.23±0.11
Methanol	28.97 ±0.12
Chloroform	16.92±0.23
Hexane	9.78±0.27
Ethyl acetate	8.95±0.11

The values are means ± SD of three replicates.

As can be seen from the Table 6, In the case of reducing power assay of *Mokara* sp. showed an activity with an absorbance value of 38.23%±0.11 to 8.95%±0.11. Among the other samples, the strongest activity was exhibited by the ethanol extract (38.23%±0.11). This is followed by methanol (16.92%±0.23) and hexane extracts (16.92%±0.23).

Conclusion

Plant phenolics constitute one of the major groups of compounds acting as primary antioxidant free radical terminators. Flavonoids, as one of the most diverse and widespread groups of natural compounds, are probably the most natural phenolics. These compounds possess a wide spectrum of chemical and biological activities including radical scavenging properties. Shimoi *et al.* [24] concluded that plant flavonoids which show antioxidant activity in vitro also functionas antioxidants in vivo. A strong relationship between total phenolic content and antioxidant activity in fruits, vegetables, grain products, and plants subject of ethnopharmacological treatments has been reported [25].

The free radical scavenging activity of *Mokara* sp. extracts were tested through DPPH method and the results are presented in the (Table 4). The role of antioxidants is their interaction depends

on oxidative free radicals. The summary of DPPH method is that the antioxidants react with the stable free radical i.e., a,a-diphenyl-b-picrylhydrazyl(deep violet color)and convert it to a,a-diphenylb-picrylhydrazine with discolouration. The discolouration indicates the scavenging potentials of the sample antioxidant such as phenolic compounds. In the present study the five extracts of *Mokara* sp. were able to decolourise DPPH and the free radical scavenging potentials of the extracts of were found to be in the order of ethanol extract >methanol extract > chloroform extract > hexane extract >ethyl acetate extract. In our study may be it appears that the five extracts from the leaves of *Mokara* sp. possess hydrogen donating capabilities to act as an antioxidant.

In our study, the order of decreasing antioxidant activity among the *Mokara* sp. extracts through all the methods was found to be ethanol extract >methanol extract > chloroform extract > hexane extract >ethyl acetate extract. This order is similar to the phenolic contents of the extracts that showed the extent of antioxidant activity of the extract is in accordance with the amount of phenolics present in that extract [26][27]. In the present study it is found that the ethanol leaves extract of *Mokara* sp. contains substantial amount of phenolics and it is the extent of phenolics present in this extract being responsible for its marked antioxidant activity as assayed through various in vitro models. Several reports have finally shown close relationship between total phenolic contents and antioxidative activity of the plants [6]. The chemical composition and chemical structures of active extract components are important factors governing the efficacy of natural antioxidants, the antioxidant activity of an extract could not be explained on the basis of their phenolic content, which also needs their characterization[25]. However, different types of actions of the phenolics present in the extracts cannot be ruled out. So far we know this is the first report that envisages the antioxidant activities of *Mokara* sp. extracts. Hence the leaves of *Mokara* sp. could be a good source of antioxidant phenolics. Further studies are needed for the isolation and identification of individual phenolic compounds and also in vivo studies are needed for better understanding of their mechanism of action as antioxidant.

References

[1] Halliwell, B., Gutteridge, J.M.C., (2007). *Free Radicals in Biology and Medicine*. Oxford University Press, Oxford.

[2] Antolovich, M., Prenzler, P., Robards, K., Ryan, D., 2000. Sample preparation in the analysis of phenolic compounds in fruits. *Analyst*., 125, 989–1009.

[3] M. Amzad Hossain. (2015).Muhammad Dawood Shah A study on the total phenols content and antioxidant activity of essential oil and different solvent extracts of endemic plant Merremia borneensis., *Arabian Journal of Chemistry*., 8, 66–71

[4] Cengiz S. , Kadir A., Bektas T., Ahmet C., Gulsah A., Ebru M., (2009). Studies on the antioxidant activity of essential oil and different solvent extracts of Vitex agnus castus L. fruits from Turkey. *Food and Chemical Toxicology*., 47, 2479–2483

[5] Cao, G., Verdon, C.P., Wu, A.H.B., Wang, H., & Prior, R.L. (1995) Clin, *Chem*., 41, 1738-1744.

[6] Vinson, J.A., Hao, Y., Zubic, S.K., (1998). Food antioxidant quantity and quality in foods: vegetables. *Journal of Agricultural Food Chemistry*., 46, 3630–3634.

[7] A. Matkowski, P. Tasarz and E. Szypuła (November 2008) Antioxidant activity of herb extracts from five medicinal plants from *Lamiaceae*, subfamily *Lamioideae Journal of Medicinal Plants Research*. 2(11). pp. 321-330.

[8] I. Fecka, D. Raj, M. Krauze-Baranowska**.** (2007)**.** *Quantitative determination of four water-soluble compounds in herbal drug from Lamiaceae using different chromatographic techniques Chromatographia*., 66,. 87–93

[9] T. Tekao, N. Watnabe, I. Yagi, and K. Sakata, (1994): A simple screening method for antioxidant and isolation of several antioxidants produced by marine bacteria from fish and shellfish. *Biosci. Biotechnol. Biochem*. 58. pp. 1780-1783.

[10] Kumarasamy, Y., Byres, M., Cox, P.J., Jasapars, M., Nahar, L., Sarker, S.D. (2007): Screening seeds of some Scottish plants for free-radical scavenging activity. *Phytother. Res*., 21, 615-621.

[11] Oyaizu, M., (1986). Studies on products of browning reactions: antioxidative activities of browning reaction prepared from glucosamine. *Japanese Journal of Nutrition*., 44, 307–315.

[12] Pulido, R.; Bravo, L.; Saura-Calixto, F. (2000). Antioxidant activity of dietary polyphenols as determined by a modified ferric reducing/antioxidant power assay., *Journal of Agricultural and Food Chemistry*, 48, 3396-3402.

[13] J.G. Elliot. (1999). Application of antioxidant vitamins in foods and beverages. *Food Technology* 53. pp. 46–48.

[14] Miller, H.E., Rigelhof, F., Marquart, L., Prakash, A., and Kanter, M. (2000) *Cereal Foods World.*, 45(2)**,** 59-63.

[15] Miller, H.E., Rigelhof, F., Marquart, L., Prakash, A., and Kanter, M. (2000) J. Am. Coll. *Nutr*., 19(3), 312S-319S.

[16] Prior, R.L., Cao, G., Martin, A., Sofic, E., McEwen, J., O'Brien, C., Lischner, N., Ehlenfeldt, M., Kalt, W., Krewer, G., & Mainland, C.M., (1998) J. Agric. *Food Chem*., 46, 2686-2693.

[17] Negi, P.S., Jayaprakasha, G.K., Jena, B.S., (2002). Antioxidant and antimutagenic activities of pomegranate peel extracts. *Food Chem*, 80, 293–297.

[18] Sharififar, F., Nudeh-Dehghn, G., Mirtajaldini, M. (2008): Major flavonoids with antioxidant activity from *Teucrium polium* L. *Food Chem*., 112, 885-888.

[19] Amarowicz, R., Pegg, B.R., Rahimi-Moghaddam, P., Bar, B., WeilL, J.A. (2003): Free-radical scavenging capacity and antioxidant activity of selected plant species from the Canadian prairies. *Food Chem*, 84, 551-562.

[20] Čandanovic-Brunet, J., Ćetkovic, G., Đilas, S., Tumbas, V., Boganovic, G.,Mandic, A., Marow, S., Cvetkpvic, D., Čandanovic, V. (2008): Radical scavenging, antibacterial, and antiproliferative activites of Melissa officinalis L. extracts. *J. Med. Food*, 11, 133-143.

[21] Borneo, R., Leon, E.A., Aguirre, A., Ribotta, P., Cantero, J.J. (2008): Antioxidant capacity of medicinal plants from the Province of Cordoba (Argentina) and their in vitro testing in model food system. *Food Chem*, 112, 664-670.

[22] Turkoglu, A.; Duru, M. E.; Mercan, N.; Kivrak, I.; Gezer, K. (2007). Antioxidant and antimicrobial activities of *Laetiporus sulphureus* (Bull.) Murril. *Food Chemistry.,* 101, 267-273

[24] K. Shimoi, S. Masuda, B. Shen, B. Furugori, N. Kinae, (1996). Radioprotective effect of antioxidative plant flavonoids in mice. *Mutation Research*. 350, 153–161.

[25] N. Buachoon and P.Sunthornsart. (2015). Development of lotion from *Albizia myriophylla* Benth. Crude ,*VRU Research and Development Journal Science and technology*. **(**10). pp.98-106.

[26] Abdille, M.H., Singh, R.P., Jayaprakasha, G.K., Jena, B.S., (2005). Antioxidant activity of the extracts from Dillenia indica fruits. *Food Chem.*, 90 (4), 891–896.

[27] Heinonen, M., Lehtonen, P.J., Hopla, A., (1998). Antioxidant activity of berry and fruit wines and liquor. *Journal of Agricultural Food Chemistry*., 48, 25–31.

Using Ground Leucaena Branches for Oyster Mushroom-Hungary Cultivation in Summer

Kannika Umpuch*, Juthathip Iamchom and Prasarn Yim-orn

Agriculture Program, Faculty of Agricultural Technology, Valaya Alongkorn Rajabhat University under the Royal Patronage, Pathum Thani 13180, Thailand

Keywords: leucaena branch**,** oyster mushroom-Hungary, yield components

Abstract. Leucaena tree (*Leucaena leucocephala)* are used by farmers in many ways such as leaves and green branches used as animal feed or green manure and stems used as fuel wood. Whereas, old/hard branches are useless left in a field. The study was set to use the useless leucaena branches by substituted for para rubber sawdust as a substrate for oyster mushroom-Hungary (*Pleurotus ostreatus* (Fr.) Kumm) cultivation. Thus, the objective of this study was to find a suitable ratio of leucaena branches and to observe yield component of oyster mushroom-Hungary in different ratios of a compost. The proportion of ground leucaena branches to para rubber sawdust was set into five that were determined as 5 treatments with 3 replications, 5 composts each, that were 0, 25, 50, 75 and 100 percent of substitution. The experiment was conducted at the Faculty of Agricultural Technology during February to May 2015. Yield and yield components were collected and statistical compared by CRD. The study revealed that an oyster mushroom-Hungary could be cultivated by ground leucaena branches substituted for para rubber sawdust in a range of 25 to 75 percent in a compost with an acceptable yield and good quality in summer.

Introduction

Leucaena is a fast-growing tree with well adaptation in the environment of Thailand. Both native and imported cultivars [1]. It is considered that leucaena is a valuable crop of Thai agricultural society and widely used in many purposes. Pods or shoots are used as raw materials for animal feed (cattle, goats, sheep, and chicken as well as pigs) and human dishes. Leucaena also be burnt to make charcoal for cooking in the household. Since the situation of energy shortages in Thailand has occurred. There are many published researches about finding the alternative resources to produce renewable energy. Leucaena wood is one option that many agencies have paid attention to generate electricity [2,3], especially a giant leucaena, which has a large stem diameter and contains the appropriate chemical composition as a fuel source [4]. However, a study about leucaena plantation for biofuel revealed that there was a residue as branches and small stems more than 500 kgDM/rai/yr (3.12 ton DM/ha/yr) left in the field that cannot enter to biofuel process [5]. Therefore, taking the residues to make value added is a challenge way and could make a benefit to the farmers.

Mushrooms are highly nutritious plants. It consists of high protein, vitamin and low fat that needed by consumers. In addition, it can be cooked in a variety of good taste for people of all ages. In addition to a high nutrient content that beneficial to the body. Some mushrooms have also been found to have medicinal properties for preventing diseases such as hypertension, atherosclerosis, respiratory diseases, obesity, hepatitis B, cancer, etc. [6]. Pleurotus mushrooms or oyster mushroom family are easy to grow and popular among people. There are many species such as white oyster mushrooms, Hungarian mushrooms or oyster mushroom-Hungary and sajor caju mushrooms as well as abalone mushrooms [7].

Oyster mushroom-Hungary can be cultivated in the same material as oyster mushrooms, such as straw, rice, corn, sawdust, soft wood, etc. Nowadays, the cultivation of mushroom has many ways such as in plastic bag, bucket, as well as sag, but a main substrate is similar that is a sawdust from softwood, para rubber tree. In some areas can use other substrate such as baggase, rice straw or fine grasses from agricultural sector [7]. Thus, mushroom cultivation use low of cost production and technology and get high income to farmer. Therefore, the aim of this study was to find an optimum of leucaena branch residues substituted for para rubber sawdust to produce oyster mushroom with an acceptable yield and good quality in summer.

Material and Method

Treatments

The mushroom substrates are para rubber sawdust (PR) and ground leucaena branches (GL). Five formulas of mushroom compost were set with three replications four bags each that were;

Formula 1 (T1): 12 bags of 100% PR
Formula 2 (T2): 12 bags of 75% PR + 25% GL
Formula 3 (T3): 12 bags of 50% PR + 50% GL
Formula 4 (T4): 12 bags of 25% PR + 75% GL
Formula 5 (T5): 12 bags of 100% GL

Mixed each formula for 25 kilograms as shown in Table 1

Table 1 Mushroom compost formulas

Ingredient	T1	T2	T3	T4	T5
PR	25 kg.	18.75 kg.	12.5 kg.	6.25 kg.	-
GL	-	6.25 kg.	12.5 kg.	18.75 kg.	1.25 kg.
Rice bran	1.25 kg.	1.25 kg.	1.25 kg.	1.25 kg.	1.25 kg.
Flour	250 g.	250 g.	250 g.	250 g.	250 g.
Lime	250 g.	250 g.	250 g.	250 g.	250 g.
Epsom salts	50 g.	50 g.	50 g.	50 g.	50 g.
Water	17.5 kg.	17.5 kg.	17.5 kg.	17.5 kg.	17.5 kg.
Size	500 g.	500 g.	500 g.	500 g.	500 g.
number of bag	48	48	48	48	48

Substrate preparation and mushroom cultivation method

1. Mix sawdust (PR or GL) with rice bran and other additives. Adjust the water content of the mixture to 60-65 percent (A rule of thumb is squeezing the mixture in the palm of your hand. When a droplet or two barely escapes, the mixture has a proper water content).

2. Fill the bags and compact. Commonly use a plastic ring to make a bottle neck for easy handling. Put a plastic ring on the bag end, pull out the bag end through the ring, fold down the pulled-out part, tied it with a rubber band and plug with cotton, paper or cotton-tapped plastic plug.

3. Pasteurize the bags in a steam boiler for 4 hours from the time temperature reaches 90-100°C.

4. Cool them and inoculate spawn from a commercial mushroom spawn (spores or root-like mycelium stored in sawdust, grain, or agar) by aseptic technique. Sorghum grain is commonly used material for a spawn carrier.

5. All the mushroom bags were set on a table covered with a plastic black to prevent a direct sunlight. In this step, the spawn is ready to colonize the substrate with mycelium.

6. Approximately two to five weeks, the white, feathery root called mycelium spread throughout the substrate. Move them to the fruiting mushroom house.

Data collection

1. Mycelium growth measured by using measuring tape every week from bottle neck to the end of mycelium.

2. Yield components that were fresh weight, cap size, stalk size and fruit per bag.

4. Ambient temperature and relative humidity in the fruiting house were recorded by using wet-dry thermometer at noon.

Experimental design and statistical analysis

Completely randomize design was used with five treatments, three replications four bags each. All raw data were analyzed and means were compared by DMRT at 95 percent of significance.

Site and duration

Faculty of Agricultural Technology, Valaya Alongkorn Rajabhat University under the Royal Patronage, Pathum Thani province. During time was February to May 2015.

Results and discussion

Ambient temperature and relative humidity in the fruiting house

In March, April and May, the mean temperatures were 30.6, 31.0 and 31.9 °C and the mean relative humidity were 87.4, 88.2 and 87.1 percent, respectively. This condition suited for growth of the mushroom that were 25-32°C with 80-90 percent of relative humidity [6].

Mycelium growth (cm.)

It took around three weeks for the mycelium fully-spread in the bags. This is a normal growth duration of oyster mushroom to spread all the bags roughly at 20-25 days [8]. In this study, the lengths of mycelium were differed from substrate ratio ($P<0.05$). The mean length of the mycelium was approximately 16 cm. for 0-75 percent of GL substitution, except in 100 percent of GL was only 13.21 cm. However, a growth trend as shown in Table 2 showed that the mycelium grew slower when the proportion of GL increased. Although a leucaena is a softwood plant like a para rubber tree, but the utilization by the oyster mushroom spawn is incomplete. A possible reason is the size of ground leucaena branch is larger than that of pararubber sawdust. Resulted in a longer period of degradation by the spawn.

However, considering the rate of growth of the mycelium each week, the oyster mushroom-Hungary's mycelium is growing very well at the second week between 2.37-3.81 times longer than that the length of the first week. The 0:100 group had a high growth rate of 3.81 times while the first group had a growth rate of only 2.70 times. In the third week, the rate of mycelium growth was reduced. It ranges from 1.53 to 1.87 times. It is possible that the spawn is degraded and consumes most of the substrate during the second week of mycelium growth.

Table 2 Mycelium growth of oyster mushroom-Hungary with different ratio of para rubber sawdust (PR) and ground leucaena branch (GL) in the substrate.

PR:GL ratio	mycelium length (cm.) with growth rate from previous week (time)		
	1st week (12-02-15)	2nd week (19-02-15)	3rd week (26-02-15)
100:0	3.93^{a}	10.66^{a} (2.70)	16.33^{a} (1.53)
72:25	2.75^{b}	9.96^{a} (3.60)	16.33^{a} (1.64)
50:50	3.00^{b}	9.83^{a} (3.23)	16.12^{a} (1.64)
25:75	2.56^{bc}	8.58^{b} (3.35)	16.04^{a} (1.87)
0:100	1.96^{c}	7.46^{c} (3.81)	13.21^{b} (1.77)

Means in the same column with the same letter are not statistically significant difference at 95 percent.

Yield and yield components

The study found that the use of ground leucaena (GL) over 50 percent substitution resulting in the fresh weight per bag was better than other ratios. The use of 50 percent of GL, the mushroom gave the best fresh weight. It was likely that the number of fruiting body was higher than that of the other groups. Although there was no statistically significant difference ($P>0.05$) was observed. On the other hand, the large number of fruiting body resulted in a small size of mushroom. Especially the size of the cap, which was one centimeter smaller than the others, as shown in Table 3.

Table 3 Yield and yield component of oyster mushroom-Hungary with different ratio of para rubber sawdust (PR) and ground leucaena branch (GL) in the substrate.

PR:GL ratio	Fresh weight (g.)	Fruit per bag (no.)	Cap diameter (cm.)	Stalk diameter (cm.)	Stalk length (cm.)
100:0	44.44^{ab}	14.44^{a}	3.22^{b}	1.20^{a}	3.24^{b}
72:25	39.50^{b}	10.25^{a}	4.76a	0.81^{ab}	5.01^{a}
50:50	54.67^{a}	14.58^{a}	3.71^{ab}	0.63^{b}	4.13^{a}
25:75	52.00^{a}	12.50^{a}	4.71^{ab}	0.67^{ab}	4.69^{a}
0:100	45.53^{ab}	11.97^{a}	3.72^{ab}	0.67^{ab}	4.17^{a}

Means in the same column with the same letter are not statistically significant difference at 95 percent.

It can be seen that ground leucaena, especially unused branches, can be used as a material of mushroom cultivation in case of the farmers cannot find or buy the rubber wood sawdust. However, a disadvantage is to provide a wood grinder or to add a process that allows the leucaena to be degraded by the spawn faster, such as soaking or fermenting in advance.

Nutrient from the substrate is an important factor to promote mushroom growth. Different types of mushrooms need different nutrients, such as champignon mushroom, grows well in compost, which comes from rice straw, mixed with horse manure and requires high nitrogen content. Wheat straw contains 0.62 percent nitrogen and horse manure has about 1.5-1.8 percent nitrogen. Carbon and nitrogen raion (C:N ratio) suitable for mushroom growth is about 17. Straw mushroom grows well in a material with high moisture content or in plant material, such as, rice straw or banana leaves. That lignocellulosic materials contain different nitrogent contents for instance, 0.58 percent in straw and 1.17 percent in banana leaves. Shitake and oyster mushrooms can grow well on wood that contains high level of lignin and low content of nitrogen. Wood contains about 0.03-1.0 percent nitrogen compared with 0.58-1.71 percent in annual crops. The carbon-nitrogen ratio in the wood ranges from 350 to 500:1, so the mushrooms are able to utilize large amounts of carbohydrates, including lignin with little amount of nitrogen [9]. Therefore, compost substrate from lignocellulosic plants can promote mycelium growth and forming a fruiting body of the mushroom.

The leucaena braches used in this study were not analyzed for carbon and nitrogen content and also have not been reported. However, in comparison with the woody stem parts, it was found that the proportion of carbon to nitrogen was about 69.7: 1, which was considered high and suitable for mushroom growth [4]. Sawdust from softwood such as rubber wood, cotton wood, and acacia wood are easy to buy, inexpensive, fast degradable and not have gum that is harmful to mushroom spawn. Hardwood sawdust, such as tamarind wood, durian, jackfruit, should be fermented first. In addition, softwood branches should be ground thoroughly before use [10]. In foreign countries, the studies were performed by using softwood such as acacia wood and eucalyptus wood that can also grow mushrooms very well [11].

Conclusion

An oyster mushroom-Hungary could be cultivated by ground leucaena branches substituted for para rubber sawdust in a range of 25 to 75 percent in a compost with an acceptable yield and good quality in summer.

Acknowledgement

The author would like to thank the Research and Development Institute, Valaya Alongkorn Rajabhat University under the Royal Patronage for fully funded.

References

[1] S. Tudsri, Tropical Forage Crops, First Edition, Kasetsart University Publishing, Bangkok, 2004.

[2] S. Tudsri, P. Sripichitt, N. Wongsuwan and K. Nakamanee, The study of biomass yield of giant leucaena used as a sustainable bioenergy resource, Research Full Report, Bangkok, 2007.

[3] W. Artharn, N. Kongkrapee, K. Rubsombat, P. Chanaroke and T. Hinsui, The study of model of small biomass plant for community, National Research Council of Thailand, Bangkok, 2007.

[4] K. Rengsirikul, A. Kanjanakuha, Y. Ishii, K. Kangvansaichol, P. Sripichitt, V. Punsuvon, P. Vaithanomsat, G. Nakamanee and S. Tudsri, Potential forage and biomass production of newly introduced varieties of leucaena (*leucaena leucocephala* (Lam.) der Wit.) in Thailand, Grassland Science. 57 (2011) 94-100.

[5] K. Rengsirikul, The Potential of Napiergrass and Leucaena as Bioenergy Feedstock in Thailand, Ph.D. Thesis, Graduate School, Kasetsart University, Bangkok, 2011.

[6] P. Yim-orn, Mushroom Cultivation, Valaya Alongkorn Rajabhat University under the Royal Patronage, Pathum Thani, 2006.

[7] P. Jantarasiri, P. Yingthongchai, N. Boontim and S. Tongsiri, Production of mushroom spawn and mushroom compost, Chiang Mai University, 2008.

[8] P. Phothirat, Mushroom Cultivation Technology, King Mongkut's Institute Technology Ladkrabang, 1989.

[9] S. T. Chang and P.G. Miles, Mushroom Cultivation, Nutrition Value, Medicinal Effect, and Environmental Impact, Second Edition, CRC Press, Florida, USA, 2004.

[10] T. Sangpakornkit, Oyster Mushroom, Tanathat Printing, Bangkok, 2009.

[11] T.E. Tisdale, Cultivation of the Oyster Mushroom (*Pleurotus sp*.) on Wood Substrates in Hawaii, Master Thesis, University of Hawaii, USA, 2004.

Isolation of actinomycetes from sugarcane rhizospheric soils with inhibitory activity against *Fusarium moniliforme* causing wilt disease in sugarcane

Janejira Detraksa[1]*, Thanaphan Ausalung[1] and Laongtip Thongmee[1]

Abstract

This research aimed to isolate and screen antagonistic actinomycetes on the control wilt disease in sugarcane. The fungi causing wilt disease were isolated from the wilted sugarcane samples and identified as *Fusarium moniliforme* S6, S7 and L11 according to morphological characteristic and ITS region of rDNA sequencing. A total of 85 actinomycete isolates were isolated from the sugarcane rhizospheric soil samples by using starch casein agar medium and screened for antagonistic properties using dual culture technique. The results showed that the actinomycete isolate S1-20 gave the highest inhibition of the mycelial growth of *F. moniliforme* S6 (76.45%), S7 (82.88) and L11 (84.41%). The isolate S1-20 was able to produce degrading enzymes, cellulase and amylase. This results suggested that actinomycetes isolate S1-20 may have the potential to be control wilt disease in sugarcane.
.

Keywords: actinomycetes, wilt disease, sugarcane, *Fusarium moniliforme*

1. Introduction

Sugarcane (*Saccharum* sp.) is a major crop cultivated in tropical and sub-tropical countries like Brazil, China, India, Thailand and Australia (1). Wilt is one of important fungal disease which causes sugarcane crop losses. Tissue discolouration and boat-shaped cavity formation are relatively more prevalent in the effected canes.Wilted canes may show root borer tunnel with borer larva inside, mostly at lower internodes. The fungus *Fusarium moniliforme* (teleomorph *Fusarium verticillioides*) and *Cephalosporium sacchari* were associated with wilt disease (2, 3). Besides sugarcane, the *F. moniliforme* also causes the disease in maize, sorghum and rice, and produce different mycotoxins (fumonisins, moniliformin and beauvericin) (4). Chemical fungicides are used to control plant diseases. In the recent years, search for alternatives to chemical control of fungal pathogens, such as biological control, gained great interest due to the emergence of fungicide resistant pathogens, irreversible environmental depletion and health concerns for both producers and consumers (5). Microbial control agents have been proposed as a more effective and environmentally friendly means of controlling soilborne diseases (6). The mechanisms of biocontrol agents are mycoparasitism, competition for space and nutrients, stimulation of the plant's defensive capacity, and secretion of bioactive compounds such as antibiotics and cell wall degrading enzymes (7, 8)
Actinomycetes are an important group of gram-positive, filamentous bacteria that play an important role in the rhizosphere by secreting a wide range of antimicrobial products, thus preventing the growth of common root pathogens. Actinomycetes, particularly several species of *Streptomyces*, have been isolated and used to control plant diseases such as the control of sheath blight of rice caused by *Rhizoctonia solani* (9) stem rot disease of chilli caused by *Sclerotium rolfsii* (7), seedling blight of barley caused by *Fusarium culmorum* (5), *Fusarium*

head blight of cereal crops caused by *Fusarium graminearum* (10). Besides biocontrol agents, actinomycetes have been proved to play vital roles as plant growth promoters, disease resistance inducers, drought tolerance stimulators (11).
The objectives of this present study were to isolate actinomycetes from rhizosphere soils of sugarcane, and determine their *in vitro* antagonism against fungal which isolate from wilted sugarcane, for their possible use as biocontrol agents against the fungi causing wilt disease.

2. Materials and Methods

2.1 Isolation of actinomycetes

Rhizospheric soils (10 samples) were collected from commercial sugarcane fields in Lopburi province. Soil pH was determined. Ten grams of rhizospheric soil from each sample were suspended in 90 ml of normal saline (0.85% of NaCl) and placed on an orbital shaker (at 100 rpm) at room temperature (28-30°C). The soil suspensions was heated at 50 °C for 6 min. Subsequently, 0.1 ml of diluted soil suspension were spread onto starch casein agar plates and incubated at room temperature for 4-10 days (12, 13). Colonies with different morphological characteristics were picked and streaked onto yeast extract malt extract agar plates for purification. Stock cultures were maintained in 20% (v/v) glycerol at -20^{o}C.

2.2 Isolation and identification of plant pathogenic fungi

The wilt infected stalk samples collected from commercial sugarcane fields in Lopburi province were isolated fungal pathogens. The plant parts were cut and surface sterilized using 10% sodium hypochlorite and soaked into sterile distilled water. And then the plant parts placed on potato dextrose agar (PDA) and incubated at room temperature until mycelial growth was observed from the plant parts. The isolated fungi were purified by point technique on PDA plates. The morphological characteristics used for identification were the shapes, formation of macroconidia, microconidia and pigmentation. The fungal isolates were maintained on PDA at 4^{o}C and as conidial suspensions stored in 20% glycerol at -20 ^{o}C.

Genomic DNA were extracted from mycelia grown in potato dextrose broth, incubated with shaking at 220 rpm for 7 days and harvested by filtration through filtering cloth and then stored at -80^{o}C (14). The fungal mycelial were extracted by CTAB method. The ITS region was the amplified by PCR technique using pair of ITS primer which were ITS1 (5' TCCGTAGGTGAACCTGCGG 3') and ITS4 (5'TCCTCCGCTTATTGATATGC 3') (15). The PCR products were sequenced by commercial sequencing company, Solgent Co, Ltd (Korea). The ITS sequences were compared with known sequences in the GenBank database using Basic Local Alignment Search Tool (BLAST) (www.ncbi.nlm.nih.gov).

2.3 *In vitro* antagonistic bioassay

All actinomycete isolates obtained from rhizospheric soils were screened for their antagonistic activity against *Fusarium moniliforme*, using a dual culture technique. The actinomycete colony on yeast extract malt extract agar was cut into a mycelium disc (5 mm in diameter) and transferred to the PDA plates, placed 3 cm away from the center of the plate. After incubation at room temperature for 7 days, a fungal disc (5 mm in diameter) was placed in the center of the plate. PDA plates with a fungal mycelia disc in the center of the plate were served as a control. The plates were incubated at room temperature until the radial growth of the fungal mycelium of the control plate reached 3 cm. The percentage of inhibition was

calculated as [(r1 − r2)/r1] ×100, where r1 is the radial of fungal mycelial growth in the control and r2 is the radial of fungal mycelial growth in the dual culture (11, 16). All isolates were tested in triplicate.

2.4 Morphology characterization of actinomycete isolate S1-20

The selected actinomycete isolate S1-20 was studied spore morphology, substrate and aerial hyphae, pigment production and colony characteristics by observed following growth on oat meal agar plates. The microscopic characterization was performed by slide culture method.

2.5 Hydrolytic enzyme production

2.4.1 Protease production

Actinomycete was streaked on casein agar (12) and incubated at room temperature for five days. At the end of the incubation, the plates was observed for clear zone around the colony.

2.5.2 Cellulase production

Actinomycete was streaked on carboxymethyl cellulose (CMC) agar and incubated at room temperature for five days. After incubation culture plates was flooded with 0.1% Congo red solution for 15 minutes and washed by flooding with 1 M NaCl for 10 minutes. A clear zone formation around the colony indicates the hydrolysis of CMC (17).

2.5.3 Amylase production

Actinomycetes was streaked on starch agar and incubated at room temperature for five days. After incubation culture plates were flooded with iodine solution and observed for halo zone around the colony.

3. Results

3.1 Isolation of actinomycetes

From 10 sugarcane rhizosphere soils, 85 isolates of actinomycete were obtained (Table 1). Rhizosphere soils collected from many area in Lopburi province were sandy loam with slightly acidic to alkaline pH (5.44-8.04). The numbers of actinomycetes from each sample were varied. The highest number of 15 isolates was recovered from sample number 7. The isolated colonies had a weft of aerial mycelium that either appeared floccose, granular or powdery. Each of the isolates were morphology colony colour included dark grey, grey, dark brown, brownish, and yellowish white.

Table 1 Number of actinomycete isolates obtained from sugarcane rhizospheric soil samples in Lopburi province.

Sample No.	Source of rhizoshere soils	soil pH	No. of actinomycete isolates
1	Chai Badan 1	7.50	7
2	Chai Badan 2	6.80	5
3	Nong Muang1	7.16	12
4	Nong Muang2	7.79	8
5	Khok Charoen1	7.57	6
6	Khok Charoen2	7.61	10
7	Sa Bot	7.50	15
8	Lam Sonthi	5.44	11
9	Tha Luang1	8.04	4
10	Tha Luang2	7.65	7

3.2 Fungal isolation

The three fungal pathogen isolates (isolate S6, S7 and L11) were isolated from the wilt infected stalk samples using PDA. The fungal colonies on PDA were appeared wolly and cottony with radial growth and have shown white and purple violet colony were observed. All fungal isolates, microconidia apiculate were formed in false heads and macroconidia were fusoid with an elongated, sharply curved apical cell. The BLAST search results indicated that the ITS sequences of fungal isolate S6, S7 and L11 were closely related to *Fusarium moniliforme* Genbank accession number KX385056, KJ028004 and KJ000438 respectively with 99% identity.

3.3 *In vitro* antagonistic bioassay

The antagonistic effect of actinomycete isolates against *F. moniliforme* which isolated from wilt infected stalks were evaluated using the dual culture technique. Table 2 showed the percentage of *F. moniliforme* S6, S7 and L11 inhibition by isolated actinomycetes. From a total of 85 actinomycete isolates from sugarcane rhizosphere soils in Lopburi province, only 2 isolates, namely S1-20 and C1-2 displayed high antifungal activity to inhibit *F. moniliforme* s7 and L11 more than 50%. Among these, isolate S1-20 showed the highest inhibition of mycelial growth of *F. moniliforme* S6 (76.45%), S7 (82.88) and L11 (84.41%). The results showed that isolate S1-20 inhibited mycelia growth of tested *F. moniliforme* in PDA, the colonies of *F. moniliforme* were stunted and deformed (Figure 1). Isolate S1-20 which showed the highest antifungal activity was selected for further study.

Table 2 Antifungal activity of actinomycete isolates against *F. moniliforme*

Isolate	Inhibition (%)		
	F. moniliforme S6	*F. moniliforme* S7	*F. moniliforme* L11
S1-20	76.45	82.88	84.41
C1-2	50.34	61.08	63.87

3.4 Morphological characteristics of actinomycete isolate S1-20

The actinomycete isolate S1-20 could grow well on oat meal agar plates and produced water soluble dark brown pigment. The aerial mycelium was white on and reverse side color was dark yellow. The spore chains were spiral type when observed under the light microscope. (Figure 2)

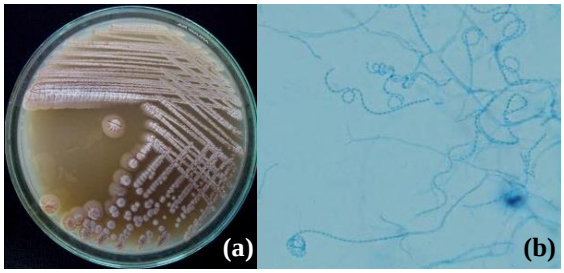


Figure 2 a) Growth pattern of actinomycete isolate S1-20 on oat meal agar media. (b) Microscopic observation of isolate S1-20 under the light microscope (1,000x).

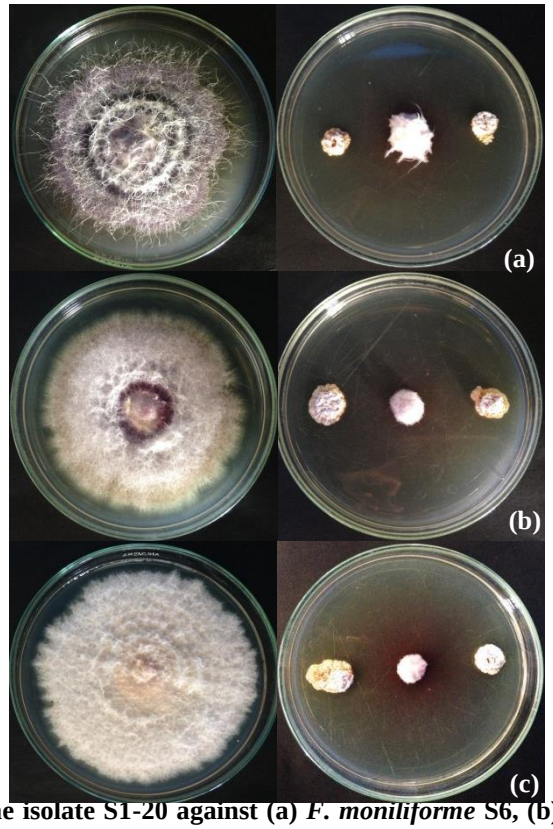


Figure 1 Antagonisms of the isolate S1-20 against (a) *F. moniliforme* S6, (b) *F. moniliforme* S7 and (c) *F. moniliforme* L11 (right) in comparison to controls (left).

3.5 Hydrolytic enzymes production

Assay of hydrolytic enzymes indicated that the actinomycetes isolate S1-20 produced cellulase and amylase by halo zone on plate (Figure 3) but not produced protease.

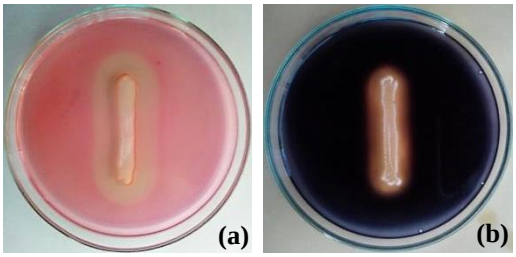


Figure 3. Production of hydrolytic enzyme by isolate S1-20. (a) Production of cellulase. (b) Production of amylase.

4. Discussions

Currently, there are widespread studies of using actinomycetes as a biocontrol agent against several fungal pathogens in various agricultural crops to reduce the use of chemicals (6, 10, 18). Antagonistic activity against fungi can be explained by several mechanisms, including antibiosis, parasitism, hydrolytic enzymes such as chitinases, glucanases or proteases, may act against the fungal cell-wall (19). In this study, from the 85 actinomycete isolates obtained from the sugarcane rhizosphere soils in Lopburi province, only 2 of them (2.35%) were found to have the antagonistic potential against *F. moniliforme* s7 and L11 more than 50% in dual culture assay. The isolate S1-20 showed the highest inhibition of mycelial growth of all tested *F. moniliforme* and there were no direct contact between fungal pathogens and actinomycete colonies in the dual-culture assay. Hence, the actinomycete S1-20 may be produced antifungal substances involved in the inhibition of mycelial growth as biocontrol agents against fungal pathogens (12). Future, the production of hydrolytic enzyme, cellulase and amylase of isolate S1-20 also act as biocontrol agents on cell wall-bearing pathogens. There are various mechanisms involved in inhibiton of fungal pathogens. Pattanapipitpaisal and Kamlandharn (7) reported *Streptomyces hygroscopicus* PACCH24 produced chitinase which catalyze the degradation of chitin, resulting in inhibition of *Sclerotium rolfsii* growth caused stem rot disease of chilli. Harikrishnan et al. (18) reported *Streptomyces aurantiogriseus* VSMGT1014 was found to produce IAA, siderophore, volatile substances and lytic enzymes such as chitinase, cellulase, protease, gelatinase, amylase, pectinase and solubilise phosphorus that involved in the primary mechanism of pathogen inhibition as used by plant growth promoting rhizobacteria (PGPR). Thus, further study needs to be addressed regarding the mechanism of action. Antifungal activity of the isolate S1-20 found in this study is importance as a candidate for further investigation in biological control of the sugarcane wilt disease.

5. Conclusion

Results of the present study indicated that actinomycete S1-20 has a potential as biocontrol agent against *F. moniliforme* that causing wilt disease in sugarcane. Further investigations need to be performed to confirm the biological control effectiveness of isolate S1-20 in field conditions focusing on fungal pathogen protection in sugarcane crop.

6. Acknowledgement

We thank Faculty of Science and Technology, Thepsatri Rajabhat University for financial support.

7. References

(1) Sindhu R, Gnansounou E , Binod P, Pandey A. Bioconversion of sugarcane crop residue for value added products. An overview. Renew. Ener. 2016;98:203-215.
(2) Viswanathan R, Poongothai M, Malathi P, Sundar AR. Sugarcane wilt: new insights into pathogen identity, variability and pathogenicity. Funct Plant Sci and Biotechnol. 2012;6(special Issue 2):30-39.
(3) Gawade DB, Pawar BH, Gawande SJ Vasekar VC. Antagonistic effect of *Trichoderma* against *Fusarium moniliformae* the causal of sugarcane wilt. American-Eurasian J Agric & Environ Sci 2012;12 (9): 1236-1241.
(4) Vishwakarma, SK, Kumar P, Nigam A, Singh A, and Kumar A. Pokkah boeng: An emerging disease of sugarcane. J Plant Pathol Microb. 2013;4(3): 1-5.

(5) Yekkour A, Sabaou N, Zitouni A, Errakhi R, Mathieu F, Lebrihi. A. Characterization and antagonistic properties of *Streptomyces* strains isolated from Saharan soils, and evaluation of their ability to control seedling blight of barley caused by *Fusarium culmorum*. Lett. Appl. Microbiol. 2012;55: 427-435.
(6) Xue L, Xue Q, Chen Q, Lin C, Shen G, Zhao J. Isolation and evaluation of rhizosphere actinomycetes with potential application for biocontrol of *Verticillium* wilt of cotton. Crop Prot. 2013; 43: 231-240.
(7) Pattanapipitpaisal P and Kamlandharn R. Screening of chitinolytic actinomycetes for biological control of *Sclerotium rolfsii* stem rot disease of chilli. Songklanakarin J. Sci. Technol. 2012; 34(4): 387-393.
(8) Intra B, Mungsuntisuk I, Nihira T, Igarashi Y, Panbangred Watanalai. Identification of actinomycetes from plant rhizospheric soils with inhibitory activity against *Colletotrichum* spp., the causative agent of anthracnose disease. BMC Research Notes 2011;4:(98)1-9. doi: 10.1186/1756-0500-4-98.
(9) Boukaew S, Prasertsan P. Suppression of rice sheath blight disease using a heat stable culture filtrate from *Streptomyces philanthi* RM-1-138. Crop Prot. 2014;61: 1-10.
(10)Jung B, Park SY, Lee YW, Lee J. Biological efficacy of *Streptomyces* sp. strain BN1 against the cereal head blight pathogen *Fusarium graminearum*. Plant Pathol. J. 2013;29(1):52-58.
(11)Himaman W, Thamchaipenet A, Pathom-aree W, Duangmal K. Actinomycetes from *Eucalyptus* and their biological activities for controlling *Eucalyptus* leaf and shoot blight. Microbiol Res. 2016;188-189:42–52.
(12)Gopalakrishnan S, Pande S, Sharma M, Humayun P, Kiran BK, Sandeep D, Vidya M S, Deepthi K, Rupela Om. Evaluation of actinomycete isolates obtained from herbal vermicompost for the biological control of *Fusarium* wilt of chickpea. Crop Prot. 2011;30:1070-1078.
(13)Prapagdee B, Kuekulvong, Mongkolsuk S**.** Antifungal potential of extracellular metabolites produced by *Streptomyces hygroscopicus* against phytopathogenic fungi. Int. J. Biol. Sci. 2008; 4:330-337.
(14)Covarelli L, Stifano S, Beccari G, Raggi L, Lattanzio VMT, Albertini E. Characterization of *Fusarium verticillioides* strains isolated from maize in Italy: fumonisin production, pathogenicity and genetic variability. Food Microbiol. 2012;31:17-24.
(15)Lin Z, Zhang Y, Que Y, Chen R, Chen B, Zhang M. Characterization of *Fusarium Verticillioide* isolates from pokkah boeng on sugarcane and the disease incidence in field. J Microbiol Exp. 2015;2(5): 00061. doi: 10.15406/jmen. 2015.02.00061.
(16)Khamna S, Yokota A, Lumyong S. Actinomycetes isolated from medicinal plant rhizosphere soils: diversity and screening of antifungal compounds, indole-3-acetic acid and siderophore production. World J Microbiol Biotechnol. 2009;25:649–655.
(17)Rathore SS, Mannivannan A, Narendhirakannan RT. Screening of cellulase producing microorganisms from lake area containing water hyacinth for enzymatic hydrolysis of cellulose. J Adv Sci Res. 2014; 5(3): 23-30.
(18)Harikrishnan H, Shanmugaiah V, Balasubramanian N, Sharma MP, Kotchoni SO. Antagonistic potential of native strain *Streptomyces aurantiogriseus* VSMGT1014 against sheath blight of rice disease. World J Microbiol Biotechnol. 2014; 30:3149–3161.
(19)Gomes RC, Semeˆdo LTAS, Soares RMA, Alviano CS, Linhares LF, Coelho RRR. Chitinolytic activity of actinomycetes from a cerrado soil and their potential in biocontrol. Lett. Appl. Microbiol. 2000; 30: 146–150.

BACTERIAL CELLULOSE PRODUCTION FROM GAC JUICE

Karan Phromthep[1,a*], Suwimon Katakul[1,b], Phinthida Na Thaisong[1,c], Tawanhatai Klaykan[1,d] and Niphaporn Suksawat[1,e]

Department of Food Science and Technology, Faculty of Science and Technology, Kanchanaburi Rajabhat University, Kanchanaburi, 71190, Thailand

Keywords: Bacterial cellulose, Gac fruit, *Acetobacter xylinum*

Abstract

The objectives of this study were to investigate the optimum formulas for bacterial cellulose (BC) production from gac juice using *Acetobacter xylinum.* The experiment compared six different formulas i.e., three levels of starter culture (10, 15 and 20 %, v/v) and two levels of sucrose (5 and 10%, w/v). After 10-day incubation period, the thickness of BC from gac juice formulas supplemented with 10% (w/v) sucrose and 15 and 20% (v/v) starter culture showed at 10.08 and 10.15 mm., respectively. In the chemical composition analysis, BC samples from gac juice formulas supplemented with 10% (w/v) sucrose and 15 and 20% (v/v) starter culture showed the highest crude fiber ($p \leq 0.05$) in crude protein, the total fat and ash were not significantly different ($p > 0.05$). Sensory evaluation of BC samples from gac juice showed that the highest scores of overall liking characteristics were gac juice formulas supplemented with 10% (w/v) sucrose, and 20 and 15% (v/v) starter culture, 7.77 and 7.70, respectively.

1. Introduction

Bacterial cellulose is a product of carbon metabolism synthesized by *Acetobacter xylinum*, and is one of the traditional foods or desserts generally consumed by people in several Asian countries including the Philippines, Indonesia, Thailand, Japan and Taiwan [1]. Interestingly, various food and beverage products could be supplemented with bacterial cellulose; for instance, ice-cream, yogurt, fruit juice, etc. Otherwise, BC could be multifunctional food ingredients of great medicinal benefits, as follows, (1) improving the rheology of food, (2) producing low-calorie food ingredient products, and (3) producing low-cholesterol products [2]. Several studies were also conducted as to investigate the production of bacterial cellulose from other materials; for example, several types of fruit juice including orange, pineapple, apple, Japanese pear, and grape [3] or food industrial by-products including citrus peel and pomace [4], cashew tree residues [5], dry olive mill residue [6]. Gac or Spiny Bitter Cucumber (*Momordica cochinchinensis* Spreng) is a tropical plant found in several countries within tropical zone, especially East and Southeast Asia region [7]. A gac fruit belongs to the melon family (*Cucurbitaceae*), it usually turns reddish when ripen.The gac fruit is rich in fatty acids and carotenoids (lycopene and β-carotene), abundantly found in aril around the seeds [8]. The fruit was claimed to be a medicinal plant according to its benefits to lower the risk of prostate and lung cancer, coronary heart disease, and eye diseases [9,10,11,12].

Generally, bacterial cellulose lacks in natural antioxidants and vitamins, the gac fruit contains benefits from natural lycopene and β-carotene. Thus, this study applied a new potential substrate from gac juice in bacterial cellulose production to increase its product nutritional value for costumers.

2. Materials and Methods

2.1 Microorganism and Starter Preparation

Acetobacter xylinum was obtained from the Institute of Food Research and Product Development, Kasetsart University, Thailand. The starter medium preparation used matured coconut water supplemented with sucrose 5% (w/v), adjusted pH 4.0 using 5% (v/v) glacial acetic acid, and sterilized medium using autoclave at 121°C for 15 minutes. The inoculated medium was incubated at 37°C for 7 days before scaling up, and then the BC pellicle showed on the top of medium [13].

2.2 Gac Juice Preparation

Ripen Gac fruits with orange-red color were obtained from fresh market in Mueang district, Kanchanaburi province. The gac fruit preparation was conducted following the method reported by [14]. The fruits were cleaned and washed by tap water. Then, the gac aril and pulp were collected from the inner part of each fruit. The preparation adapted the method formulated by [15], the gac aril and pulp were mixed with drinking water, 1: 2 ratio by weight, and blended together using laboratory blender for 5 minutes. The juices were pasteurized at 80°C for 20 minutes and kept in a sealed container, then immediately cooled and stored at 4°C for further experimental investigation.

2.3 Study of Optimum Formulas for BC Production from Gac Juice

2.3.1 Production of BC from Gac Juice

Six different formulas of BC from gac juice were shown in Table 1, this experiment divided the starter culture into 3 levels (10, 15, and 20%, (v/v)), and the sucrose amount into 2 levels (5 and 10% , (w/v)). In the process of BC production, the prepared gac juice was supplemented with varied sucrose, 0.5% ammonium sulfate, and the medium were pasteurized at 80°C for 20 minutes.

Then, medium was immediately cooled and adjusted pH to 4.0 using 5% (v/v) glacial acetic acid. Each amount of starter cultures was added and the inoculated medium was poured into sterilized plastic tray (15×25×8 cm.; W×L×H) and covered by muslin cloth. The BC samples were statically incubated at room temperature for 10 days.

Table 1. Six differences of bacteria cellulose formulas

BC Formulas	Starter culture (%, v/v)	Sucrose (%, w/v)
1	10	5
2	10	10
3	15	5
4	15	10
5	20	5
6	20	10

2.3.2 Study of Physicochemical Properties of Bacterial Cellulose from Gac Juice

The BC pellicles were formed after incubation for 10 days, then collected and the physicochemical properties of bacterial cellulose samples were analyzed. The physical characteristics i.e., the thickness of BC was measured by using Vernier caliper, the color (ΔL^* , Δa^* , Δb^*) was measured by HunterLab (ColorFlex). The chemical compositions of BC samples i.e., moisture contents, total fat (extraction system), crude protein (Kjeldahl Method), crude fiber, ash were analyzed [14].

2.4 Sensory Evaluation of Bacterial Cellulose from Gac Juice

In the preparation process of six various BC pellicle samples, the pellicles were cut into 1.0 cm^3 cubes and cooked in boiling water for 5 minutes. The cubes were washed and boiled for 3 times to removed sour taste, and then the cooked BC cubes were submerged in the 15% (w/v) syrup for 24 hours before analyzing. Sensory evaluation was measured using a 9-point hedonic scale [17]. The 30 panelist members compared six various BC samples and carried out to estimate product-specific characteristics i.e., appearance, color, odor, flavor, texture, and overall liking.

2.5 Statistical Analysis

The study of physicochemical analysis (thickness, color measurement, and chemical compositions) was statistically planned as 3×2 factorial experiment in completely randomized design (CRD) and the sensory analysis was also planned as 3×2 factorial experiment in randomized complete block design (RCBD). Results were presented using ANOVA (analysis of variance) and computed by SPSS for Windows (SPSS, Chicago, Ill., U.S.A.). Duncan's New Multiple Range Test (DMRT) was used to determine significant differences among results, and statistical significance was accepted at 95%, with probability ($p \leq 0.05$).

3. Results and Discussions

3.1 Physicochemical Properties of BC from Gas Juice

Table2. Thickness and color parameter of BC from gac juice with six various formulas after the 10-day incubation period

Formulas	Thickness (mm)	Color parameter		
		ΔL*	Δa*	Δb*
1	7.05±0.60[c]	63.64±1.99 [a]	14.36±1.92 [a]	29.30±4.10 [a]
2	9.05±0.42[b]	63.99±2.72 [a]	13.70±3.83 [a]	28.06±6.79 [a]
3	7.06±0.11[c]	62.47±1.08 [a]	17.20±1.38 [a]	34.16±2.82 [a]
4	10.08±0.03[a]	63.50±3.65 [a]	16.08±5.48 [a]	33.81±1.87 [a]
5	8.96±0.98[b]	61.82±1.39 [a]	18.82±1.87 [a]	38.21±5.40 [a]
6	10.15±0.42[a]	63.34±0.01 [a]	16.18±0.54 [a]	34.66±2.80 [a]

Different letters in the same column denote significant differences according to the DUNCAN test ($p \leq 0.05$)
Values are the means ±standard deviation ($n = 3$)

After the 10-day incubation period, the BC pellicles were formed and the thickness were shown around 7.0 to 10.0 mm (table 2). When the six various formulas were compared, it was found that the thickness of formulas 4 and 6 were greater than other formulas; 10.08 and 10.15 mm, respectively ($p \leq 0.05$). BC from coconut fermentation formed 8 mm of thickness after the 11-day incubation period [18], and the optimum sucrose for BC production medium was10% (w/v), the BC starter culture amount 10 to 20% (v/v) was suitable for BC production from coconut water [19]. Whereas, the amount of starter culture and sucrose did not affect the color characteristics of each BC pellicle sample and the color parameter values (ΔL*, Δa*, Δb*) were not significantly different ($p > 0.05$).

3.2 Chemical Composition of BC from Gac Juice

Table 3. Chemical compositions of BC from six various gac juice formulas

Formulas	Moisture content (%)	Ash (%)	Fat (%)	Protein (%)	Fiber (%)
1	94.02 ± 0.76^{b}	0.11 ± 0.04^{b}	0.14 ± 0.05^{a}	0.12 ± 0.00^{a}	0.90 ± 0.06^{c}
2	96.11 ± 1.56^{a}	0.18 ± 0.01^{ab}	0.16 ± 0.00^{a}	0.16 ± 0.06^{a}	1.03 ± 0.03^{b}
3	94.12 ± 0.71^{b}	0.13 ± 0.06^{ab}	0.14 ± 0.03^{a}	0.20 ± 0.08^{a}	0.93 ± 0.05^{c}
4	96.52 ± 0.83^{a}	0.18 ± 0.03^{ab}	0.17 ± 0.03^{a}	0.15 ± 0.05^{a}	1.12 ± 0.06^{ab}
5	94.52 ± 0.00^{b}	0.14 ± 0.01^{ab}	0.17 ± 0.03^{a}	0.19 ± 0.04^{a}	0.92 ± 0.01^{c}
6	96.98 ± 0.16^{a}	0.20 ± 0.01^{a}	0.17 ± 0.01^{a}	0.19 ± 0.02^{a}	1.15 ± 0.02^{a}

Different letters in the same column denote significant differences according to the DUNCAN test ($p \leq 0.05$)
Values are the means ±standard deviation ($n = 3$)

Chemical compositions of BC from six various gac juice formulas were shown in table 3. These pellicle samples showed no significant differences in the amount of crude protein and fat. In ash content, it showed the highest value in BC sample from formula6.

According to BC samples formulas 2, 4 and 6, it pointed out the higher moisture contents than formulas 1, 3 and 5 ($p \leq 0.05$), and the results accorded with the crude fiber. Due to the more thickness, more fiber, and high density of BC, it could hold more water or moisture in its structure [19]. In addition, cellulose contained high hydrophilicity and could absorb more water contents in BC structure [20].

3.3 Sensory Evaluation of Bacterial Cellulose from Gac Juice

Table 4. Sensory characteristics among six various BC formulas evaluated by 30 panelists using 9-point hedonic scale

Formulas	Appearance	Color	Odor	Flavor	Overall liking
1	5.93 ± 1.53^{c}	6.40 ± 1.25^{c}	6.67 ± 1.49^{a}	7.03 ± 1.30^{b}	6.87 ± 0.97^{b}
2	7.00 ± 1.23^{b}	6.43 ± 1.33^{bc}	6.63 ± 1.33^{a}	7.23 ± 1.04^{b}	7.07 ± 0.98^{b}
3	6.07 ± 1.39^{c}	6.70 ± 1.32^{bc}	6.80 ± 1.47^{a}	6.90 ± 1.56^{b}	6.93 ± 1.05^{b}
4	7.63 ± 1.00^{a}	6.80 ± 1.42^{ab}	6.70 ± 1.47^{a}	7.70 ± 0.84^{a}	7.70 ± 0.84^{a}
5	6.23 ± 1.19^{c}	6.57 ± 1.30^{bc}	6.60 ± 1.33^{a}	7.23 ± 1.07^{b}	7.03 ± 0.89^{b}
6	7.37 ± 1.61^{ab}	7.10 ± 1.69^{a}	6.93 ± 1.36^{a}	7.80 ± 1.03^{a}	7.77 ± 1.57^{a}

Different letters in the same column denote significant differences according to the DUNCAN test ($p \leq 0.05$)
Values are the means ±standard deviation ($n = 30$)

Sensory evaluation of six various BC formulas was measured using a 9-point hedonic scale and the scores of each sensory characteristic were evaluated by 30 panelists (table 4) . There were no significant differences of color and odor characteristics ($p > 0.0.5$).Appearance characteristics of BC formulas 2, 4, and 6 obtained higher sensory scores than formulas 1, 3, and 5 ($p \leq 0.05$).The overall liking characteristic score of BC samples from gac juice (formulas 4 and 6) reported higher mean

scores than other formulas, 7.70 and 7.77, respectively ($p \leq 0.05$). In addition, the BC formulas 4 and 6 showed higher scores than formula 7, and both formulas were accepted by 30 panelists.

4. Conclusion

According to the study of bacterial cellulose from gac juice, it was found that the optimum sucrose content was 10% (w/v) and optimum starter culture was at least 15% (v/v). After 10-day incubation period, the pellicles of each BC were formed, and the best formulas provided the 10-mmthickness or greater. In the chemical composition analysis, the BC formula 6 with 20% (v/v) starter culture and 10% (w/v) sucrose content showed the highest moisture content and crude fiber. Additionally, the sensory evaluation results of the formulas showed the highest scores in all sensory attributes, and the results were accepted by 30 panelists.

5. Acknowledgments

I would like to gratefully acknowledge the Department of Food Science and Technology, Faculty of Science and Technology, Kanchanaburi Rajabhat University, Thailand for productive supports and facilitation in this research study.

6. References

[1] S. Kongruang, *Bacterial cellulose production by Acetobacter xylinum strains from agricultural waste products*, Applied Biochemistry and Biotechnology, 148 (2008) 245-256.

[2] Z. Shi, Y. Zhang, G.O. Phillips, G. Yang, Utilization of bacterial cellulose in food, Food Hydrocolloids 35 (2014) 539-545.

[3] A. Kurosumi, C. Sasaki, Y. Yamashita, Y Nakamura. Utilization of various fruit juices as carbon source for production of bacterial cellulose by *Acetobacter xylinum* NBRC 13693. Carbohydrate Polymers. 76(2009) 333-335.

[4] X. Fan, Y. Gao, W. He, H. Hu, M. Tian, K. Wang, S. Pan, Production of nano bacterial cellulose from beverage industrial waste of citrus peel and pomace using *Komagataeibacter xylinus.* Carbohydrate Polymers 151 (2016) 1068–1072.

[5] G. Pacheco, C.R. Nogueira, A.B. Meneguin, E. Trovatti, M.C.C. Silva, R.T.A. Machado, S.J.L. Ribeiro, E.C. da Silva Filho, H.D.S. Barud, Development and characterization of bacterial cellulose produced by cashew tree residues as alternative carbon source, Industrial Crops & Products 107 (2017) 13–19.

[6] F.P. Gomes, N.H.C.S. Silva, E. Trovatti, L.S. Serafim, M.F. Duarte, A.J.D. Silvestre, C.P. Neto, C.S.R. Freire, Production of bacterial cellulose by *Gluconacetobacter sacchari* using dry olive mill residue. Biomass and Bioenergy 55 (2013) 205-211.

[7] M. Iwamoto, H. Okabe, T. Yamauchi. Studies on the constituents of *Monordica cochinchinensis* Spreng. Chemical and Pharmaceutical Bulletin 33 (1985) 1-7.

[8] X.T. Tran, S.E. Parks, P.D. Roach, J.B. Golding, M.H. Nguyen, Effects of maturity on physicochemical properties of Gac fruit (*Momordica cochinchinensis* Spreng.), Food Science & Nutrition (2016) 305–314.

[9] S. Songtummin, B.* Leenanon, Survival of *Lactobacillus acidophilus* TISTR1338 and *Lactobacillus casei* TISTR390 in probiotic Gac ice cream, International Food Research Journal 23(2016) 790-796.

[10] T.C. Kha, M.H. Nguyen,P.D. Roach, Effects of spray drying conditions on the physicochemical and antioxidant properties of the Gac (*Momordica cochinchinensis*) fruit aril powder. Journal of food engineering 98(2010) 385-392.

[11] D.T.T. Nhung, P.N. Bung, N.T. Ha, T.K. Phong, Changes in lycopene and beta carotene contents in aril and oil of gac fruit during storage. Food Chemistry 121 (2010) 326-331.

[12] H. Phan-Thi, Y. Wache, Isomerization and increase in the antioxidant properties of lycopene from *Momordica cochinchinensis* (gac) by moderate heat treatment with UV-Vis spectra as a marker. Food Chemistry 156(2014) 58-63.
[13] S. Hestrin, M. Schramm, Factors affecting production of cellulose at the air/liquid interface of a culture of *Acetobacter xylinum.* J .Gen .Microbiol 11(1954) 123–129.
[14] T. Auisakchaiyoung, T. Rojanakorn, Effect of foam-mat drying conditions on quality of dried Gac fruit (*Momordica cochinchinensis*) aril, International Food Research Journal 22(2015) 2025-2031.
[15] W. Angkananon, V. Anantawat. Effects of Spray Drying Conditions on Characteristics, Nutritional Value and Antioxidant Activity of Gac Fruit Aril Powder. Rev. Integr. Bus. Econ. Res. 4(2015) 1-11.
[16] AOAC .Official methods of analysis of AOAC international .17th ed .Association of official analytical chemists .Gaithersburg, MD,USA, (2000).
[17] T.K. Majumdar, D.D. Wadikar, C.R. Vasudish, K.S. Premavalli, A.S. Bawa, Effect of Storage on Physico-Chemical, Microbiological and Sensory Quality of Bottlegourd-Basil Leaves juice. American Journal of Food Technology 6(2011) 226-234.
[18] D.A. Nugroho, P. Aji, Characterization of Nata de Coco Produced by Fermentation of Immobilized *Acetobacter xylinum* .Agriculture and Agricultural Science Procedia 3(2015) 278 – 282.
[19] A.A. Jagannath, S.S. Kalaiselvan, P.S. Manjunatha, P.S. Raju, A.S .Bawa, The effect of pH sucrose and ammonium sulphate concentrations on the production of bacterial cellulose (Nata-de-coco) by *Acetobacter xylinum*, World J .Microbiol .Biotechnol, (2008) 2593-2599.
[20] D.G. White, R.M. Brown, Prospects for the commercialization of the biosynthesis of microbial cellulose. In: Schuerech, C (ed) Cellulose and Wood-Chemistry and Technology. Wiley, New York. (1989).

Broken-Milled Riceberry Drinking Yogurt

Nomjit Suteebut [a*], Chayapat Kee-ariyo [b], Katsarin Meetam
Munin Nuchnang, Nopporn Sakulyunyongsuk [c]
and Thanapop Soteyome[d]

Food Science and Technology, Home Economic Technology,
Rajamangala University of Technology Phra Nakhon, Bangkok, Thailand

Abstracts

The objectives of this research were to develop drinking yogurt from broken-milled Riceberry. For Preparation of set yogurt, the cooked rice was mixed with water for 3 different levels. This product compost of broken-milled Riceberry, pasteurized milk, natural plain yogurt sugar, guar gum and xanthan gum. The highest acceptance (9-Point Hedonic Scale) from the panelist showed that broken-milled Riceberry at 10% was proper for this beverage than 5% and 10% ($p \leq 0.05$). There was a water soluble solids at 21°Brix with a pale purple. Yogurt odor was stronger than the smell of rice. The viscosity of the product was increased as broken-milled Riceberry increasing. Then study the ratio between yogurt and syrup for drinking yogurt production, three different levels were used: 30:70, 40:60, and 50:50. The highest acceptance from sensory test showed the best recipe is the formula use a ratio of 50:50 which pH was 4.18, soluble solids were 18 °Brix.This drinking yogurt from broken-milled Riceberry had moisture, ash, fat, protein, fiber and carbohydrate was found to be 82.28±0.58, 0.16±0.15, 0.32±0.34, 0.07±0.00, 0.36±0.04 and 6.81±0.03, respectively. Moreover, it was safe with the microbiological properties at 15 days storage at 4±1 ℃ passed quality standards (Notification of Ministry of Public Health. 353/2013).

Keywords: broken-milled rice, Riceberry, yogurt

1. Introduction

In recent years, consumers are more interested in heath products, especially foods with health promoting such as riceberry with antioxidant properties. Riceberry is a newly rice variety from Thailand originated from a cross -breed between Khao Hom Nin (JHN), the local non-glutinous purple rice, and Khoa Dawk Mali 105 (Jasmine rice), the Thai Hom Mali Rice. Riceberry rice is rich in many antioxidant that help our immune system stay healthy. It contains significant levels of beta-carotene, gama oryzanol, vitamin E, folic acid, tannin, zinc, fiber and bran oil. These antioxidants, vitamins and minerals give riceberry rice a nutrition profile that is relatively unique. The dark purple colour it matures signifies it high nutrition content and gives it a unique appearance when served. [1]

Yogurt (Fermented milk) is one of the dairy product, derived from the animal milk or derivatives of milk that pasteurized for destroying pathogens in milk. Yogurt is a nutritious dairy product [2]. Nowadays, the company has developed a variety of valuable products as an alternative for consumers who love health. Global interest in cereal-based fermented products is increasing due to low fat/cholesterol, high minerals, dietary fibers, and phytochemicals content [3]. Therefore, researchers are interested in increasing the value of rice broken rice with nutritional value, but not expensive. By developing a sour milk from yogurt Rice. To add value to both products to be popular with consumers of all ages. Get the full benefits of the product.

2. Introduction

In recent years, consumers are more interested in heath products, especially foods with health promoting such as riceberry with antioxidant properties. Riceberry is a newly rice variety from Thailand originated from a cross -breed between Khao Hom Nin (JHN), the local non-glutinous purple rice, and Khoa Dawk Mali 105 (Jasmine rice), the Thai Hom Mali Rice. Riceberry rice is rich in many antioxidant that help our immune system stay healthy. It contains significant levels of beta-carotene, gama oryzanol, vitamin E, folic acid, tannin, zinc, fiber and bran oil. These antioxidants, vitamins and minerals give riceberry rice a nutrition profile that is relatively unique. The dark purple colour it matures signifies it high nutrition content and gives it a unique appearance when served. (Rice Science Center, 2016)

Yogurt (Fermented milk) is one of the dairy product, derived from the animal milk or derivatives of milk that pasteurized for destroying pathogens in milk. Yogurt is a nutritious dairy product. Nowadays, the company has developed a variety of valuable products as an alternative for consumers who love health. Global interest in cereal-based fermented products is increasing due to low fat/cholesterol, high minerals, dietary fibers, and phytochemicals content (Blandino et al, 2003). Therefore, researchers are interested in increasing the value of rice broken rice with nutritional value, but not expensive. By developing a sour milk from yogurt Rice. To add value to both products to be popular with consumers of all ages. Get the full benefits of the product.

2. Materials and Methods

2.1 Raw material

Broken-Milled Riceberry (Ban Bangtachom Community, Sing Buri Province)**,** Plain set yogurt (Meiji Bulgaria), Pasteurized whole milk (Meiji), Sugar (Mitr phol), Guar Gum (CTi & SCIENCE Co.,Ltd.) Xantangum (CTi & SCIENCE Co.,Ltd.)

2.2 Quality analysis

2.2.1 Physical analysis: The color of samples were measured by CM-3500d spectrophotometer (Konika Minolta). The total soluble solid was measured by RHB–32 ATC Refractometer. The viscosity was measured by Brookfield

2.2.2 Chemical analysis: pH was measured by pH meter. The Total acidity was measured and expressed as a percentage of lactic acid.

2.2.3 Microbiological analysis: Yeast and mold, coliform, and total plate count [4]

2.2.4 Sensory evaluation

The sensory properties of yogurt was designed as Randomized Complete Block Design (RCBD). Sensory evaluation was conducted by using untrained 50- panelist, who were student at Rajamankala University of Technology. Yogurt samples were scores in terms of color,

aroma, flavor, taste, texture, and overall preference, using the 9–Point hedonic scale ranging from 1 (dislike extremely) to 9 (like extremely). Then, data from this experiment were analyzed by analysis of variance (ANOVA) and differences among treatments were determined by Duncan's New Multiple Range test (DMRT).

2.3 Method

2.3.1 Optimum concentration of broken-milled riceberry milk

Broken-milled riceberry milk was prepared by mixing the boiled broken-milled riceberry with 3 levels of water (5, 10 and 15%). The soaked riceberry was next homogeneously blended, and filtered to remove coarse material. Then, the broken-milled riceberry milk was boiled at 65°C for 15 minutes. Thereafter, treatments were prepared according to the ingredients in Table 1.

Table 1 Composition of the treatments

Ingredients	**Amount (%)**		
	Treatment 1	**Treatment 2**	**Treatment 3**
Broken-Milled Riceberry Milk	63.35 (5%)	63.35 (10%)	63.35 (15%)
Pasteurized whole milk	20	20	20
Plain yogurt	9	9	9
Sugar	7	7	7
Guar gum	0.25	0.25	0.25
Xanthangum	0.4	0.4	0.4

Adapted from [5]

From the formula in Table 1, three different levels of broken-milled riceberry were used to produce set yoghurt from broken-milled riceberry as shown in Figure 1. Then, the set yogurts were analyzed physical qualities, chemical qualities, and sensory evaluation. Take the recipe that was the best recipes for manufacturing drinking yoghurt from broken-milled riceberry.

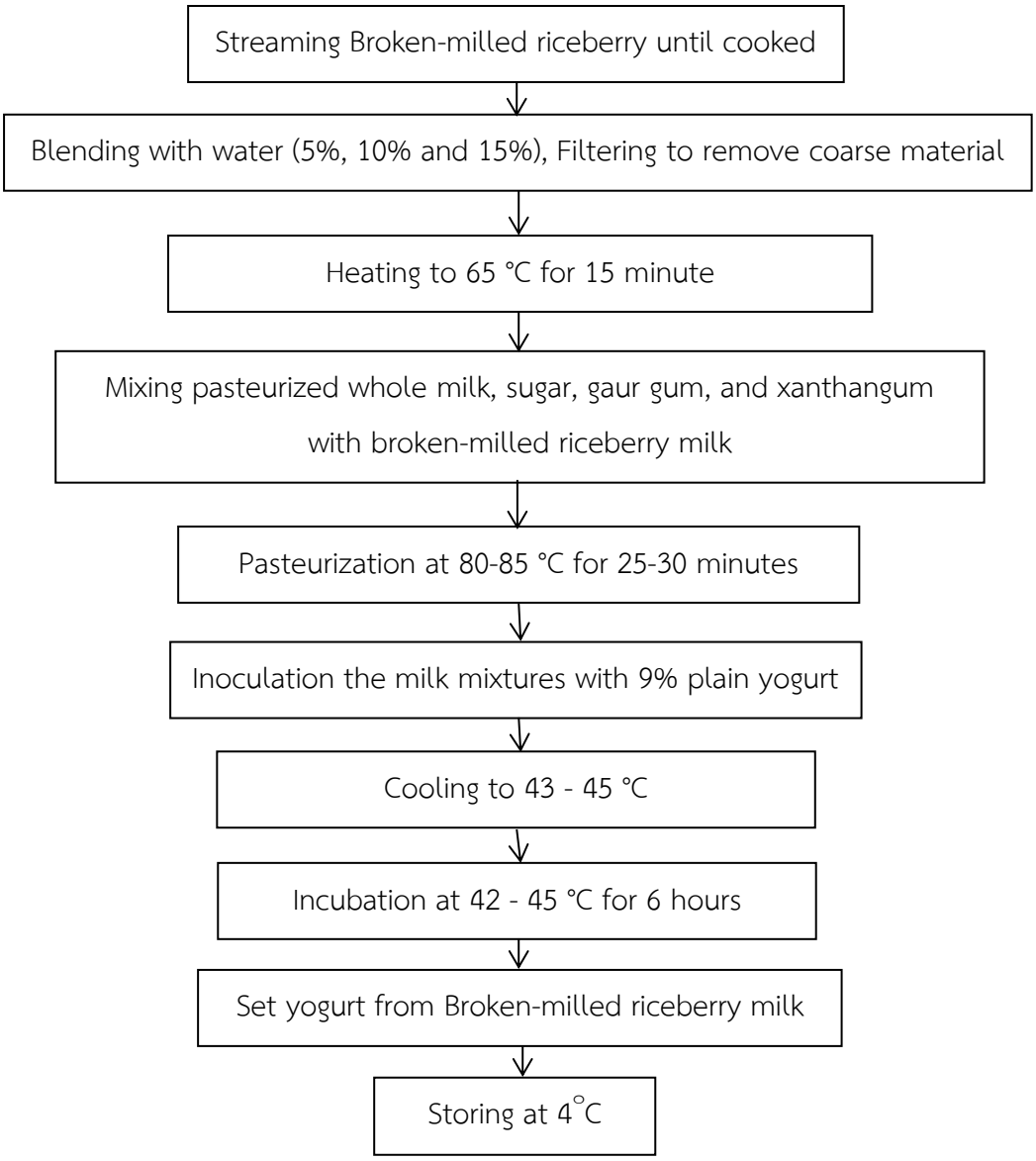


Figure 1 Production steps for set yogurt from broken-milled riceberry
Adapted from Adapted from [5]

2.3.2 Optimum ratio between the set yoghurt from broken-milled riceberry and syrup for producing drinking yogurt

Study on the optimum ratio between the set yoghurt from broken-milled riceberry and syrup, 15˚Brix. The selected formulation of set yogurt obtained from 2.3.1 was examined. The experiment was composed of the different ratio of set yoghurt from broken-milled riceberry to syrup (0:70, 40.:60, and 50:50) was conducted to produce drinking yogurt according to the method in Figure 2, and then to analyze physical, chemical, microbiological and sensory aspects of drinking yogurt

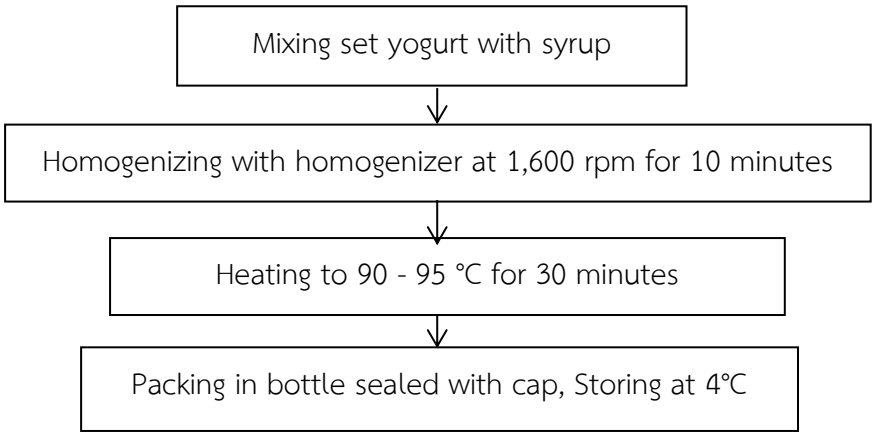


Figure 2 Production steps for drinking yogurt
Adapted from Adapted from [6]

2.3.3 Chemical analysis of broken-milled riceberry drinking yogurt

Analysis of moisture content, protein, ash, fat, fiber, and carbohydrate contents was performed according to [2] .All analyses were carried out in triplicate.

2.4 Shelf life of broken-milled riceberry drinking yogur

Broken-milled riceberry drinking yogurt that was the most acceptance from 3.2, was packed in a plastic bottle with cap, and stored in a refrigerator at 4±1° C for 15 days. Samples were collected every 5 days for 1, 5, 10 and 15 days. Physical, chemical, microbial and sensory qualities were analysed every 5 days.

3. Results and Disscussion

3.1 Optimum concentration of broken-milled riceberry milk

The quality of set yogurts with different concentrations of broken-milled riceberry milk in three treatments were shown in table 2, and the sensory characteristics of three broken-milled riceberry drinking yogurts were shown in table 3.

Table 2 The quality of set yogurts with three different concentrations of broken-milled riceberry milk

Qualities	concentrations of broken-milled riceberry milk		
	5%	10%	15%
Total Soluble Solid (ºBrix)	22.0±0.0[a]	21.0±0.0[b]	19.0±0.0[c]
Color			
- L*	73.81±0.01[a]	72.60±0.07[b]	61.43±0.51[c]
- a*	6.29±0.03[c]	6.68±0.05[b]	10.43±0.04[a]
- b*	7.80±0.03[b]	7.67±0.03[b]	8.40±0.02[a]
pH	4.05±0.02[b]	4.13±0.02[a]	4.13±0.28[a]
Total acidity (%)	0.94±0.01[a]	0.84±0.04[b]	0.81±0.06[b]
	Pale purple	Pinkish purple	Dark purple
	Weak rice flavor	Moderate rice flavor	Strong rice flavor
			

Note: Values in a row within the same group followed by different letters were significantly different treatments ($p \leq 0.05$).

From table 2, when the concentrations of broken-milled riceberry milk was increased, it found that decreases in total soluble solid, L*, and total acidity, and increase in a*, b*, and pH were observed. Color of all treatments had purple and the more concentrations of broken-milled riceberry milk was, the darker color was. Due to anthocyanin was a dark purple pigment, set yogurt had dark purple. [1] [6]

From table 3, the result of sensory evaluation of broken-milled riceberry drinking yogurts with different levels of broken-milled riceberry milk (5%, 10% and 15%) revealed that three treatments were significant difference ($p \leq 0.05$) in all characteristics. The panelists preferred 10% recipe in all attributes so this recipe was selected to next experiment.

Table 3 Sensory characteristics of three broken-milled riceberry drinking yogurts

Sensory characteristics	concentrations of broken-milled riceberry milk		
	5%	10%	15%
Color	6.22±1.09[b]	6.68±0.81[a]	6.44±0.90 [ab]
Aroma	6.34±0.93[a]	6.54±0.93[a]	5.84±0.95[b]
Flavor	5.98±1.23[b]	6.62±0.80[a]	5.76±1.31[b]
Taste	5.64±1.25[b]	6.66±1.06[a]	5.78±1.18[b]
Texture	5.86±1.17[b]	6.82±0.84[a]	6.14±1.09[b]
Overall preference	6.14±1.56[b]	7.52±1.16[a]	5.84±1.16[b]

Note: Values in a row within the same group followed by different letters were significantly different treatments ($p \leq 0.05$).

3.2 Optimum ratio between the set yoghurt from broken - milled riceberry and syrup for producing drinking yogurt

Table 4 revealed that three treatments showed an increasing trend of the physical qualities, especially color (L*, a*, and b*) and viscosity, when more ratio between the set yoghurt from broken - milled riceberry and syrup. Meanwhile, total soluble solid was decreased when amount of syrup was decreased [7].

Table 4 Quality of broken-milled riceberry drinking yogurt with different syrup levels

Qualities	broken-milled riceberry drinking yogurt: syrup		
	30:70	40:60	50:50
Color			
- L*	46.22±0.01[b]	48.21±0.00[a]	48.05±0.01[a]
- a*	10.65±0.00[b]	12.62±0.01[a]	12.30±0.01[b]
- b*	8.43±0.02[c]	9.56±0.01[a]	9.43±0.01[b]
Total Soluble solid (°Brix)	21.00±0.00 [a]	18.40±0.00 [b]	18.00±0.00 [b]
Viscosity (cp)	56.06±0.30 [c]	77.16±0.56 [b]	94.20±0.17 [a]
pH	4.41±0.00 [a]	4.27±0.00 [b]	4.27±0.00 [b]
Total acidity (%)	0.12±0.01[c]	0.21±01.0[a]	0.18±00.0[b]
Yeast and mold (CFU/ml)	<10	<10	<10
Coliform (MPN/ml)	<3	<3	<3
Total plate count (CFU/ml)	<10	<10	<10

Note: Values in a row within the same group followed by different letters were significantly different treatments ($p \leq 0.05$).

The results of the chemical analysis showed that all treatments were significantly difference in total acidity (% lactic acid) and pH.

Analysis of the microbiological qualities showed that total plate count was less than 10 CFU/ml, and the yeast and mold were less than 3 CFU/ml which passed the standard [2] (Notification of Ministry of Public Health 353/2013).

The results of microbiological analysis of drinking milk products after 15 days showed that total plate count was less than 10 CFU/ml, and the yeast and mold were less than 10 CFU/ml which was less than the standard. - it must not exceed 100 colonies per 1 gram sample for non-fermented yogurt. Moreover, the coliform content is less than 3 MPN/ml, which meets criteria. (Industrial Product Standards, 2004).

Table 5 Sensory characteristics of three broken-milled riceberry drinking yogurts with different syrup levels

Sensory characteristics	**Drinking yogurts with different syrup levels**		
	30: 70	**40 : 60**	**50 :50**
Color	6.66±0.98^{b}	6.62±0.92^{b}	7.60±0.96^{a}
Aroma	6.22±0.86^{b}	6.42±0.97^{b}	7.26±0.96^{a}
Flavor	6.22±0.92^{b}	6.42±0.86^{b}	7.26±0.99^{a}
Taste	6.00±0.94^{b}	6.52±1.21^{b}	7.30±1.16^{a}
Texture	6.06±0.86^{b}	6.26±0.75^{b}	7.40±0.92^{a}
Overall preference	5.92±0.82^{b}	6.32±0.93^{b}	7.50±0.81^{a}

Note: Values in a row within the same group followed by different letters were significantly different treatments ($p \leq 0.05$).

Sensory scores presented in Table 5. The panelists preferred ratio of yogurt: syrup at 50:50 to 30:70 and 40:60 in all attributes. Besides, all attributes of both drinking yogurts (30:70 and 40:60) were not significantly different ($P \geq 0.05$). The lower viscosity and higher soluble solid of this drinking yogurt (Table 4) may be the factor responsible for the higher scores. A better taste result of drinking yogurt could have been achieved by adding syrup that reduced the sour taste.

3.3 Chemical analysis of Broken-Milled riceberry Drinking Yogurt

Chemical compositions of broken-milled riceberry drinking yogurt were moisture content of 82.28±0.58 percent, ash of 0.16 ±0.15 percent, fat of 0.32±0.34 percent, protein of 0.17±0.00 percent, protein of 0.34 percent, fiber of 0.36±0.04 percent, and carbohydrate of 16.81±0.03 percent, respectively.

3.4 Shelf life of broken-milled riceberry drinking yogurt

The results of microbiological analysis of drinking milk products after 15 days showed that total plate count was less than 10 CFU/ml, and the yeast and mold were less than 10 CFU/ml which was less than the standard. - it must not exceed 100 colonies per 1 gram sample for non-fermented yogurt. Moreover, the coliform content is less than 3 MPN/ml [2], which meets criteria. (Industrial Product Standards, 2004). Results obtained suggest that broken-milled riceberry drinking yogurt stored at refrigerator temperatures (4 ± 1 °C) can be stored up to 15 days.

Table 6 Qualities of broken-milled riceberry drinking yogurt during storage at 4±1°C

Qualities	Storage time (Days)			
	1	**5**	**10**	**15**
Color				
- L* ^{ns}	48.80±0.06	48.85±0.0	48.83±0.11	48.85±0.0
- a* ^{ns}	12.30±0.02	2	12.55±0.01	2
- b*	9.43±0.01^{b}	12.35±0.0	9.35±0.02^{a}	12.48±0.0
Total soluble solid (°Brix)^{ns}	18.00±0.00	8	18.00±0.00	8
pH	4.18±0.00^{a}	9.32±0.01^{b}	4.13±0.00^{a}	9.34±0.00^{b}
		18.00±0.0		18.00±0.0
		0		0
		4.18±0.02^{a}		3.98±0.10^{b}
Lactic acid (%)^{ns}	0.15±0.00	0.15±0.00	0.16±0.00	0.16±0.01
Total plate count (CFU/ml)	<10	<10	<10	<10
Yeast and mold (CFU/ml)	<10	<10	<10	<10
Coliform (MPN/ml)	<3	<3	<3	<3

Note: Values in a row within the same group followed by different letters were significantly different treatments ($p \leq 0.05$).

^{ns} represents non-significance ($p > 0.05$)

4. Conclusion

The recipe of drinking yogurt, the most acceptance, was made from rice milk with cooked rice content (10% water content). The optimal ratio between the set yoghurt from broken - milled riceberry and syrup for manufacture drinking yogurt was 50:50 percent. The chemical compositions of broken-milled riceberry drinking yogurt were 82.28 ± 0.58 % moisture content, 0.16 ± 0.15% ash, 0.32 ± 0.34% protein, 0.17 ± 0.00% protein, 0.36 ± 0.04% fiber and 16.81 ± 0.03% carbohydrate. Moreover, it was safe with the microbiological properties at 15 day storage at 4±1°C passed quality standards (Notification of Ministry of Public Health. 353/2013).

5. Acknowledgments

Authors would like to thank the Faculty of Home Economics, Research and Development Institute, Rajamangala University of Technology Phra Nakhon for the financial support of investigations presented in this article.

6. References

[1] Information on Rice Science center and rice gene discovery. (2016, August 23) "Riceberry". Kasetsart University. [Online]. Available: http://dna.kps.ku.ac.th/v2016/index.php

[2] Information on Fermented Milk, Notification of the Ministry of Public Health No. 353, 2013.

[3] A. Blandino, M.E. Al-Asceri, S.S. Pandiella, D. Cantero, C. Webb. Cereal based fermented foods and beverages. Food Res Int, 36, 2003, pp. 527–543.

[4] AOAC. Official Methods of Analysis of AOAC International, 17th ed. Gaithersgurg, MD, USA: Official Methods. 2000.

[5] A. Chaikulsareewath, W. Chauykaew, and N. Sakulsong, 2014. Hom Nin Brown Rice Yoghurt Production. Agricultural Sci. J. 45(2) (Suppl.) 2014, pp. 273-276.

[6] A. Chaikulsareewath and O. Korsaked. Production of brown rice drinking Yoghurt. Journal of Food Technology, Siam University. 4 (1) 2008, pp. 34-41

[7] N. Suteebut, T. Yimyong, A. Meethongkham and S. Pusombun. Application of Yam Bean Syrup in Drinking Yogurt Product**.** RMUTP Research Journal, Vol. 11, No. 1, January-June 2017.

Study the ability to tolerate salt and produce proteins based on microorganisms isolated from soy sauce residue

Thanapop Soteyome* , Chayapat Kee-ariyo , Nomjit Suteebut and Nopporn Sakulyunyongsuk

Department of Food Science and Technology, Faculty of Home Economics,
Rajamangala University of Technology Phra Nakhon, Bangkok 10300, Thailand

Abstract

Study the ability to tolerate salt and produce proteins based on microorganisms isolated from soy sauce residue on this time had using Zygosaccharomyces rouxii Y isolated from soy sauce residue. Cultured with agar Yeast extract peptone dextrose (YPD) added sodium chloride, 0%, 5%, 10%, 15%, 20%, 25%, 30% and 35%. The results showed that the yeast can tolerate highest salt in medium added with 15% sodium chloride (Nacl). Maximum cell concentration at 0.947 has 4.167 grams of protein per liter at 288 hours and y east produce highest proteins at 11.590 grams per liter and the concentration of cells Equal to 2.582 in medium that does not add sodium chloride at 144 hours.

Key words: proteins, tolerate salt Yeast

1. INTRODUCTION

Due to the price of energy, Protein shortage and pollution from wastes are increasing; solve only one problem may not have enough effective. The current solution is a combination. For example, animal manure, agricultural wastes, wastes from industrial, particularly the food industry, such as pineapple peel, tomatoes peel, soy sauce Residue etc. Which replace the elimination of these wastes is the most widely used as animal feed zhan li (2009). However, those wastes' nutritionally are not enough for animals. Therefore, if it can be increase the nutritional value of protein in waste used as animal feed, this solution would be a combination of the elimination of waste and pollution along with a lack of protein. In order to solve the problem of shortage of dietary protein source and develop animal feed's product to be effective and efficient, it need to research and development of other protein sources to replace the natural source of protein, which will take a long time to produce. Production of single cell protein is an option that has been interested. because the processing time is short; using the area in production is less compared with the production of natural proteins and high quantities

of protein in the waste has come out with microorganisms, which many types of microorganisms have high protein content.

The soy sauce production process, using the fermentation process by take advantage of the salt tolerant's microorganisms. Making soy sauce is mixed the soybean, wheat flour or rice flour, the flour may be roasted before use, create flavor from the browning reaction. Such as, Maillard reaction or Caramellization then spread the mixture on the threshing by controlling the temperature and humidity, to have good flow of the air at surface that suitable for the growth of microorganisms. Control temperature at 25-35^{o}C incubation for 24 hours. Bring mixture in the first step mixed with salt water then left to fermentation, which is a natural fermentation, no temperature control, the fermentation will take about one year.

The longer it is fermented soy sauce with greater flavor. The temperature control during the fermentation in the range of 35-40^{o}C Yan ling dong (2012), which is the optimum temperature for the growth of microorganisms, will shorten the duration of the fermentation and maturing down. During the fermentation requires mixing periodically to increase the oxygen. At the end of fermentation and maturing, Soy sauce will be separated by filtration may be mixing the filter aid to help speed up the filter. Separated liquid residue may add salt water again for the second fermentation to achieve the second soy sauce which will be low quality and has lightened flavor than first fermentation soy sauce. To make soy sauce safe for microorganisms those cause spoilage and microorganisms those pathogens. Soy sauce after the fermentation will sterilized by pasteurization at a temperature of 70-80^{o}C by pasteurization can be used in container pasteurization, bottled before pasteurization. If produce a large quantities can use in-line pasteurization method Figure 1.

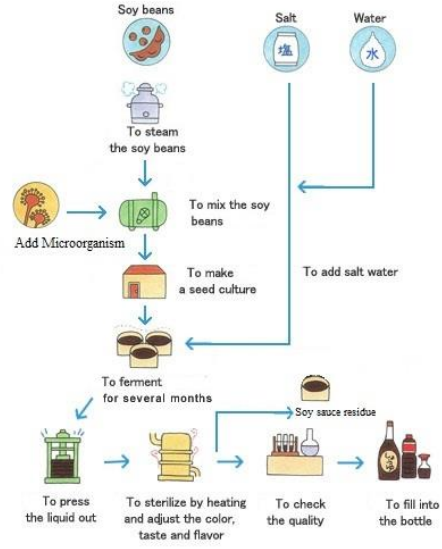


Figure 1. Process of Soy sauce Production

When the product come out it will contained with the waste, also containedwith these microorganisms. These microorganisms have the ability to tolerate salt and dietary protein. If having the study and research of these microorganisms will be able to take advantages and develop protein and pollutants salt in the environments as well.

2. Materials and Methods

2.1 Microorganisms used in the research

Zygosaccharomyces rouxii Y6 which is the salt tolerant yeast and non-flocculation, Courtesy from Asst. Prof. Feng Wen Hu , South China University of Technology, Guangdong, Guangzhou, China.

2.2 Medium

2.2.1 The medium for storage and feed the bold yeast Y6 are Yeast extract peptone dextrose agar (YPD agar) and Yeast extract peptone dextrose (YPD)

2.2.2 Foods for the culturing yeast Y6 to measure the growth and amount of protein. The YPD agar containing sodium chloride, 0%, 5%, 10%, 15%, 20%, 25%, 30% and 35% sterilization temperature is at 121^0c for 15 minutes.

2.3 Yeast preparation

2.3.1 Culturing on selective medium Zygosaccharomyces rouxii Y6 in YPD, incubated on a shaker at 200 rpm at 30^{o}C for 18-24 hours, then take the yeast spread on YPD agar to isolate single colonies, stocks and kept in YPD agar slant

2.3.2 Bring the yeast stored in slant for 2 loops, spread the yeast cultured in YPD, incubated on a shaker at 200 rpm at 30^{o}C for 18 hours, bring the yeast measure the concentration. The turbidity measured with Spectrophotometer absorbance at 660 nm. Absorbance about 0.1 < Volume of cell 1x108 cell/ml>

2.4 Study the growth of yeast Y6 in medium.

The yeast is prepared, cultured in medium YPD supplemented with NaCl 0%, 5%, 10%, 15%, 20%, 25%, 30% and 35% volume of 250 ml. In flask 500 ml. has passed, sterilized at 121 ^{o}C for 15 minutes, then incubated on a shaker with a speed of 200 rpm at 30^{o}C collected every 24 hour and analyzed the growth of the yeast protein.

3. The Analysis

Analyzed the growth of yeast by Analysis the means of cell concentration by turbidity measurements at 660 nanometer Takashi, H., M. Sugishita, Y. Fukushima, T. Fukase and H. Motai. (1991).

Analysis of protein content, analysis by way of Lowry method. Kim, H. R., Kim D.S. and Godber J. S. (1999).

Results

The growths of the yeast Y6 in medium, Yeast extract peptone dextrose added with sodium chloride, 0%, 5%, 10%, 15%, 20%, 25%, 30% and 35% found that medium added sodium chloride, 20%, 25%, 30% and 35% the yeast Y6 cannot growth but medium added sodium chloride, 0%, 5%, 10%, 15% the yeast can growing with the maximum cell concentration of 2.258, 2.399,1.665, and 0.947 at 144, 240,192 and 264 hours, Shown in Figure 2,3,4 and 5. The protein content that yeast created, Yeast has produce proteins differently according to the time change which is equal to 11.590, 10.488,7.326 and 4.167 grams per liter at 144, 240,192 and 264 hours, shown in Figure 6,7,8 and 9. The protein content was created by the yeast is consistent to the concentration of yeast in the same direction, which is the higher concentration of yeast; protein content will increase as well.

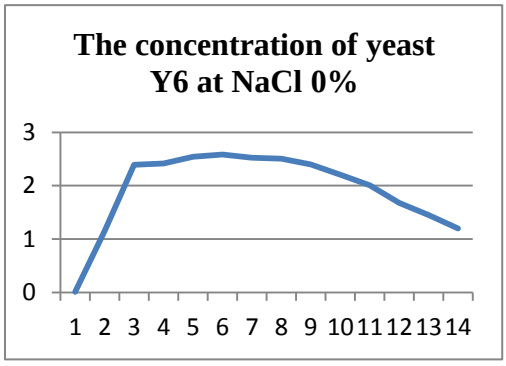


Figure 2 The concentration of yeast Y6 at NaCl 0%

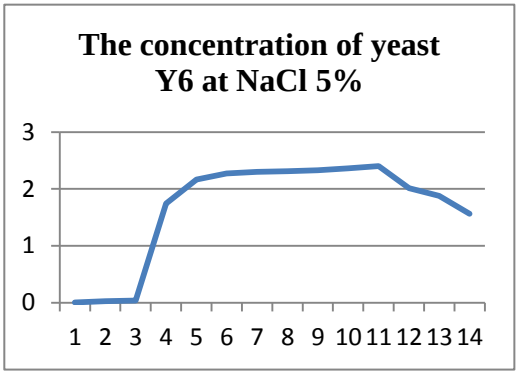


Figure 3 The concentration of yeast Y6 at NaCl 5%

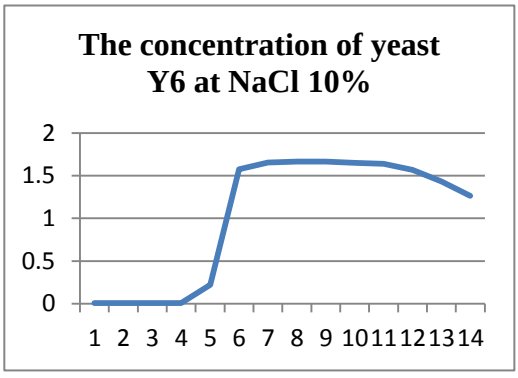


Figure 4 The concentration of yeast Y6 at NaCl 10%

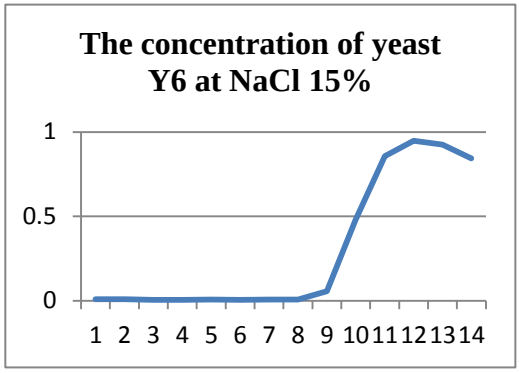


Figure 5 The concentration of yeast Y6 at NaCl 15%

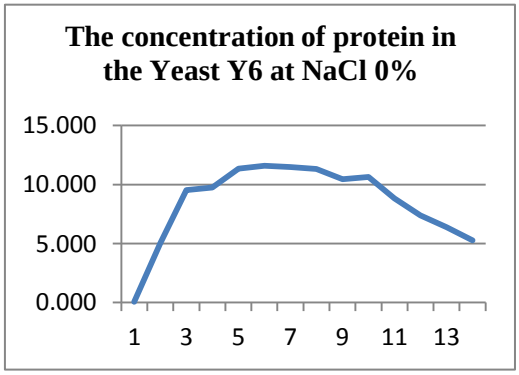


Figure 6 The concentration of protein in the Yeast Y6 at NaCl 0%

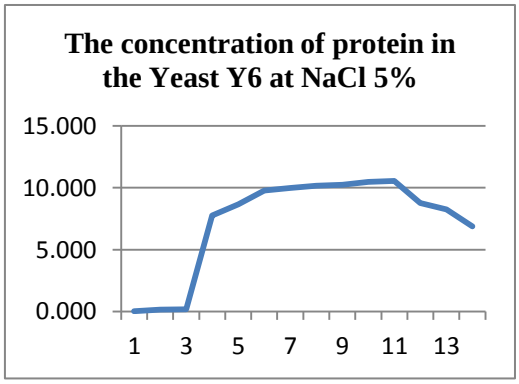


Figure7 The concentration of protein in the Yeast Y6 at NaCl 5%

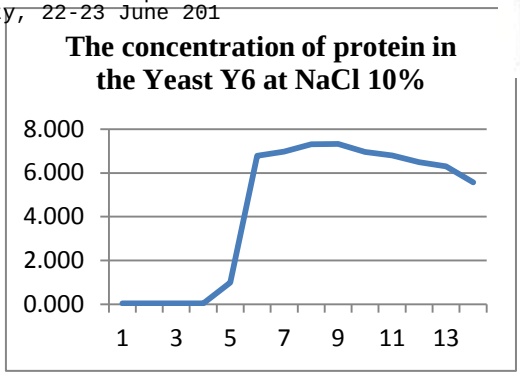


Figure 8 The concentration of protein in the Yeast Y6 at NaCl 10%

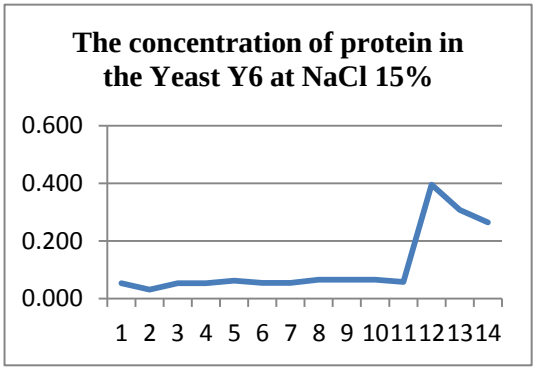


Figure 9 The concentration of protein in the Yeast Y6 at NaCl 15%

Conclusion

From experiment cultured Zygosaccharomyces rouxii Y6 in YPD containing sodium chloride, 0%, 5%, 10%, 15%, 20%, 25%, 30% and 35% found that yeast are able to tolerate in the highest salt at 15% sodium chloride at a cells concentration of 0.947 and protein content equal to 4.167 grams per liter at 288 hour also have highest concentration of cells and concentration of protein without the addition of sodium chloride. By concentration of cells was equal to 2.582 and 11.590 grams of protein per liter, at 144 hour, having the concentration of protein in the same direction as the concentration of yeast. The results obtained if the yeast Zygosaccharomyces rouxii Y6 cultured in waste from industries mixed with huge quantities of sodium chloride can take advantage and develop protein product and pollutants in the environment with salt as well and will increase the option to create protein nutrients for animal and can reduce the cost of farmers to buy protein supplements in another way.

Acknowledgement

Research Study the ability to tolerate salt and produce proteins based on microorganisms isolated from soy sauce residue. It can be successfully with the kind support of the National Research Council of Thailand, which has endorsed the evaluation of research proposals. This research was supported in year's budget by Rajamangala University of Technology Phra Nakhon and we're great to THANK YOU Rajamangala University of Technology Phra Nakhon for providing the opportunity and funding of this research to the research's team. Thank you to all of our research team who are dedicated to working hard and working to resolve all the problems.

References

Alasheh, S. and Duvnjak Z. (1995). Phytase production and decrease of phytic acid content in canola-meal by Aspergillus carbonarius in solid-state fermentation process. Biotechnology progress. 10(4) : 353-359.

Bradford, M. M. (1976). A rapid and sensitive method for the quantitation of micro-gramquantities of protein utilizing the principle of protein-dye binding.Anal. Biochem. 72 : 248-254.

Cromwell, G. L., T. S. Coffey, H. S. Monegue and J. H. Randolph. (1993). Efficacy of phytase in improving the bioavail-ability of phosphorus in soybean meal and corn-soybeanmeal diets for pigs. J. Anim. Sci. 71 : 1831-1840.

Ebune, A., Alasheh, S. and Duvnjak Z. (1995). Production of phytase during solid-statefermentation using Aspergillus ficuum NRRL 3135 in canola-meal. BioresoureTechnology. 53(1) : 7-12.

Kerovuo, J., Lauraeus, M., Nurminen, P., Kalkkinen, N. and Apajalahti J.(1998).Isolation, characterization, molecular gene cloning and sequencing of a novelphytase from acillus subtilis. Appl. Environ. Microbiol. 64(6) : 2079-2085.

Kim, D. H., Oh, B. C., Choi, W. C., Lee, J. K. and Oh T. K. (1999). Enzymatic evaluation of Bacillus amyloliquefaciens phytase as a feed additive. Biotech. Lett.21(11) : 925-927.

Kim, H. R., Kim D.S. and Godber J. S. (1999). Culture conditions for a new phytase producing funus. Biotechnol. Lett. 21 : 1077-1081.

Sharma, Girish Kumar. Potential cost saving for leprosy patients in seeking local leprosy care in Nepal. Bangkok : Chulalongkorn University, 1994. 57 p. (T E8494)

Takashi, H., M. Sugishita, Y. Fukushima, T. Fukase and H. Motai. (1991). Continuous Production of Soy Sauce by a Bioreactor System. Press Biochem. 26 (1991) : 39-45.

Yan ling dong. (2012). Extraction and Purification of Soybean Isoflavone from Soy Sauce Residue south china university of technology

Yoon, S. J., Choi Y. J., Min H. K., Cho K. K., Kin J. W., Lee S. C. and Jung Y.H.(1996). Isolation and identification of phytase – producing bacterium, Enterobacter sp. 4 and enzymatic properties of phytase enzyme. Enzyme Microb. Technol.18 : 449-454.

zhan li. (2009) Fermentation of soy sauce residue protein，foshan ,school of Science and Technology

Innovative Technology and Sustainability Engineering

DEVELOPMENT OF THE EXPERIMENTAL SETUP FOR MEASURING THE SPEED OF SOUND

Sirin Sirathanakul[1, a*]

[1] Division of Science, Faculty of Science and Technology, Rajamangala University of Technology Phra Nakhon, Bangkok, 10800, Thailand

Keywords: experimental setup, measuring the speed of sound, resonance method

Abstract. Sound is a mechanical wave which cannot be seen. It is difficult to measure the speed of sound wave directly. However, when waves of equal amplitude travel in opposing directions, they produce wave resonance. Human ear can hear sound frequencies from approximately 20 to 20,000 hertz. The purpose of this study was to design and develop an experimental setup for measuring the speed of sound in air. In this paper, frequencies of sound from a function generator was used range about 500-1600 hertz, and the sound was detected by human ears and surface of water was used in reflecting the sound. The wavelength of sound in a pipe was controlled with pump and the gravity. The changing of water level changes the effective length of the resonance. The experimental data were statistically analysed by means and percentage error. The experimental setup was found to provide quick, clear and accurate detection of the sound. This finding indicates that the experiment setup for measuring the speed of sound is performed correctly, easily and fast. Furthermore, the percentage of error by comparing between the experimental value and the theoretical value found less than 5.

Introduction

Principle of physics laboratory and Principle of physics are courses available for students in the Computer Science Division and Environmental Science and Natural Resource Division, Faculty of Science and Technology and Food Science and Technology Division, Faculty of Home Economics Technology, at Rajamangala University of Technology Phra Nakhon (RMUTP). Principle of physics laboratory attempts to enhance necessary knowledge for a complete understanding and to develop science process skills in Principle of Physics. Effective and efficient instrument influences greatly on experimental result, reliable and valid measuring [1]. In this study, the experimental setup was designed and developed for measuring the speed of sound by using the resonance method. A sound waves traveling through air are longitudinal wave. In longitudinal waves, the particles of the medium move in a direction parallel to the direction of energy transport

Sound travels through an elastic medium: slowly in gases; faster in liquids than in gases and faster in solids than in liquids. For ideal gas, sound travelling depends only on its temperature, pressure and density have no effect on the speed of sound [2]. Air, a mixture of oxygen and nitrogen, constitutes a non-dispersive medium. Thus, at a fixed pressure, speed of sound is the same for all frequencies [3]. In physics, resonance is a phenomenon in which a vibration system or external force drives another system to oscillate with greater amplitude at specific frequencies [4]. A resonance in air can be set up in a pipe by interference at the same frequency between sound waves traveling in opposite directions; incident wave and the wave reflected from one end closed of the pipe, [5]. A loud sound is produced when resonance occurs.

Applications of basic theories and laws of Physics can be proved by methods or experiments, which may vary between different universities, for some suitable reasons, such as basic knowledge, budgets and etc. In this paper, the speed of sound was measured by resonance method. Source sound from a function generator was used and the sound was detected by human ears**.**

Research Methodology

The process of design and construction the instrument setup for measuring the speed of sound at atmosphere pressure was as follows:

1. To study and collect the data toward the experiment related with measuring the speed of sound on the document and internet resources.

2. To define a clear scope of the research instrument. The scope and the instrument setup in this research were devised into 3 parts as follow:

Part1. Source and frequencies of sound: The sound source in this research used Function Generator, because outgoing of sound provides continuously and can control the intensity of sound. If the source sound is tuning fork, the signal is discontinuous and the intensity of sound can't be controlled. The speaker was connected with Function Generator. The sound experiment frequencies were used in range from 400 to 1600 hertz (Hz).

Part2. Types and sizes of pipes: The pipe in the experiment used acrylic plastic, acrylic has many valuable properties for this choosing discuss from optically transparent forms, abrasion resistant, resistant to ultraviolet and easily clean pipe [6]. The internal diameter of acrylic pipe is suitable the diameter of speaker. The length of the pipe was determined by range of frequencies experiment in resonance method theory. If the frequencies are used lower than 400 Hz, the pipe must be longer. On the other hand, if the frequencies are used more than 1600 Hz, the results of the experimental may be obtained high percentage of error, although this advantages is to make the pipe shorter.

Part3. The air tube of pipe: The instrument set was designed to quickly perform, accurate and low cost. The researcher used the ears for detecting the sound. However, the incoming resonance must be a good quality sound. Wherewith, hard and smooth surfaces are particularly good quality at reflecting sound. The surface of the water is a good reflector. Because of fluid molecules reduce energy by encircling other molecules, and this energy creates a force, surfactant. The length of air tube was adjusted by changing of water level in pipe. Increasing the length of the air tube is achieved by allowing the water in pipe flow through the valve. Thus, valve help to control the flow rate of water. The decreasing of the air tube used the pressure from pump; resist earth's gravitational attraction.

3. Construct and assembling the instrument setup: The pump's power determines the height of water level in pipe. The pipe was cut; the length of pipe must be longer than length of air tube as a resonance and longer than length of the water level from pump's power for preventing the overflow of water. This experiment used power of pump at 20-23 watt (W), determined from a benefit between the both reasons and pumps are sold in the market. Scale line of distance was stickled beside pipe from the top end to down by default value from 0 to 120 centimetre (cm). An aluminium rod was cut by Cutting saw. After that it was put in the Milling Machine and Lathe Machine. As a result of that, aluminium rods is formed a work piece for getting the pipe. Inside of work piece was drilled and put the O-ring for seal, prevent flowing out of the water. Moreover, aluminium work piece was drilled and tapping. The rubber tube connects with aluminium work piece; the other ends connect with a valve. This valve is two way directions. The other end of valve connects with other rubber tube and passes to the pump. Then other components of instrument setup were compiled together.

4. The validity and reliability of the instrument setup; This experiment setup have a detail as follow: set up the range of frequency from Function Generator used during 400 Hz-1600 Hz, length of acrylic pipe used about 130 cm and 3.2 cm, power of pump used 20-23 W, type of valve used two way direction for the water flow in and flow out, power suppy used separate switch socket, the capacity of bucket used at least about 3 litre (L), because the whole of pump sink in water all time in experiment; so capacity of bucket must be more than the volume of pipe. Finally in doing the

experiment, place speaker over the pipe at 0.50 cm. Caution: Don't touch to power supply while your hand wet the water.

For the purposes of improving and editing defects. After the trial found that it is not easy in work. So that researcher modified the length of the pipe. In addition, the power of pump and frequerncies in experiment must be changed. The length of pipe used about 65 cm, power of pump used 8-9 W and frequency used during about 500-1600 Hz, as shown in Fig 1.

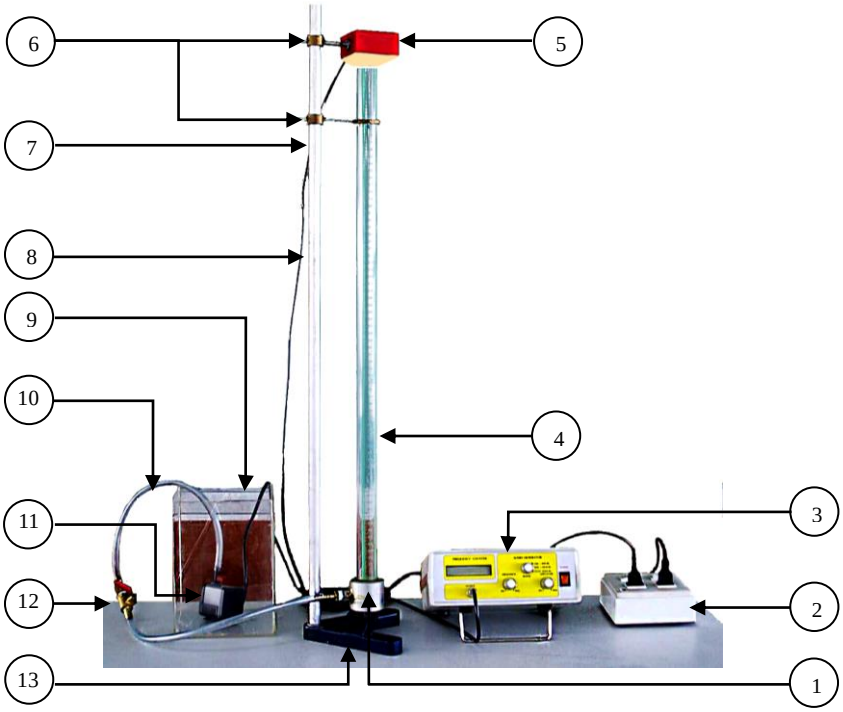


Figure 1 The components of the instrument setup in a measuring the speed of sound respectively are: 1 instead of Aluminum work piece, 2 instead of Power supply, 3 instead of Function Generator, 4 instead of Pipe, 5 instead of Speaker, 6 instead of Clamping devices, 7 instead of Rod of Stand, 8 instead of Electric wire, 9 instead of Bucket for water, 10 instead of Rubber tube, 11 instead of Submersible Pump, 12 instead of Valve, and 13 instead of Base.

The research tools were evaluated by three experts and validated by using the item-objective congruence (IOC) and found that IOC was greater than the standard score as 0.50. The obtained reliability with Cronbach's alpha was 0.85. The experimental setup was tried out by 3 students of Rajamangala University of Technology Phra Nakhon (RMUTP). Then, it was customizable and applied to use in the research.

5. Experimental: The students of RMUTP were selected by sampling, 9 undergraduate. They were divided into 3 groups, equal. Each group was advice relating the experiment by one expert teacher. The details of the experiment were conducted as procedure;

5.1 Fill the water into the bucket, approximately 3 L. The pump must be sinking in water.

5.2 Open the valve and switch on the power supply of pump. As a result, the water will be aspirated into the pipe until almost full. That is the water replaces the air in the pipe.

5.3 Close the valve and switch off the pump.

5.4 Switch on Function Generator and set the frequency. Hence, the sound wave was emitted from a speaker.

5.5 Open the valve in opposite direction from the first. As a result, earth's gravitational attraction will allow the water flow out and pass the rubber tube into the bucket. Size of openings

valve was used to control the rate of water flowing. An adjusting the water level is one method to adjust the length of air tube. This technique can adjust the length of air tube quickly.

5.6 Listen, awareness of the increasing sound and observe the range of position while happen the resonance, loud sound.

5.7 Repeat step 5.2 to 5.6 for adjusting the length of air tube again. In this time, the water was allowed is slowly flow by opening the valve a few. Observe the position that hearing the loud sound.

5.8. Reading directly from the scale lines of distance and record the resonance position into the table of result at the first and second, as the position 1 and position 2, respectively.

5.9 Repeat step of procedure above with another frequency of experiment that it was assigned.

5.10 Reading and record the temperature of air while doing the experiment.

Results and Discussion

The sample group measured the speed of sound for testing 5 frequencies and corrected the data at the position of the resonance 2 position for 1 frequency. The mean of each group results follows in the tables 1. The temperature at the experiment was measured as 25.50°C.

Tables 1 The result of experiment by student of 3 groups;

Frequency f [Hz]	Position 1 X_1 [cm]	Position 2 X_2 [cm]	Length of wave $\lambda=2(X_2-X_1)$ [cm]	Speed of sound $v=\lambda f$ [m/s]
557	14.00	45.10	62.20	346.454
615	12.60	40.70	56.20	345.63
1000	7.20	24.50	34.80	346.00
1250	5.40	19.30	27.80	347.50
1600	3.90	14.70	21.60	345.60
			mean of the speed	346.23

During the position of hearing loud sound is a distance of half a wavelength. Thus, the wavelength is double distance between position 2 and position 1. From the measuring, the speed of sound can calculate from relates to the speed of sound (v), the wavelength (λ) and the frequency of a wave (f), the equation for the speed is $v = \lambda f$. At experiment found the average of the speed of sound in air from experiment value as 346.23 m/s. The theoretical value of physics equations is $v = (\gamma kT/m)^{1/2}$ [7], where γ is the adiabatic index which an idea gas = 1.40, k is Boltzmann's constant = 1.38×10^{-23} J/K, T is the absolute temperature for this experiment at 25.50°C = 25.50+273.15 = 298.65 K and m is the molecular mass of air = 4.8×10^{-26} kg, obtains the value of the speed of sound is 346.71 m/s. The percentage error of the speed of sound was measured by comparing between that two values obtained as 0.13.

The results of measurement the speed of sound had percentage error less. The successful of results may be depend on 3 parts; students, instructor, and instrument setup. The students completed their work with his group and had also been interested in the experiment; carefully carried out to avoid and quiet during did the experiment. The attitudes of the students showed in the willingness to work are affective domain [8]. The availability of instructor by possible knowledge, underlying theory of the experiment, the procedure in experiment had been obtained before starting. This teaching method help the students acquire that in way to observe and carefully carried out to avoid errors. Finally, instrument setup was designed for 3 groups collecting data of the resonance positions for 1 frequency. This average from 3 groups help to obtain more accurate result in

experiment. In many cases above it is reasonable to help removes the barriers of the experiment. Moreover, measuring the speed of sound is accordance with the experiment of Preya Anupongarge in the method and result of experiment but that study adjusted the air tube by moving the rod handle and used oscilloscope for detecting the sound [9]. Thus, this instrument setup can be measured the speed of sound perform correctly, easily and fast and furthermore the percentage of error as 0.13.

Conclusion

From the findings of instrument setup is height effective; the following conclusions were made;

1) In terms of design, this instrument setup was designed for suitability requirements in using, strength, storage and maintenance. Moreover, it is a low cost and also accurate result. So that, the experimental setup was designed the process of production as well.

2) In terms of efficiency, the length of air tube can adjust easily and fast. This indicated that the experimental setup has ability to help effectively learning. The efficient of instrument setup is enough for our purpose of this study.

Recommendation

The measuring the speed of sound has many methods such as echo method, resonance method, etc. Although used the same of method, but the tool was used different type. In this study used the resonance method. This method is suitable for students whom have not background knowledge to use the oscilloscope for detecting the sound and condition of this study is to use mixture gas, low temperature and low humidity. Finally, researcher needs a few experiment tools as possible and to spend less time through experiment. However, this experiment setup is a good tool which it is not only one that gives a result in agreement with other measurements but also it can be used very easy, fast and low cost.

Acknowledgments

In addition, the authors would like to thank the Division of Science; Faculty of Science and Technology, Rajamangala University of Technology Phra Nakhon (RMUTP) for promoting this this study, Division of Tool and Die Engineering of RMUTP for the assistance of constructing the experimental setup and all teachers for offering counselling in many procedures of study.

References

[1] A.A. Ronquillo and O.K. Koc, Laboratory Manual For Physics 211(Engineering), Philippine copying by University of The East, 151 (1999) iii.

[2] The speed of sound is a term used to describe the speed of sound waves passing through an elastic medium on https://www.sciencedaily.com/terms/speed_of_sound.htm, access day on June 10, (2017)

[3] Speed of sound, https://en.wikipedia.org/wiki/Speed_of_sound, access day on May 30, (2017).

[4] Resonance on https://en.wikipedia.org/wiki/Resonance, access day on May 30, (2017).

[5] A.R. Serway, W.J. Jewett, Physics for Scientists and Engineers, sixth ed., Brooks Cole Inc., United States, 1283 (2004) 559.

[6] Acrylic Properties on http://www.emachineshop.com/machine-shop/Acrylic-Properties/page61.html, access day on May 30, (2017)

[7] J.D. Cuttnell, K.W. Jonson, Physics, fifth ed., John Wiley & Son Inc., United States of America, 1002 (2001) 461.

[8] Bloom's Taxonomy: The Affective Domain on http://www.nwlink.com/~donclark/hrd/Bloom/affective_domain.html access day on May 30, (2017)

[9] P. Anupongarge, Development of the Experiment Set for Studying Wave Resonance in Closed End Tube, Journal of Rangsit University : Teaching & Learning , (2011) 19. (in Thai)

AEROSOLIZATION OF HYDROXYL RADICALS AS A SAFE ALTERNATIVE DISINFECTION FOR BACTERIAL REDUCTION

Adjima Chayasitthisophon[1,a], Kenneth W. Foster[2,b], Nopphon Weeranoppanant[3,c] and Aluck Thipayarat[1,4,d*]

[1]Department of Food Engineering, Faculty of Engineering, King Mongkut's University of Technology Thonburi, 126 Pracha-u-Tid Road, Bangmod, Tungkru, Bangkok 10140, Thailand.

[2]Department of Physics, Syracuse University, 225 Physics Building, Syracuse, NY 13244-1130, USA.

[3]Department of Chemical Engineering, Faculty of Engineering, Burapha University, 169 Long-Hard Bangsaen Road, Saensook, Muang, Chon Buri 20131, Thailand.

[4]Office of Education, Faculty of Engineering, Burapha University, 169 Long-Hard Bangsaen Road, Saensook, Muang, Chon Buri 20131, Thailand.

Keywords: disinfecion**,** hydrogen peroxide**,** aerosolization

Abstract. Hydrogen peroxide (H_2O_2) fumigation has recently been explored and tested to be a good fumigant replacement of formaldehyde. This technique has been proven safer, less irritating and requires shorter exposure times. Aerosolization by aerosol generators have been used to produce aerosols containing hydroxyl radicals of hydrogen peroxide. The dispersal of this highly oxidizing mist of micron-size droplets destroyed *Escherichia coli (E. coli)* and *Aspergillus niger (A. niger)* colonies that have been artificially spiked on surfaces. The H_2O_2 mist produced some *E. coli* inactivation graded with concentration. In comparison, *A. niger* was more susceptible to H_2O_2 mist than *E. coli*. However, substantial inactivation of *A. niger* was achieved with 1% H_2O_2 concentration; total annihilation of *A. niger* with a 10-min fumigation with only H_2O_2 aerosolization. At 5% H_2O_2 treatment, the fumigation time was reduced from 8 to less than 2 min. Hence, it is possible to use a minimal H_2O_2 concentration making residual chemical least toxic due to rapid disappearance of the highly unstable free radicals.

Introduction

High bacterial contamination on any food processing areas can signify poor sanitation practices and generate detrimental consequences on human health. Possibly a few of these microbes may act as human pathogens or instigate allergic reactions [1,2,3,4]. Poor hygiene and improper food preparation practices have previously been demonstrated as contributing to many foodborne diseases and outbreaks [5]. At the presence of high microbial counts, cross-contamination allows the transfer of microorganisms (bacteria, virus, parasites, or fungi) from one contaminated area to the other. Unfortunately, people in the food supply chain occasionally have little awareness of surfaces and equipment with high microbial contamination, which are unable to detect by simple visual inspection and take several hours to days to validate [5,6,7]. Unsanitized processes or areas post high risk to transmission and the occurrence of sporadic foodborne outbreaks. Thorough sanitizing of food processing surfaces and areas serves as an

effective precaution to prevent the chance of cross-contamination and eliminate the risk for humans to ingest contaminated food and become ill. The highly effective antimicrobial activity but minimal toxicity of the residual chemical (e.g., organic acids, chlorine dioxide, hydrogen peroxide, and ozonated water) have been done to find alternative aqueous sanitizers [8,9,10]. Each of these chemicals has its own unique characteristics and drawbacks. Hydrogen peroxide (H_2O_2) solution, for instance, has been proven effective in controlling the bacterial contamination. Its toxicity is due to its capacity as an intermediate in oxygen reduction to generate more reactive oxygen species such as the hydroxyl radical. As a highly active biocide, H_2O_2 exhibits antimicrobial activity through the generation of hydroxyl free radicals that penetrate the cell wall to attack lipids, proteins and DNA [11]. Owing to its non-selective biocidal property, it is active against viruses, spores and fungi as well as bacteria [12]. H_2O_2 rapidly degrades into oxygen and water (nontoxic products) upon contacting organic material to form toxic residues, thus having no long-term residual activity. [13]. H_2O_2 is classified as generally regarded as safe for use in food products as a bleaching agent, oxidizing and reducing agent and antimicrobial agent [14]. The use of H_2O_2 (aqueous solution with 0.1–10% concentration) to sanitize whole and fresh-cut produce has been investigated in recent years for example alfalfa sprouts, apples, cantaloupes , honeydew melons, asparagus spears, green peppers, cucumber, zucchini and mushrooms [14,15,16]. Variable effectiveness of H_2O_2 application on raw fruits and vegetables has been reported by many researchers. So the disinfection potential of H_2O_2 is of paramount interest for food industry since this technology can effectively treat harmful microorganisms with minimally effects on sensorial quality of food products and spontaneously decompose leaving practically no trace of toxic byproducts. The knowledge of these hydroxyl radicals enhancement to improve the efficacy of H_2O_2 fumigation was limited. It is depending on the concentration, pH, temperature and other environmental factors [17]. The influence of these factors on H_2O_2 action may be one of the reasons for the variability mentioned above. However, quantitative data about the effect of exposure time, sanitizer concentration and environmental factors on the surviving number of microorganisms are lacking. More systematic research must be done to determine the optimal conditions for the application of H_2O_2 treatments [18].

In this paper, an aerosolize making use of aerosol generator was utilized to create and disperse a disinfectant aerosol and carry hydroxyl radicals with the water mist in micron size to disinfect food processing surfaces as well as difficult-to-reach areas, especially overhead surfaces, cracks and crevasses of food equipment and so on. The aimed of this research was to demonstrate the effectiveness of highly oxidizing mist to eradicate artificially spiked *E.coli* and *A. niger* on surfaces. [19,20,21]. The use of fumigation to lower the toxicity of residual H_2O_2 can facilitate the application of highly efficient disinfecting protocol for food processing area and surfaces with minimal toxicity of chemical residue.

Methods

Bacterial strains and frozen stocks

E. coli DMST 4609 and *A. niger* cultures were obtained from either the Department of Medical Sciences Thailand (DMST, Bangkok, Thailand) or Thailand Institute of Scientific and Technological Research (TISTR, Bangkok, Thailand). All pure cultures were multiplied on tryptic soy agar (TSA, Lab M, UK) to obtain pure single colonies all of a particular strain. One loopful of *E. coli* was transferred into 100 ml of tryptic soy broth (TSB, Lab M, UK) and *A.*

niger also was transferred in potato dextrose broth (PDB, Lab M, UK) in a 250-ml Erlenmeyer flask (Corning™ PYREX™, Corning, NY) and incubated under a isothermic condition at 37 ± 1 °C for 24 h. Before freezing, the inoculum was mixed well with glycerol to achieve 10 – 15% (v/v) final concentration of glycerol. The 1 ml aliquots of inoculum stock in 1.5 ml sterile Eppendorf tubes were kept at -20°C.

Preparation of lysate of *E. coli* and *A. niger* strains

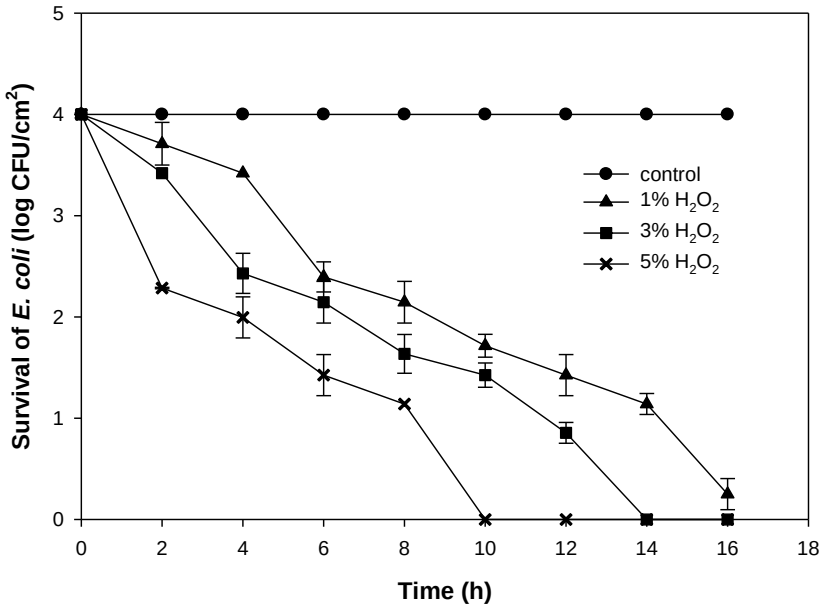
E. coli DMST 4609 and *A. niger* cultures from frozen stock was enriched in 100 ml of TSB and PDB in a 250-ml Erlenmeyer flask incubated at 37±1 °C, 200 rpm for 18 – 24 h. The 10-fold serial dilutions were done in 0.1% w/v peptone water (PW, Difco, Laboratories, Sparks, MD) to obtain cell density around 10^1 - 10^4 log CFU/ml. A 20 µl aliquot of each diluent (*E. coli* and *A. niger)* was spread directly on TSA and PDA plates respectively. All agar plates from both strains were incubated at 37±1 °C for 18 – 24 h. The uniformly-distributed colony of proper dilution had to be selected to determine the efficacy of hydroxyl radical fumigation generated in the experiments.

Aerosolization by Hydrogen peroxide

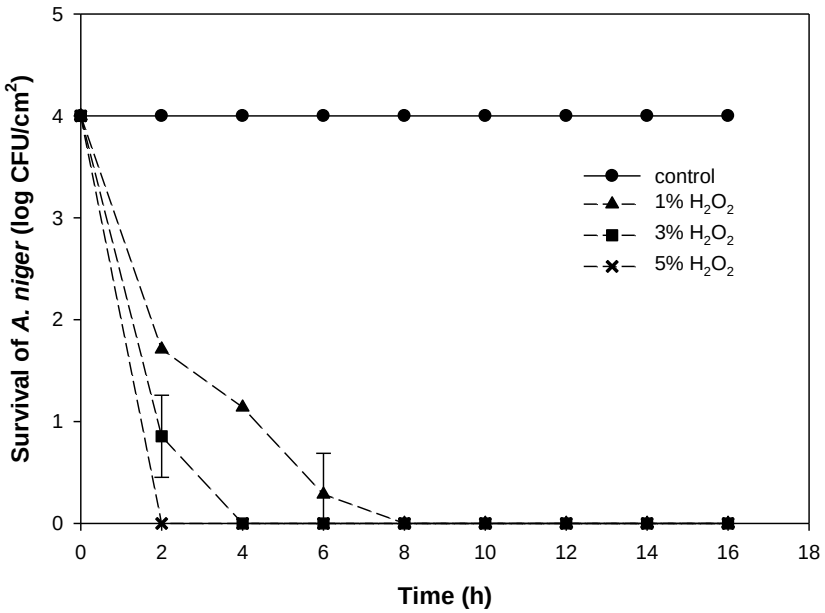
The generation of OH• was achieved using a patent-pending technology Thailand-1701000719, initiated by the mixture between H_2O_2 and DI water is into a reservoir with 10 liter working volume. In the reservoir, there are 3 aerosol generators with water evaporation of 133 ml per hour to make H_2O_2 aerosols. And there is also a fan attached to the top of the reservoir to disperse the vapor. The aerosols distribution inside the chamber with the dimension 34x34x34 cm^3. The excess aerosols escaped the chamber through the outlet opening.

Results and Discussion

The bactericidal property of liquid H_2O_2 has been well documented to increase with its higher concentration. The application of H_2O_2 fumigation, especially in vaporized hydrogen peroxide (VHP) fumigation using high H_2O_2 concentration (more than 35%), has shown to be effective in microbial surface decontamination [22]. In this work, an alternative concept of H_2O_2 fumigation (i.e., ultrasonic fumigation using low concentration of H_2O_2) was demonstrated to inactivate microbial contamination on spiked surfaces using two models of microorganisms including bacteria (*E. coli*) and mold (*A. niger*). Micron-size H_2O_2 droplets generated by ultrasonic transducers were launched by a forced air mechanism and dispersed well in the testing chamber. The bacteria and molds were inoculated on the top surface of the chamber at 4 log CFU/cm^2 and Fig. 1 (a and b) showed the different degrees of inactivation comparing the effectiveness of this H_2O_2 fumigation on bacterial and mold samples using different concentrations of H_2O_2 fume (i.e., 1, 3 and 5% H_2O_2). Although the use of H_2O_2 at high concentration via VHP process has been shown to be effective against a wide array of microbial contamination (e.g., bacteria, yeast, fungi, viruses, bacteria spores, etc.) [23,24], the antimicrobial effectiveness with the H_2O_2 vapor phase at lower concentrations has not been fully understood on the killing activity of selected microorganisms, especially using ultrasonic frequency in creating minuscule aerosols [25,26].



a) Inactivation of *E. coli* treated with different concentration of H_2O_2



b) Inactivation of *A. niger* treated with different concentration of H_2O_2

Figure 1 Normalized inactivation of a) *E.coli* and b) *A. niger* (the initial contamination at 4 log CFU/cm^2) on surfaces of a test chamber (34x34x34 cm^3) after H_2O_2 fumigation treatment at various of H_2O_2 concentrations (1, 3 and 5 percent).

The ultrasonic sound waves was able to energize H_2O_2 solution and successfully create stream of H_2O_2 aerosols producing substantial microbial reduction within 15 minutes. It was evident that higher H_2O_2 concentration resulted in faster and greater microbial inactivation [18]. Increasing the antimicrobial agent concentration generally improved the effectiveness of the treatment. With the 4 log CFU/cm^2 of the initial *E. coli*/coliform contamination, only 5% H_2O_2 fume enabled total disinfection of the spiked surfaces within 12 min whereas the lower concentrations (i.e., 1% and 3% H_2O_2) were unable to produce complete sterility (Figure 1a). For mold, the H_2O_2 fume as low as 1% H_2O_2 was able to eliminate artificial *A. niger* contamination within 10 min where the same H_2O_2 fume only produced 20-25% inactivation of *E. coli*/coliform on the normalized scale (Figure 1b). Higher concentration of H_2O_2 fume (i.e., 5% H_2O_2) shortened the treatment time to 4 min. From the results of *A. niger* survivability (Fig 2), this H_2O_2 fumigation was more effective towards mold inactivation than the bacteria Seemingly, mold was more susceptible to this low-concentration H_2O_2 fumigation than bacteria. Similar results has been reported in the commercial VHP systems that this technology is highly effective to rapidly oxidize fungal vegetative forms and spores; although, bacterial spores are more resistant than fungal spores.

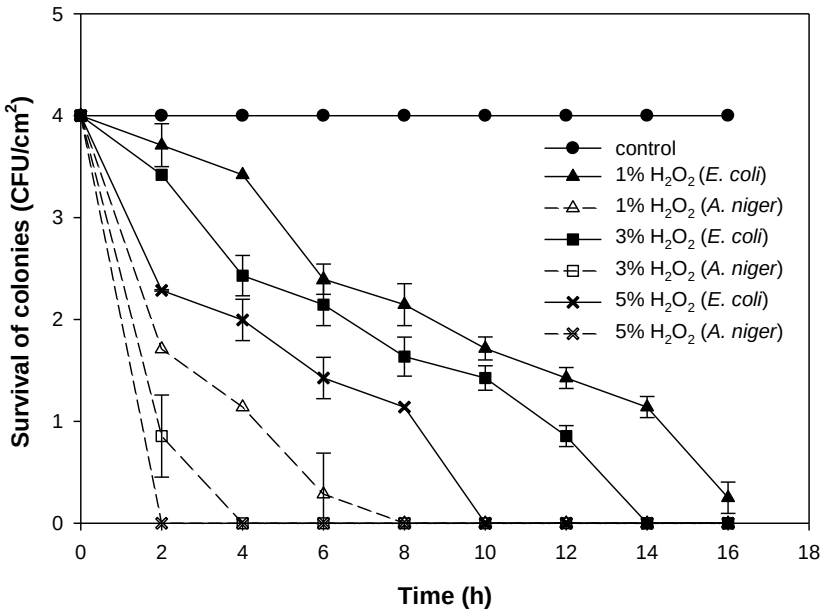


Figure 2 Comparison the efficiency of the H_2O_2 fumigation treatment on the survival of *E. coli* and *A. niger* at various H_2O_2 concentrations (1, 3 and 5 percent).

In this experiment, bacterial and mold inactivation effects were observed in the H_2O_2 vapor phase similar to that happening in its liquid phase. [26] summarized the research works demonstrating the same effectiveness of H_2O_2 in vapor phase in sterilizing against different microorganisms as follows: vegetative bacteria and highly resistant bacteria endospores [12,24,25,27,28,29,30,31,32,33], viruses [23], fungi [34], yeast, amoebae, infective proteins and other microorganisms [30,35,36]. Similar to what observed in the commercial VHP systems, it is assumed that the inactivation activity of H_2O_2 follows the same oxidation mechanism. When

the H_2O_2 fume comes into contact with microorganisms, H_2O_2 produces destructive hydroxyl free radicals oxidizing membrane lipids, DNA and other essential cell components. The concentrations of H_2O_2 and free radical derivatives must overwhelm the natural cell defense mechanism (e.g., indigenous catalase) so that the oxidation fails cell's regulatory functions.

Summary

The antimicrobial action of H_2O_2 was investigated in aqueous systems as a basis for understanding the only effects of H_2O_2 concentration levels without considering other factors involved in sanitization such as produce surface characteristics, microorganism attachment features and presence of organic material other than the bacteria. Effectiveness of H_2O_2 solutions against *E. coli* and *A. niger* appeared to improve at the agar surface by H_2O_2 aerosolization. The effects of H_2O_2 concentration on the exposure time needed to reduce the microbial population by 4 log CFU/cm^2. The results present could help understanding the effect of H_2O_2 aerosolization level on *E. coli* and mold inactivation in aqueous solutions. The hydroxyl radicals availability was hypothesized to provided strong oxidation potency of this successful H_2O_2 treatment scheme. Further similar studies using different *E. coli* strains, biofilms and fresh produce are in progress in order to evaluate the effect of vegetable surface characteristics and optimize H_2O_2 treatments avoiding or minimizing adverse effects on the quality of fresh produce. The antimicrobial action of H_2O_2 have been successfully applied in many environmental treatment applications for the sanitation of microbial contamination in processing line and other hazardous toxic organic pollutants, either to less harmful compounds or to their complete mineralization.

Acknowledgements

This work was financially supported by the research grant of Burapha University through National Research Council of Thailand (Grant No. 115/2560). We are grateful to have strong industrial partnership with Thai Asia Rice product Co., Ltd. Special thanks are extended to the Thailand Research Fund, namely Research and researcher for industry: MAG master research Grants (Grant No. MSD58I0110).

References

[1] T. Husman, Health effects of indoor-air microorganisms. Scand. J. Work Environ. Health. (1996) 5-13.

[2] J. C. Kash, J. K. Taubenberger, The role of viral, host, and secondary bacterial factors in influenza pathogenesis. The American journal of pathology, 185 (2015) 1528-1536.

[3] R. Kaushik, R. Balasubramanian, Discrimination of viable from non-viable Gram-negative bacterial pathogens in airborne particles using propidium monoazide-assisted qPCR. Sci. Total. Environ. 449 (2013) 237-243.

[4] X. Wu, C. Han, J. Chen, Y. W. Huang, Y. Zhao, Rapid detection of pathogenic bacteria from fresh produce by filtration and surface-enhanced raman spectroscopy. Jom. J. Min. Met. Mat. S. 68 (2016) 1156-1162.

[5] M. C. Erickson, J. Liao, J. L. Cannon, Y. R. Ortega, Contamination of knives and graters by bacterial foodborne pathogens during slicing and grating of produce. Food. Microbiol. 52 (2015) 138-145.

[6] S. F. Bloomfield, Home hygiene: a risk approach. Int. J. Hyg. Envir. Heal. 206 (2003) 1-8.

[7] F. Pérez-Rodríguez, A. Valero, E. Carrasco, R. M. García, G. Zurera, Understanding and modelling bacterial transfer to foods: a review. Trends. Food. Sci. Tech. 19 (2008) 131-144.

[8] L. A. Keskinen, A. Burke, B. A. Annous, Efficacy of chlorine, acidic electrolyzed water and aqueous chlorine dioxide solutions to decontaminate Escherichia coli O157: H7 from lettuce leaves. Int. J. Food. Microbiol. 132 (2009)134-140.

[9] Y. Huang, H. Chen, Effect of organic acids, hydrogen peroxide and mild heat on inactivation of *Escherichia coli* O157: H7 on baby spinach. Food Control. 22 (2011) 1178-1183.

[10] D. H. Kingsley, E. M. Vincent, G. K. Meade, C. L. Watson, X. Fan, Inactivation of human norovirus using chemical sanitizers. Int. J. Food. Microbiol. 171 (2014) 94-99.

[11] G. McDonnell, A. D. Russell, Antiseptics and disinfectants: activity, action, and resistance. Clin. Microbiol. Rev. 12 (1999) 147-179.

[12] S. S. Block, Peroxygen compounds. In: Disinfection, sterilisation and preservation. Forth ed., Philadelphia, New York, 1991

[13] J. Herdt, H. Feng, Aqueous antimicrobial treatments to improve fresh and fresh-cut produce safety. Microbial safety of fresh produce. A John Wiley & Sons Publication Inc., Iowa, 2009, pp. 169-190.

[14] G. M. Sapers, G. F. Simmons, Hydrogen peroxide disinfection of minimally processed fruits and vegetables. Food Technol. 52 (1998) 48-52.

[15] R. Montville, Y. Chen, Schaffner, D. W. Glove barriers to bacterial cross-contamination between hands to food. J. Food. Protect. 64 (2001) 845-849.

[16] D. O. Ukuku, G. M. Sapers, Effect of sanitizer treatments on Salmonella Stanley attached to the surface of cantaloupe and cell transfer to fresh-cut tissues during cutting practices. J. Food. Protect. 64 (2001) 1286-1291.

[17] S. Beuch, B. Boelcke, L. Belau, Effect of the organic residues of Miscanthus× giganteus on the soil organic matter level of arable soils. J. Agron. Crop. Sci. 184 (2000) 111-120.

[18] S. Raffellini, S. Guerrero, S. Alzamora, Effect of hydrogen peroxide concentration and pH on inactivation kinetics of Escherichia coli. J. Food. Safety. 28 (2008) 514-533.

[19] O. Dvir, I. Farber, M. Rom, A. Rachman, D. Beno-Moualem, S. Meir, A. Lichter, Low humidity after harvest changes the abscission site in bunch cherry tomatoes. J. Sci. Food. Agr. 89 (2009) 1519-1525.

[20] O. A. Karabulut, K. Ilhan, U. Arslan, C. Vardar, Evaluation of the use of chlorine dioxide by fogging for decreasing postharvest decay of fig. Postharvest. Biol. Tec. 52 (2009) 313-315.

[21] A. Daus, B. Horev, O. Dvir, S. Ish-Shalom, A. Lichter, The efficacy of ultrasonic fumigation for disinfestation of storage facilities against postharvest pathogens. Postharvest. Biol. Tec. 62 (2011) 310-313.

[22] T. Kimura, Effective Decontamination of Laboratory Animal Rooms with Vapour-phase (â€ œVaporizedâ€) Hydrogen Peroxide and Peracetic Acid. Scand. J. Lab. Anim. Sci. 39 (2012) 17-23.

[23] R. A. Heckert, M. Best, L. T. Jordan, G. C. Dulac, D. L. Eddington, W. G. Sterritt, Efficacy of vaporized hydrogen peroxide against exotic animal viruses. Appl. Environ. Microb. 63 (1997) 3916-3918.

[24] A. Kahnert, P., Seiler, M. Stein, B. Aze, G. McDonnell, S. H. Kaufmann, Decontamination with vaporized hydrogen peroxide is effective against Mycobacterium tuberculosis. Lett. Appl. Microbiol. 40 (2005) 448-452.

[25] B. Unger-Bimczok, V. Kottke, C. Hertel, J. Rauschnabel, The influence of humidity, hydrogen peroxide concentration, and condensation on the inactivation of Geobacillus stearothermophilus spores with hydrogen peroxide vapor. J. Pharm. Innov. 3 (2008) 123-133.

[26] P. Kačer, J. Švrček, K. Syslová, J. Václavík, D. Pavlík, J. Červený, M. Kuzma, Vapour phase hydrogen peroxide-method for decontamination of surfaces and working areas from organic pollutants. In: Organic Pollutants Ten Years after the Stockholm Convention-Environmental and Analytical Update., Croatia, 2012, pp. 400-30.

[27] G. L. French, J. A. Otter, K. P. Shannon, N. M. T. Adams, D. Watling, M. J. Parks, Tackling contamination of the hospital environment by methicillin-resistant Staphylococcus aureus (MRSA): a comparison between conventional terminal cleaning and hydrogen peroxide vapour decontamination. J. Hosp. Infect. 57 (2004) 31-37.

[28] L. Hall, J. A. Otter, J. Chewins, N. L. Wengenack, Use of hydrogen peroxide vapor for deactivation of Mycobacterium tuberculosis in a biological safety cabinet and a room. J. Clin. Microbiol. 45 (2007) 810-815.

[29] M. D. Johnston, S. Lawson, J. A. Otter, Evaluation of hydrogen peroxide vapour as a method for the decontamination of surfaces contaminated with Clostridium botulinum spores. J. Microbiol. Meth. 60 (2005) 403-411.

[30] N. A. Klapes, D. O. N. A. L. D. Vesley, Vapor-phase hydrogen peroxide as a surface decontaminant and sterilant. Appl. Environ. Microb. 56 (1990) 503-506.

[31] M. Kokubo, T. Inoue, J. Akers, Resistance of common environmental spores of the genus Bacillus to vapor hydrogen peroxide. *PDA* J. Pharm. Sci. Technol. 52 (1998) 228.

[32] J. V. Rogers, C. L. K. Sabourin, Y. W. Choi, W. R. Richter, D. C. Rudnicki, K. B. Riggs, M. L. Taylor, J. Chang, Decontamination assessment of Bacillus anthracis, Bacillus subtilis, and Geobacillus stearothermophilus spores on indoor surfaces using a hydrogen peroxide gas generator. J. Appl. Microbiol. 99 (2005) 739-748.

[33] G. M. Sapers, P. N. Walker, J. E. Sites, B. A. Annous, D. R. Eblen, Vapor-phase Decontamination of Apples Inoculated with *Escherichia coli*. J. Food. Sci. 68 (2003) 1003-1007.

[34] C. F. Forney, R. E. Rij, R. Denis-Arrue, J. L. Smilanick, Vapor phase hydrogen peroxide inhibits postharvest decay of table grapes. HortScience. 26 (1991) 1512-1514.

[35] G. Fichet, E. Comoy, C. Duval, K. Antloga, C. Dehen, A. Charbonnier, G. McDonnell, P. Brown, C. I. Lasmézas, J. P. Deslys, Novel methods for disinfection of prion-contaminated medical devices. The Lancet. 364 (2004) 521-526.

[36] S. Vassal, L. Favennec, J. J. Ballet, P. Brasseur, Hydrogen peroxide gas plasma sterilization is effective against Cryptosporidium parvum oocysts. Am. J. Infect. Control. 26 (1998) 136-138.

Evaluated investment projects using ERP (Enterprise Resource Planning). A case study of industrial plants By Real option.

Kunyaluk Nuchprayoon[1*] , Chayathach Phuaksaman[2]

[1]*Department of Industrial Engineering, Faculty of Industrial Engineering, King Mongkut's University of Technology North Bangkok, Bangkok 10800, Thailand*

[2]*Department of Industrial Engineering, Faculty of Industrial Engineering, King Mongkut's University of Technology North Bangkok, Bangkok 10800, Thailand*

Abstract

This research project aims to evaluate investment decisions using ERP (Enterprise Resource Planning) and to create a model to analyze the ratio of return on investment for ERP systems and assess the value of the return on investment. To offer alternatives to invest ERP system by means of research, by means of a computer model, discounted cash flow (Discounted Cash Flow Model), the financial indicators (NPV) test and simulation scenarios. Monte Carlo by calculation of the change of key variables simultaneously using the sample data, which varies according to the nature of the distribution of those variables. For results of the distribution of financial returns that provide a basis for decision making. The research found that Factors that are sensitive to investment returns, most of the modules due to high net worth and changes over time, which is related to the rate of change of the module and investment, and to assess the level of risk based on simulations. Monte Carlo The investment using ERP (Enterprise Resource Planning) module, the FA has only a few fragments. The simulations showed that the chance that the NPV is less than zero, so 17.86 million baht investment alternative than investing in alternatives.

Keywords: Real Option, ERP, Monti Carlo

1. Introduction

In today's business climate Information technology has to play a major role. To change the behavior of enterprises in the management of the organization. The new business is created and controlled by the computer. Is the primary tool linking information between agencies Internal and external. The ability to control communication. The process is to decide properly and ready to face the competition in digital.

For the industry, ERP (Enterprise Resource Planning). It is a system that is gaining popularity. It is a computer system to control processes in all departments of the organization and linking data from all related departments into a central database. The data operations are coordinated, consistent, unified company.

Today it has expanded the definition of an Extended ERP or ERP to ERP Plus. This means covering the rest of the system. A CRM system, which means the management resources of both organizations before the sale. Such as aggressive sales (Sales Force Automation) aftermarket. And customer support system (Customer Support) system ERP Plus. Used to control the various business processes (Business Processes). Almost every organization within the organization, whether they are single or multiple transactions. Related to the manufacturing, distribution and finance. The system supports the decision of the executive (Executive Information System).

Although systems, ERP (Enterprise Resource Planning) system that enables organizations to become more efficient. But there are many organizations that do not invest with ERP. Due to many factors such as the relatively high price. There is functionality in several modules. The value of investments and so on from the above research, this study aims to evaluate investment decisions using ERP (Enterprise Resource Planning). And modeling to analyze the ratio of return on investment using ERP (Enterprise Resource Planning). And assess the value of the return on investment. To offer an alternative to investing using ERP (Enterprise Resource Planning) guidance to operators as an alternative to investing using ERP (Enterprise Resource Planning).

2. Objectives

1. To study the project investment decisions using ERP (Enterprise Resource Planning).
2. To create a model to analyze the ratio of return on investment using ERP (Enterprise Resource Planning) and assess the value of the return on investment. To offer an alternative to investing using ERP (Enterprise Resource Planning).

3. Benefits

1. The guidelines for the valuation of investments using the ERP (Enterprise Resource Planning).
2. Data on the proportion of the return on investment using ERP (Enterprise Resource Planning) and assess the value of the return on investment. To offer an alternative to investing using ERP (Enterprise Resource Planning).

4. Scope of Research

1. Learn how to evaluate the form factor of choice for investors using ERP (Enterprise Resource Planning).
2. Modeling the valuation to determine the value of the investment system, ERP (Enterprise Resource Planning).

5. Literature Review

The concept of enterprise resource management system

Organizations or companies that engage in business activities, delivering products or services to customers. The business is to create value for the business resources to be used. Or services delivered value is divided into sections or departments. Each division will be responsible for their part. And the final value of coordination between the individual parts together. The coordination is called Value Chain (Value Chain) (Ban, 2546: 3).

When the chain link together often a problem of waste. And inefficiency, a problem that often occurs. The difficulty in recognizing the work of the various departments immediately. Make investment decisions and manage resources. The organization is made more difficult Difficulty in managing such as

1. Expanding the scope of activities linked. When an organization grows larger departments. Related to the value adds up. Activities linked to longer.

2. The linking structure of more complex activities. When an organization grows larger, the division of labor associated activities. More complicated

3. The loss of activity. And the rapid decline in performance. When the connection of events. Larger and more complex. Causing the front wall The loss of activity The relationship between horizontal activity will slow down. Efficiency in all links below.

4. Recognition of the link activity difficult. When linked to a larger and more complex. The perceived condition or results of activities in various divisions more difficult to export data to management immediately recognize.

5. investment and management resources to maximize difficult. When management cannot decide It will contribute to the management and implementation of activities. According to various problems in the organization.

Enterprise Resource Planning systems (Enterprise Resource Planning) system to help manage the unified organization. By controlling the operation and management of resources in the organization, coordination. Accounting, HR, finance, manufacturing, warehousing, customer tax automatically. The computer systems to process control in every way. Corporate entities and associated data from all related departments into a central database. The data operations are coordinated, consistent, unified whole company (Dejpongsaem: 2004). Such a system would include aspects of business operations. In the form of a single database. But look model works like a real business. The entire system can be connected to each other realtime (Ban, 2546: 7). In other words, Enterprise resource planning system The system is used to manage and plan the use of resources of the organization by systematically linking systems. Enterprise together The system of accounting and finance. HR System Production management system The distribution system To help plan and manage enterprise resources efficiently. It also reduces the time and process them.

Factors to consider when deciding to invest in ERP software are as follows.

1. The use of software or software development itself.
2. Technology and architectural design of the ERP.
3. Functions of ERP will need to respond to business and build a successful organization.
4. Manually editing software (Customization)
5. Maintenance software (Software maintenance).
6. Cost of ownership ERP (Cost of Ownership).

Real Option method.

Typically the rate of return projects. Often used traditional financial metrics such as NPV or IRR. Alternatively, use metrics such as ROI assembled in the accounting decisions. But the use of these indicators alone. Cannot respond to changes Ditto This is because IT projects Is associated with the uncertainty in many forms. Uncertainty in terms of the process of project management. Uncertainty in terms of the technology itself. Or the prospect of future changes and so on. To calculate the return on investment traditionally has always been critical in terms of the ability to predict these uncertainties.

In practice, the decision to invest in IT projects the organization is "Options" in the judgment in many ways, for example. May cancel the project failed to fewer losses. Or may delay investment decisions in the future until the time was right. In some cases, organizations have the need to invest in a project with a negative NPV compensation if the project could cause the project to create business opportunities for many future projects.

Analysis of return Projects with Real Options Analysis, it is seen to be an alternative. The analysis yields a more close. The uncertainty reflects the nature of IT projects. And now, many organizations have begun to use Real Options Analysis to evaluate the return on IT investments already. Although some may face difficulties in implementing them. But the trend in the future, we should see the results of research on the increase. Enabling a better understanding of the accuracy even more.

Simulation Monte Carlo

Monte carlo analysis is a technical analysis of the Monte Carlo. As a way to emphasize the statistical research. The relationship with the possibilities of what can be. Happened to prioritize them.

Step implementation of the Monte Carlo building up of random variables. With distribution The probability that a given process modeling, analysis and solution of the problem by the Monte Carlo method to generate random events. Replication using Program packet Using simulation techniques In a statistical study

Monte carlo is a technique used to help simulation business problems using a random number table or a computer to help to generate random number method.

1. distributions result (outcome) to the experimental one.
2. Define the probability of impact it will have in the first.
3. Determination of the number represents the probability of each outcome is in line with the second.
4. Use a random number table or computer to generate random number in each iteration.
5. The figures random number to determine the third. During that match any And indicate any effect under Article 1.
6. Go back to four in order to iterate the next time.
7. At the end of the stipulated sum up and interpret.

Technical analysis of the Monte Carlo help in the decision making process as such.

1. To help determine the framework for decision making. For planning
2. The decision to seek help to prevent the problem. Expected to happen before the problem becomes critical.
3. enables the assessment of alternatives. Policy and Practice
4. Add the chance to choose from many options that it offers.

6. Tools used in research

1. models, discounted cash flow (Discounted Cash Flow Model).
2. Test Simulation Monte Carlo.

7. Methodology

1. The decision to invest in the education system, ERP (Enterprise Resource Planning) and other factors. Affecting Investment Systems ERP (Enterprise Resource Planning).

2. Determination of the return on investment of these five choices are given.

Option 1 choice for ERP (Enterprise Resource Planning) vendors every module.

Option 2 uses ERP (Enterprise Resource Planning) module only FA and MM.

Option 3 uses ERP (Enterprise Resource Planning) module only FA.

Option 4 is developed by the internal.

Option 5 Current conditions

3. Use assessed using discounted cash flow models (Discounted Cash Flow Model) financial indicators (NPV), which is used in this research is the weighted average cost of WACC is based on case studies calculate.

The decision of the (NPV)

If NPV is greater than zero should yield investment program because of the added value over the cost of the investment project.

If the NPV is less than or equal to zero. Should not invest because there is no added value from the investment.

It also uses the internal rate of return, IRR (Interal Rate of Return), and compared between the two indicators of the five cases.

4. Determination of the sensitivity of the various input variables. The impact on the financial return of the project. To assess whether The uncertainty of the variables that affect the risk of, or changes very little financial return of the project, respectively.

5. Test Simulation Monte Carlo by calculating the effect of changes in key variables simultaneously using the sample data, which varies according to the nature of the distribution of those variables. For results of the distribution of financial returns that provide a basis for decision making.

8. Conclusion

1. Analysis of changes.

From analyzing the ROI of the investment project. Keeping in mind the factors that affect the investments of investors using ERP (Enterprise Resource Planning) by Maria Ortega option. The study has several industrial enterprises, affecting the decision. And many factors are difficult to control because the investor is unable to determine the various factors which have such an effect on the analysis of investment projects. Analysis of changes in factors It has changed the factors that increase and decrease. To study the dynamics of the financial criteria.

Table 1 changes the value of the NPV.

factor	The changes
Investments	-10%
Inflation	-5%
The rate of increase in the FT	0%
Module number	-20%
The rate of change of module.	-15%

From Table 1, factor analysis to find factors that are sensitive to investment returns, most of the modules due to high net worth and changes over time, which is related to the rate of change of the module and investments. the investment in the ERP system to factor in the number of modules.

2. The results of Monte Carlo simulations.

Simulation analysis of events under uncertainty. How is that different from the sensitivity analyzes. And let's analyze the bigger picture analysis Scenario modeling to project and monitor the results to determine the feasibility of different variables. The changes are simultaneously the Monte Carlo simulation can evaluate the level of risk in each situation.

Table 2 Assessment of the level of risk.

Sizing investment	Investments (CAPEX) million baht.	Return (NPV) million baht.	Internal Rate of Return (IRR)%.	Standard Division (S.D).	The NPV lowest potential (million baht).	No. risks of investing very little.
The ERP (Enterprise Resource Planning) vendors every module.	20.00	26.00	7.45	3.86	-12.23	2
The ERP (Enterprise Resource Planning) module only FA and MM.	16.00	22.00	6.56	3.00	-16.30	3
The ERP (Enterprise Resource Planning) module only FA.	13.00	17.00	4.78	2.33	-17.86	4
Within development	30.00	32.00	10.11	7.89	-9.80	1
Current conditions (do nothing).	0	0	0	0	0	5

Table 2 Assessment of the risk level in ascending order of analysis, simulation Monte Carlo. This value is the distribution of the NPV Chance thick. Which indicates the level of risk in any way with the distribution of the NPV is less risky to invest less. The result of the simulation is divided assessed as two parts: the choice is the NPV <0 is used to assess the likelihood of probability to the NPV <0 and the choices the NPV> 0 is viewed from. distribution of the NPV

The results of a Monte Carlo simulation showed that investors use ERP (Enterprise Resource Planning) module, the FA has only a few fragments. The simulations showed that the chance that the NPV is less than zero, so 17.86 million baht investment alternative than investing in alternatives. The returns show the simulation in this alternative investment. It is likely that the return is less than zero, which is the return on investment will be greater than zero.

9. References

[1] Ford, D. N., Lander, D. and Voyer, J. (2001) The Real Options Approach to Valuing Strategic Flexibility in Uncertain Construction Projects. Construction Management and Economics. 20(4): 343-352.
[2] Amram, M. and Kulatilaka, N. (1999a) Real Options, Managing Strategic Investment in an Uncertain World. Harvard Business School Press. Cambridge, MA.
[3] Benaroch, M. (2001) Option-Based Management of Technology Investment Risk. IEEE Transactions on Engineering Management. 48(4): 428-444.
[4] Brennan, M. J. and Trigeorgis, L. (2000) Project Flexibility, Agency, and Competition, New Developments in the Theory and Application of Real Options. Oxford University Press. New York.
[5] Kemna, A.G.Z. (1993) Case Studies on Real Options. Financial Management. 22(3): 259-270. Autumn 1993. [12] Trigeorgis (ed.), L. (1995) Real Options in Capital Investment. Prager. New York, N.Y.
[6] Lander, D.M. and Pinches, G.E. (1998) Challenges to the Practical Implementation of Modeling and Valuing Real Options. The Quarterly Review of Economics and Finance. 38:537-67.
[7] Amram, M. and Kulatilaka, N. (1999) Real Options - Managing Strategic Investment in an Uncertain World, Harvard Business School Press, Boston, MA.
[8] de Neufville, R., Scholtes, S., and Wang, T. (2005) "Valuing Real Options by Spreadsheet: Parking Garage Case Example," ASCE Journal of Infrastructure Systems, in press. de Weck, O.,
[9] Luenberger, D. G. (1998) Investment Science, Oxford University Press, New York, NY. Myers, S.C. (1984) "Finance theory and financial strategy," Interfaces, 14, Jan-Feb, pp. 126-137.
[10] Trigeorgis, L. (1993) "Real Options and Interactions with Financial Flexibility," Financial Management, Autumn, pp. 202 – 224.
[11] Wang, T. and de Neufville, R. (2004) "Building Real Options into Physical Systems with Stochastic Mixed-Integer Programming," 8th Annual Real Options International Conference, Montreal, Canada.
[12] Wang, T. (2003) "Analysis of Real Options in Hydropower Construction Projects – A Case Study in China," Master of Science Thesis, MIT, Cambridge, MA.

Dynamic Multi-product Multi-level Capacitated Lot Sizing Using Heuristic Method

Songwut Prakaiwichien[1, a *], Vichai Rungreunganun[2,b]

[1] Department of Industrial Engineering, Faculty of Engineering, King Mongkut's University of Technology North Bangkok 10800, Thailand

[2] Department of Industrial Engineering, Faculty of Engineering, King Mongkut's University of Technology North Bangkok 10800, Thailand

Keywords: Dynamic Multi-product Multi-level Capacitated Lot Sizing, Heuristic Method .

Abstract.

The paper studies the multi-level lot sizing problems for assembly production in capacitated, dynamic and deterministic cases. Our focus is on minimizing the sum of setup costs, inventory costs and overtime costs by using the Heuristic method which will consume very less time but results are nearly or equal to optimal solution. The algorithm of this Heuristic method is to determine the lot sizing of each product at all periods from high to low level by weighing number of set up cost with holding cost and overtime cost occurred in each period. The result confirms that the best solution of the proposed technique in comparing with the optimum solution by Lingo Software does not exceed than 5% but use processing time less than 1 second even though it is the large scale problems. The processing time is nearly the same as LINGO when run large scale problems.

Introduction

Material requirement planning (MRP) is an inventory management system based on computer software which create for the production manager in order to plan and place orders for items of dependent demand. Dependent demand items are components which determined by bill of material (BOM), for example raw materials, component part, subassemblies, which are the dependent demand that we need for determining the inventory needed and these factors depend on the level of production of the final product.

MRP are commonly used by multi level product industry in order to provide components both type and quantity in all level of production which results in sufficient material in time. MRP is design to answer what, how much and when it is needed. Therefore, MRP start with the amount of finished goods and time it is needed. Then calculate what are the requirements for the subassemblies in order to produce the final products such as component parts, raw materials etc.

Dynamic Multi-Product Multi-Level Capacitated Lot-Sizing problems are important problems occurred in lot sizing procedure. Since MRP must consider 2 important issues ie the limitations of the production resources and taking into account of total cost which compose of holding cost and setup cost . However, the MRP system is not concern about the above – mentioned issues. As a result, MRP system does not provide an optimal production plan in the aspect of total cost.

To lot sizing problems in assembly production systems by using dynamic programming algorithms and a branch and bound algorithms to obtain the optimal solution, Crowston and Wagner (1973) . Tempelmerier and Derstroff (1996) develop a Lagrangean heuristic. They also started with a Wagner-Within solution and then used a smoothing procedure to try to find a feasible solution. Ozdamar and Barbarosoglu (2000) presented another heuristic using Lagrangean Relaxation and Simulated Annealing. Dellaert , Jeunet and Jonard (2000) propose A genetic

algorithms to solve the general multi-level lot-sizing problem with time-varying costs by develop a binary encoding genetic algorithm and design five specific genetic operators to ensure that exploration takes place within the set of feasible solutions. Dellaert and Jeunet (2003) proposed A randomized multi-level lot sizing heuristic for general product structures.

The objective of this paper is to minimize total cost for Dynamic Multi-Product Multi-Level Capacitated Lot-Sizing problems under assembly production systems while no backlogging is allowed by using Heuristic method to find the solution.

Methods

Model formulation

The dynamics multi level capacitated lot sizing problem is aimed at minimizing variable production costs over a finite planning interval. The variable production costs which are considered comprise of holding costs and setup costs. The planning interval is divided into several periods and limited by the planning horizon T.

For each period in the planning interval, the end item demand is assumed to be known and has to be fulfilled without backlogging. Inventory holding costs are calculated based on the end of period inventory. Setup costs and setup times accrue for an item in each period of production.

Minimize

$$\sum_{j=1}^{j}\sum_{t=1}^{T}(sc_j Y_{jt} + h_j I_{jt} + oc_j O_{jt}) \quad (1)$$

Subject to

$$I_{j(t-1)} + X_{jt} = D_{jt} + \sum_{k\in S_j} r_{jk} X_{kt} + I_{jt} \quad , j = 1,\ldots,J ;\ t = 1,\ldots,T \quad (2)$$

$$\sum_{j=1}^{J} a_{mj} X_{jt} \le C_{mt} + O_{jt} \quad , m = 1,\ldots,M ;\ t = 1,\ldots,T \quad (3)$$

$$X_{jt} \le B_{jt} Y_{jt} \quad , j = 1,\ldots,J ;\ t = 1,\ldots,T \quad (4)$$

$$I_{jt}, O_{jt}, X_{jt} \ge 0 \quad , j = 1,\ldots,J ;\ t = 1,\ldots,T \quad (5)$$

$$Y_{jt} \in \{0,1\} \quad , j = 1,\ldots,J ;\ t = 1,\ldots,T \quad (6)$$

Indices and index sets:

j	Items or operations	, *j* =1, ..., *J*
m	Resources	, *m*=1, ..., *M*
t	Periods	, *t*=1, ..., *T*
Sj	Set of immediate successors of item *j* in the bill of material	

Where the known parameters are

amj	Capacity needed on a resource m for one unit of item *j*
Bjt	Large number, not limiting feasible lot-sizes of item *j* in period *t*
Cmt	Available capacity of resource *m* in period *t*
hj	Holding cost for one unit of item *j* in a period
ocjt	Overtime cost for one unit of item *j* in period *t*

Djt External demand for item j in period *t*

r_{jk} Number of units of item *j* required to produce one unit of the immediate successor item *k*

scj Setup cost for a lot of item *j*

and the decision variables are

Ijt Inventory of item *j* at the end of period *t*

Ojt Amount of overtime of item *j* used in period *t*

Xjt Amount of item *j* produced in period *t*

Yjt Binary variable indicating where production is allowed for item j in period t (=1, if item *j* is produced in period *t*, 0 otherwise)

The objective (1) is to minimize the sum of holding, setup and overtime costs. Equations (2) are the inventory balances to make sure that no backlogging will occur. For multi-level production, a lot-size of item *k* will result in a dependent demand for its immediate predecessor items *j*. Required capacities for lot-size production must not exceed available normal capacities (possibly extended by overtime; (3). Capacity requirements result from both production time per item times the amounts produced as well as setup times incurred with each lot. Setup constraints (4) enforce binary variables *Yjt* to unity, in case a lot of item *j* is produced in a period *t*. All variables are restricted to non-negative or binary values, respectively (5),(6).

Heuristic Method

Step 1

To choose the item for determining the lot size according to bill of material structure (BOM) from the highest to the lowest level. First item is finish products. Next item is from the next level which is subassembly of the first item. Do the same process until all items are chosen.

According to the BOM, in order to determine lot size items are divided into 3 groups:

Group 1 Finished products

Group 2 Subassemblies ,which are immediate successor, will influence the number of items which are the predecessors.

Group 3 Component Parts and Raw material which have not any parts to be made before.

Step 2

To determine lot size of group 1 (finished products). This group will produce in lot for lot according to the demand in that period.

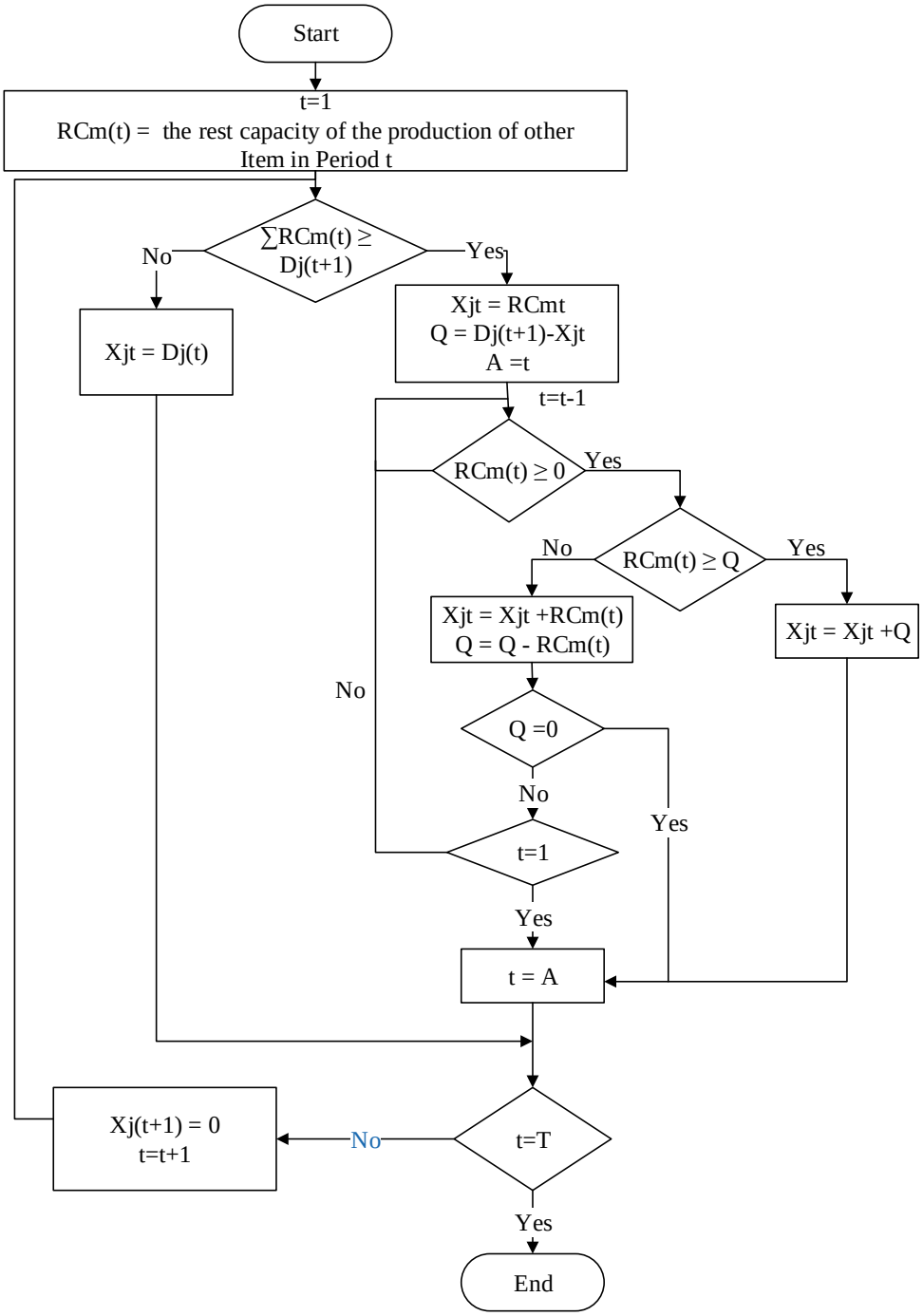
Step 3

To determine lot size of group 2 (Subassemblies). To consider which item has to share limited resources in production. Lot sizing determination can be devided into 2 cases

1) Holding cost per unit of each item are the highest. This item will be produced in lot for lot according to the demand in that period.

2) Holding cost per unit of each item are the lowest. Starting from production in lot for lot according to the demand in first period. Then, recheck accumulate remaining production capacity of resources sharing with other item. If accumulate capacity from the first period until present period is higher or equal to demand in the next period, To produce this item at present period as much as allowance capacity. Then, go back to the prior period which has available capacity and increase the amount of production step by step back until the amount of accumulate production are equal to the demand of next period. The objective of this step is to make the amount of production in next period been zero which make setup cost been zero. However, this method have to be rechecked whether it make total holding cost higher than setup cost or not. If yes, production in lot for lot according to the demand at present period. And follow the same procedure until the last period. The procedure of this method will be as shown in Fig.1.

Fig.1, Flowchart on Production Procedure on Step 3 case 2



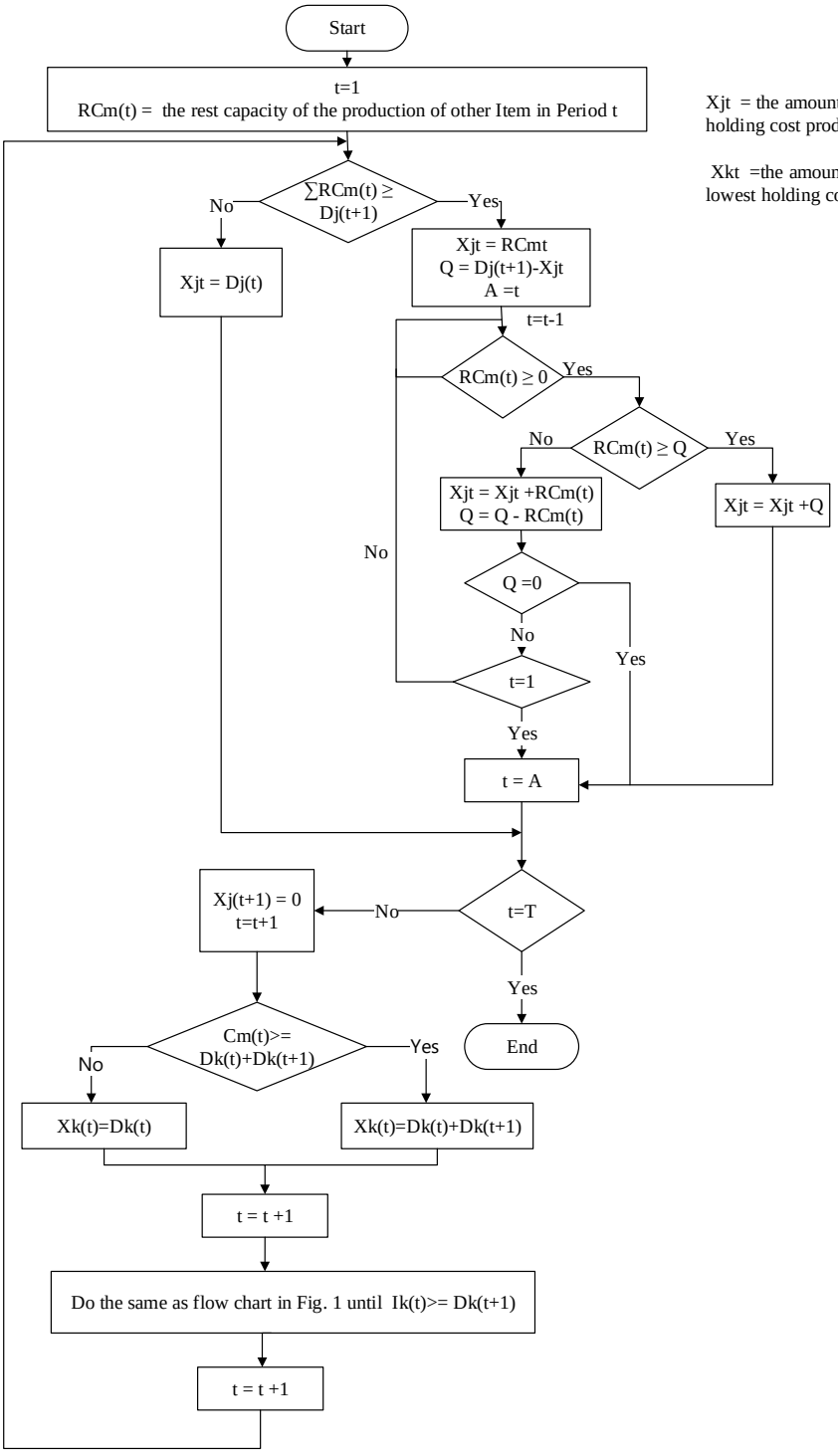
Step 4

To determine lot size of group 3 (Component Parts and Raw material). To consider which item has to share limited resources in production. Lot sizing determination can be divided into 2 cases

1) If the item which share resources with this group is not in group 3, this item will be produced in lot for lot according to the demand in that period. However, the item in group 3 will be produced by following the procedure shown by the Fig.1.

2) If the item which share resources with this group is in group 3, the production will be as shown in Fig.2.

Fig.2, Flowchart on Production Procedure on Step 4 case 2



Step 5

To calculate cumulative total cost

Result and Discussion

The performance of the Heuristics Method described in the previous section was evaluated on a set of testing problems. The algorithm was programmed with Matlab (Version 7.0) running under Windows 7 on a compatible personal computer (Model Intel(R) CORE(TM)2 Duo CPU T5250@ 1.50 GHz) . The experiments revealed that Heuristics Method could obtain good approximation solution of Dynamic Multi-Product Multi-Level Capacitated Lot-Sizing Problems in consumes processing time less than 1 second even though it is the large scale problems. Following are the examples of the experiments.

Example: (Assembly System) For assembly product structures shown in Fig.3 (4 items) constrained by two resources (A , B). The test instance data of each test set is shown in Table 1 and 2.

Fig.3, The Product structure for Example

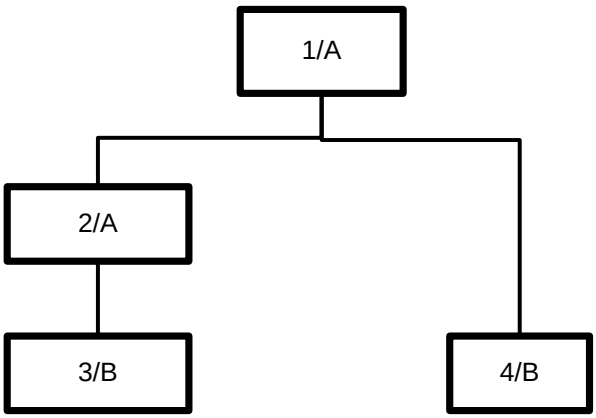


Table 1, The test instance data for all Test set

	Item 1	2	3	4
Holding cost/unit/period	6	3	2	1
Setup cost for item	200			
Overtime cost/unit/period	100			
Capacity A (all period)	30			
B (all period)	32			

Table 2, The External Demand for each test set

Test set No.	External Demand for item 1
1	15,15,16,13,17,14
2	14,15,16,13,17,14
3	13,15,16,13,17,14
4	12,15,16,13,17,14
5	11,15,16,13,17,14
6	10,15,16,13,17,14
7	9,15,16,13,17,14
8	8,15,16,13,17,14
9	7,15,16,13,17,14
10	16,15,16,13,17,14
11	17,15,16,13,17,14
12	18,15,16,13,17,14
13	19,15,16,13,17,14
14	20,15,16,13,17,14
15	15,15,20,13,17,14
16	15,15,21,13,17,14
17	15,15,22,13,17,14
18	15,15,23,13,17,14
19	15,15,24,13,17,14
20	12,15,14,13,17,14,12,11
21	12,15,14,13,17,14,12,11,14
22	12,15,14,13,17,14,12,11,14,12
23	12,15,14,13,17,14,12,11,14,12,13
24	12,15,14,13,17,14,12,11,14,12,13,12
25	12,15,14,13,17,14,12,11,14,12,13,12,13
26	12,15,14,13,17,14,12,11,14,12,13,12,13,1

Table 3, Results of the Numerical Experiments

Test set No.	Best Solution found by		Difference of Solution	Times(sec)	
	Heuristic	Lingo6		Heuristic	Lingo6
1	2812	2812	0%	<1.0	<1.0
2	2424	2424	0%	<1.0	<1.0
3	2424	2424	0%	<1.0	<1.0
4	2388	2374	0.59%	<1.0	<1.0
5	2294	2291	0.13%	<1.0	<1.0
6	2197	2197	0%	<1.0	<1.0
7	2197	2117	3.78%	<1.0	<1.0
8	2121	2019	5.0%	<1.0	<1.0
9	1960	1954	0.3%	<1.0	<1.0
10	3212	3212	0%	<1.0	<1.0
11	4012	4012	0%	<1.0	<1.0
12	4812	4812	0%	<1.0	<1.0
13	5612	5612	0%	<1.0	<1.0
14	6412	6412	0%	<1.0	<1.0
15	5218	5218	0%	<1.0	<1.1
16	6018	6018	0%	<1.0	<1.2
17	6818	6818	0%	<1.0	<1.3
18	7618	7618	0%	<1.0	<1.4
19	8418	8418	0%	<1.0	<1.5
20	2816	2816	0.0%	<1.0	1
21	3144	3079	2.11%	<1.0	3
22	3456	3345	3.32%	<1.0	5
23	3782	3634	4.07%	<1.0	19
24	4094	3927	4.25%	<1.0	58
25	4420	4235	4.37%	<1.0	414
26	4734	4500	5.2%	<1.0	1440

Conclusion

In the development of Heuristic Method in solving Dynamic Multi-Product Multi-Level Capacitated Lot-Sizing Problems, the result shows that to use the Heuristic Method in finding solution. The experiment reveals that when comparing to the solution calculated by Lingo6 software does not exceed than 5 %. While this method consumes processing time less than 1 second even though it is the large scale problems.

For further research, more numerical experiments can be made and to develop the method in solving problems by using the results from Heuristic Programming. This method considering only the alternative which has potential to improve the solution and reduce processing time.

Acknowledgements

I would like to acknowledge and extend my heartfelt gratitude to Assoc. Prof. Vichai Rungreunganun for his useful comments and suggestions which have made the completion of this paper possible.

References

[1] W.B. Crowston, M.W. Wegner, Dynamic Lot Size Models for Multi stage Assembly Systems, Management Science. 20(1)(1973) 14-21.
[2] N. Dellaert, J. Jeunet, N. Jonard, A genetic algorithms to solve the general multi-level lot-sizing problem with time-varying costs, International Journal of production economics. 68(2000) 241-257.
[3] N.P. Dellaert, J. Jeunet, Randomized multi-level lot sizing heuristic for general product structures, European Journal of Operational Research. 148(2003) 211-228.
[4] L. Ozdamar, G. Barbarosoglu, An integrated lagrangean relaxation simulated annealing approach to multi-level multi-item capacitated lot sizing problem, International Journal of Production Economics. 68(2000) 319-331.
[5] H. Tempelmeier, M. Derstroff, A Lagrangean-based heuristic for dynamic multi-level multi-item constrained lotsizing with setup times, Management Science. 42(5) (1996) 738-757.

Properties of Rice Grain after mild heat treatment by Radio Frequency heating for killing insects

Pisith Sok[1, a], Sirichai Songsermpong[1, b*]

[1]Department of Food Science and Technology, Faculty of Agro Industry, Kasetsart University, 50 Nam Won Wan road, Chatuchak, Bangkok, 10900 Thailand

Keywords: Radio frequency heating, rice weevil, 2-acetyl-1-pyrroline, quality

Abstract. Radio Frequency (RF) heating is an alternative physical method to replace the conventional fumigation method. Rice quality such as physical, physico-chemical, cooking properties, and aroma after treated by 27.12 MHz RF heating machine were determined. Different rice varieties were tested including Khao Dok Mali 105 (milled and brown rice), waxy rice and colored rice (riceberry, hom nin, and sang yod). Insect mortality was studied to find the optimum condition of treatment. Crack and chalky grain were slightly increased after long exposure to RF heating, the results remain below 7%. There is no significant effect of RF heating on amylose content. Hardness and adhesiveness were slightly increased and decreased, respectively, that signify the effect of aging after RF treatment. Peak viscosity and final viscosity of treated samples compare to control showed decrease and increase, respectively, which also support the aging process accelerated by RF heating. The relationship of rice flour pasting viscosities and amylose content was also discussed. Aroma of Jasmine rice, 2-acetyl-1-pyrroline (2AP) was decreased for long exposure of RF heating. RF heating for 20 to 30 seconds is sufficient for killing insects which still keep acceptable quality of rice grains.

Introduction

Rice (*Oryzae sativa* L.) is one of the major staple foods in the world and is the most important export product in Thailand. Before consumption rice quality was reduce by insect infestation [1]. Due to the economic benefit chemical fumigation method was use widely until now [2]. However, this conventional chemical disinfestation method has led to adverse effects on human health and environment and it will be banned in the near future [3]. Therefore, alternative disinfestation methods have been investigated and developed to replace the chemical method.

Many investigations have been attempted to prove the alternative of chemical fumigation, including ionizing irradiation, controlled atmosphere, cold storage, low pressure and dielectric heating [4,5,6,7,8,9,10]. Microwave was employed to achieve 100 % insect mortality when the final rice temperature was higher than 55 °C [11]. Even though the microwave method can be successfully employed for insect control, the resulting quality of treated rice has to be considered.

The application of infrared heating has been studied, however the improvement is needed more since the insect mortality is limited to maintain the acceptable quality [12].

The feasibility of using Radio Frequency (RF) energy for disinfesting milled rice was investigated [1]. Additionally, the results showed insignificant changes in rice quality in terms of moisture, protein, fat, starch, and color. Zhou and Wang [13] further investigated the RF heating uniformity and validated the developed protocols for disinfesting not only milled rice, but also rough and brown rice.

Those works described above have proven that radio frequency heating is a promising technique that achieves not only high rice drying efficiency and improved storage stability, but also an efficient disinfestation and disinfection of rough rice. However, detailed research on rice quality after using RF as a disinfestation method has not yet been reported clearly which is questioned by

the rice mills whether the aroma is gone, the quality of rice grains and the cooking quality is changed. Consequently, the objectives of this work were (1) to study optimum RF treatment condition for controlling insects in rice grain, and (2) to compare the quality of 6 different varieties of rice before and after RF treatments at different time intervals.

MATERIAL AND METHODS

1. RF treatment and Insect mortality

Rice samples were treated by RF heating machine (Eureka Agro, model RW-X1, Thailand) in batch process with the frequency of 27.12 MHz and Power 15 kW. Duration of treatment to control the insect infestation was varied in the range of 10, 20 and 30 seconds with holding 30 seconds. Studied samples were Jasmine white rice (WR), Jasmine brown rice (BR), Sticky rice (SR), Riceberry (RB), Hom Nin (HN), Sang Yod (SY) which provided by Patum Rice Mill and Granary Public Co.Ltd, Bangkok, 10110 Thailand.

The rice weevil insects (*Sitophilus Oryzae*) were obtained from Department of Entomology, Faculty of Agriculture, Kasetsart University. During radio frequency treatment 30 adult rice weevils were mixed into 5 kg of rice sample that was subjected to Radio Frequency heating immediately [14]. Insects were checked visually 1 day after treatment and insects were not moving was suppose died. Percentage of Insect mortality (IM) was calculated by:

$$\text{IM}\ (\%) = \frac{\text{Dead insect}}{\text{Total insect}} \times 100 \quad (1)$$

2. Moisture content

Moisture Content. The moisture content (MC) of rice samples was determined according to the standard method of AOAC [15]. Five grams of the sample was dried at 105 °C in a hot air oven for 72 h, and weight difference was measured and calculated in percent wet basis. The average value obtained from three replicates for each experiment was presented.

3. Crack and chalky grain

100 grains of rice sample were placed on the glass over the light bulb to analyze the crack and chalky grains visually and weighed in order to calculate the percentage of cracked rice. After analyzing crack grain, the chalky grain was also visualized continuously. The identified chalky grain was weighted and the percentage of chalky rice was calculated [16].

4. Color

Rice grain of 100 g was sampling and the color was measured by a colorimeter (Hunterlab, UltraScan PRO, USA) for all color determinations. The color was measured in CIELAB color system. L* is a measure of the lightness, a* describes red-green color and b* describes yellow-blue color. Color difference (ΔE) was also calculated as showed in equation (2) between control and RF treated samples [17].

$$\Delta E = \sqrt{\left(L^{*}-L_{0}^{*}\right)^{2}+\left(a^{*}-a_{0}^{*}\right)^{2}+\left(b^{*}-b_{0}^{*}\right)^{2}} \quad (2)$$

5. Amylose content (AC)

Amylose content of milled rice determination was followed by the method of Juliano et al., [18]. Brown rice AC was estimated as 96% milled rice: AC * 0.96 [19].

6. Texture analysis

Cooked rice was measured with a Texture Analyzer (Texture Technologies Corp., TA-XT Plus, UK). Rice was weighted 75 g and cooked by an electric rice cooker (Panasonic SR-3NA, Thailand) and hold 10 min. Weight and water ratio of studied samples including Jasmine white rice(WR), Jasmine brown rice (BR), Sticky rice (SR), Riceberry (RB), Hom Nin (HN), Sang Yod (SY) were

1:1.7, 1:2.3, 1:1, 1:2.3, 1.2.5, 1.2.3, respectively. The 3 cooked rice grains were placed on compressing base with 1 centimeter distance from each kernel. The initial height of the compression cylindrical probe P/36R was set at 14.5 mm. The probe speed during compression was 1 mm/s and with compression strain 90%. During retraction, the probe speed was 1 mm/s. Hardness and adhesiveness were interpreted from texture graph as the maximum force in Newton (N) and negative force value in Newton second (N.s), respectively [20,21].

7. Viscosity

Pasting properties of rice flour determination was followed by Tirawanichakul et al [22], using a Rapid Visco Analyzer, RVA (Pertern 4500, Australia). RVA parameters including Peak viscosity and final viscosity were expressed in centipoise [cP].

8. Aroma of Jasmine rice, 2-acetyl-1-pyrroline (2AP)

The determination of 2AP was done by Rice Chemistry Research Lab and Center of Excellence for Innovation in Chemistry, Chiang Mai University. The methods include preparation of rice samples and determination by Headspace-Gas chromatography. [23].

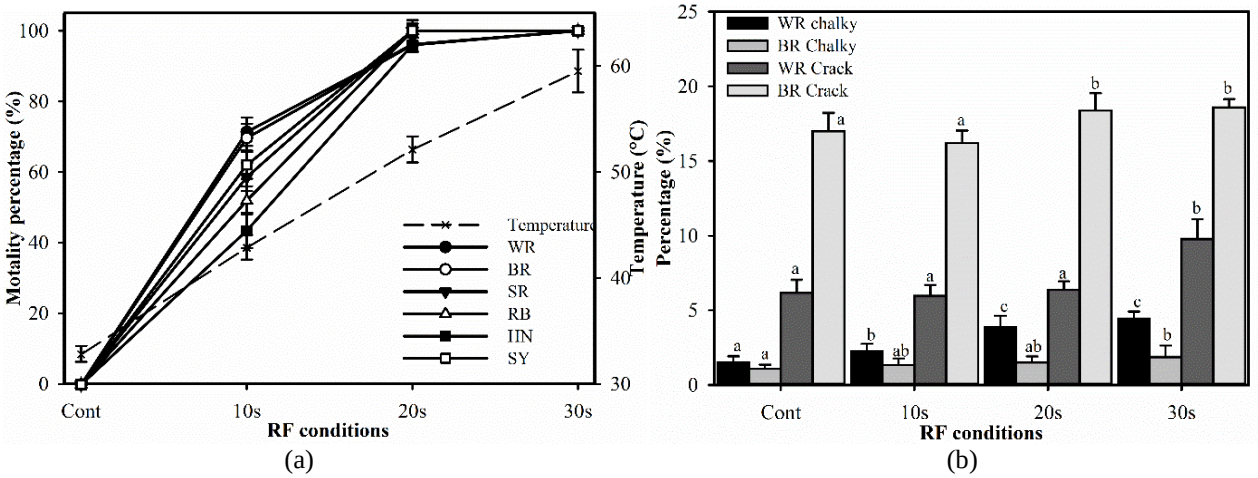
9. Statistical analysis

Analysis of variance (ANOVA) was performed to see the significant difference by using a statistical software SPSS (Version 16; SPSS Inc.; Chicago, IL, USA). Comparison between means were examined using Duncan multiple range test (DMRT) at $P \leq 0.05$ significance level. The results (three replications) were presented as means values with standard deviations.

RESULT AND DISCUSSION

1. Insect mortality

Figure 1(a) show the percentage of insect (*Sitophilus Oryzae*) mortality before and after RF heating. All the adult insect was killed completely after 30 seconds treatment for every studied rice varieties. In addition, after 20 seconds treatment the infested *Sitophilus Oryzae* show 100% killed for Sticky rice, Riceberry, and Sang Yod rice. The temperature after treatment also indicate. The temperature of Control, 10s, 20s, and 30s show in average 32.83, 42.89, 52.14, and 59.53 °C, respectively. Based on the obtained data after 10s treatment the temperature is not sufficient to control weevils completely. The previous study reported that temperature increase to 50 °C with 6 minutes holding time can reach 100% mortality [24] which correspond to this data the temperature more than 50 ° with shorter time holding.



(a) (b)

Figure 1 (a): insect mortality and temperature after RF heating, (b) Crack and chalky grain after treatment

2. Crack and Chalky grain

Cracks and chalkiness can be affected by many factors such as growing method, drought, rain, postharvest handling, drying, quality of milling, fluctuation of temperature and moisture during storage and transportation. Cracks and chalkiness lead to low market price of rice grain.

In this study Figure 1 (b), Jasmine white rice chalkiness after treatment for 10s, 20s, 30s the data showed an increase of 0.03, 0.38 and 0.45 %, respectively. However, percentage of chalky and crack found in WR remain below 7% which is the acceptable according to Thai agricultural standard of Khao dok mali 105 white rice. This finding follow the same trend with published work [12] which use infrared as a heating source to control insects. The chalky and crack tend to increase with the temperature increase and longer RF exposure. This could be due to the stress formation resulting from moisture gradients. Higher heating might result in samples with internal cracks and fissures leading to more leached out components during cooking and then resulting in lower stickiness. [16].

3. Moisture content

Moisture content of rice sample before and after exposure to RF was presented in Figure 2. Based on the result, moisture content of studied samples were significantly reduced. The highest moisture reduction was found after 30 seconds treatment by 0.65, 0.22, 0.18, 0.24, 0.55 and 0.38 for WR, BR, SR, RB, HN and SY, respectively.

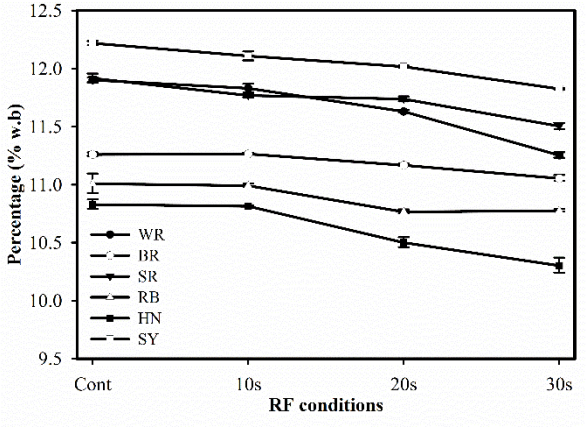


Figure 2: Moisture content of studied sample after RF heating

Current finding agreed with the study of Duangkhamchan et al [12] state that grain moisture were reduce significantly after infrared irradiation at 60 °C for 3 minutes. Prior to the above studies, Wanitchang and Wanitchang [25] found from their experiment that drying rice grain at high temperature could reduce moisture content more rapidly and achieve lower level of final moisture in rice grain compared to the lower alternative temperature. Similarly, Janhang et al [26] found that the use of 27.12 MHz RF at 85 °C for 3 minutes could reduce the moisture content of the treated rice grain by 9.3% to the lowest possible level.

4. Color

Physical property is the main and primitive quality of rice; therefore, Figure 3 shows the color of rice grains before and after radio frequency heat treatment. By observing the graph, color difference of colored rice such as RB and HN highly changed compared to other varieties after RF heating for 30 seconds by1.87±0.22 and 2.02±0.43, respectively. Meanwhile, color difference of normal rice including WR, BR and SR at these conditions was slightly changed during haft minutes treatment by 0.86±0.13, 0.18±0.02 and 0.50±0.06, respectively. However, the finding in this study remain below $\Delta E = 2.3$ which corresponds to Just Noticeable Difference (JND) [27)].

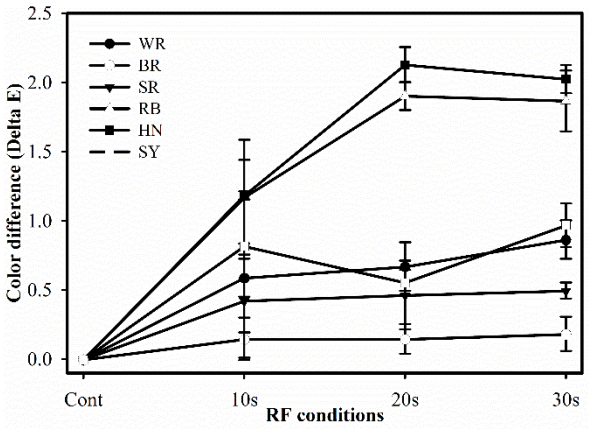


Figure 3 : Color difference (ΔE) after RF treatment

In this case, the color change may be due to the higher moisture gradients corresponding with high temperature generated inside the rice kernel that facilitate the pigment of color moving from bran layer to endosperm [28, 29]. In addition, the condensation of water during RF heating might lead to an increasing the amount of free amino acid [30]. With rapid increase in temperature, Maillard reaction and the infusion of color substances from rice bran into endosperm may be accelerated [31]. In Jasmine rice, the outer layer is removed before heating and the water loss and change in chalky area of rice grains might be correlated to the increased whiteness of rice. This phenomenon is well-known and consistent with report of Rordprapat et al [32]. The possible explanation could be the fact that the material properties in a rice kernel change when grain temperature surpasses the glass transition temperature, ~50 °C for rice with MC of 12 % wb [33], at which the starch goes from a glassy into a rubbery state, and also typically occurs in rice drying [16, 33].

5. Texture properties

Texture properties of cooked rice play an important role in consumer preference. Hardness and stickiness of cooked rice treated at different RF heating conditions are presented in Figure 4.

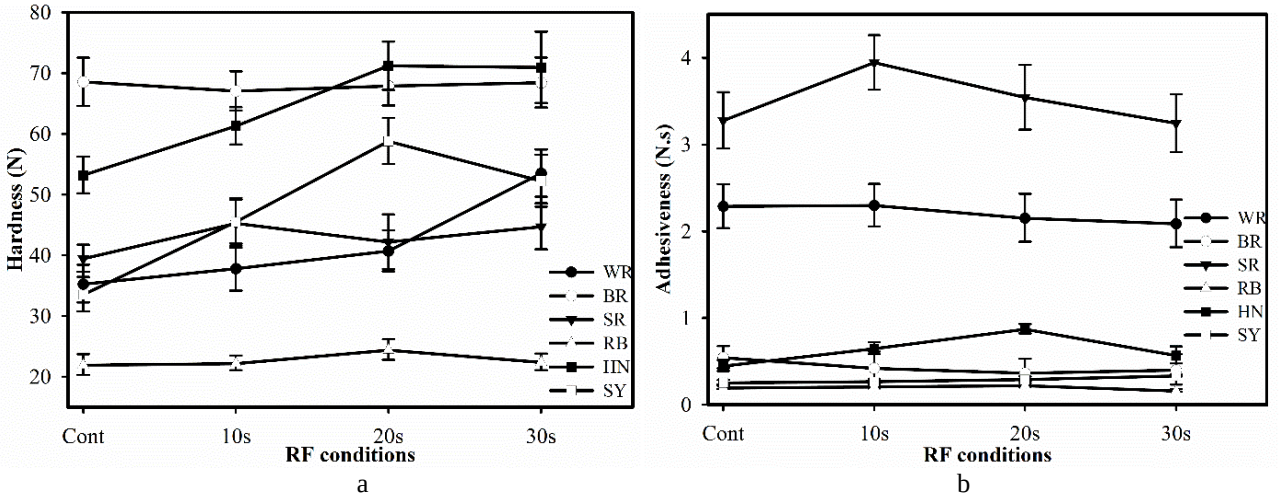


Figure 4 (a&b) : the effect of RF treatment on cooked rice texture.

The hardness of treated samples under 30 seconds were higher than those of untreated rice in this study. In contrary, Duangkhamchan et al [12] report that after heated by infrared heating hardness of cooked rice sample was significantly reduce. Apart from other physical characteristics, the texture of cooked rice was also dependent on chemical characteristics of raw kernels such as amylose content and temperature of gelatinization [34]. The hardness and stickiness were affected by amylose and short chain of amylopectin leached out during cooking process [35]. Adhesiveness values decreased in all studied samples with higher temperature and longer exposure time, except

HN which remain slightly increase. Reduction of stickiness of cooked rice samples followed the same trend of previous study [12]. Another researcher that work with microwave energy also reported the diminution or stickiness and augmentation of hardness of cooked rice sample, this effect was stated as aging effect on rice grain [36].

6. Amylose content

represents the amount of amylose content in percentage and the effect of radio frequency heating. According to the obtained data, it is more likely unable to observe the effect of RF treatment. However, the data play a fundamental role to provide the amylose content information of the different varieties rice in Thailand. Amylose content is a chemical characteristic of rice that can affect cooked rice texture [34]. Sticky rice (SR) is a very low amylose content rice (6.32±0.06%), and Jasmine milled/brown rice (WR/BR), Riceberry (RB), Hom Nin (HN), Sang Yod (SY) are low amylose content rice (15.11±0.14, 15.37±0.27, 14.56±0.22, 16.86±0.33, 15.40±0.22, respectively).

Table 1: Amylose content after RF treatment*

Rice samples	Amylose content (%)			
	Control	10s	20s	30s
WR	15.11±0.14[a]	15.13±0.17[a]	15.05±0.28[a]	15.15±0.11[a]
BR	15.37±0.27[a]	15.60±0.27[a]	15.67±0.05[a]	15.34±0.00[a]
SR	6.32±0.06[a]	6.40±0.06[a]	6.41±0.06[a]	6.38±0.06[a]
RB	14.56±0.22[a]	14.48±0.11[a]	14.66±0.07[a]	14.64±0.00[a]
HN	16.86±0.33[a]	16.83±0.16[a]	17.09±0.22[a]	16.88±0.14[a]
SY	15.40±0.22[a]	15.10±0.43[a]	15.25±0.33[a]	15.29±0.05[a]

*Different letters in the same column denote significant difference ($p<0.05$)

7. Viscosity

The values of Rapid Visco Analysis (RVA) pasting properties of six different rice cultivars with four conditions of RF heating are presented (centipoise “cP”and Standard deviation) in the Figure 5.

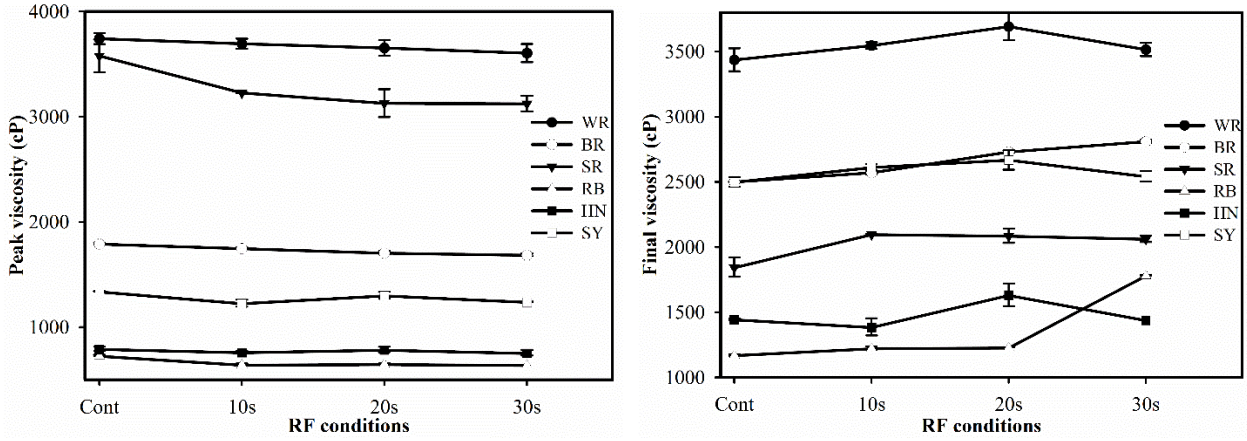


Figure 5: Viscosities of rice flour samples after RF treatment

Peak viscosity of WR remained highest compare to other varieties and decrease during treatment time by 3742.50±51.62, 3694.00±48.08 3654.00±74.95 and 3605.50±85.56 cP for untreated, 10s, 20s and 30s, respectively. This finding was inconsistent to the previous work [36] stated the peak viscosity was increase after dielectric heating (microwave energy). Peak viscosity, which reflects the swelling extent of the starch granules, was higher for WR flour in relation to their higher starch and lower protein content. The amylase activity of the BR flours certainly played a significant role in reducing peak viscosity, also taking into consideration the low heating gradient adopted. Another observation found that viscosity of flour sample that did not contain bran layer give higher peak

viscosity which is agree with the study of Wu et al [21] suggest that viscosity of samples that contain bran layer are significantly lower than bran polished rice. In addition, viscosity after cooling (final viscosity) was higher for the milled rice, in particular for those varieties that presented a higher percentage of amylose. After treated by RF heating final viscosity of all flour samples were increase which was agreed to the study of Le [36]. Other researchers stated that final viscosity increase is the essential change that could indicate rice aging phenomena [37, 38, 39]

8. 2-Acetyl-1-Pyrroline (2AP)

Khao Dok Mali 105 (KDML105) or Jasmine white rice (WR) and brown rice (BR), in addition to its good physical quality, has been well known as the most popular aromatic Thai rice. Therefore, it is essential to determine 2AP content (presented in Figure 6) affected by different RF temperatures and heating times.

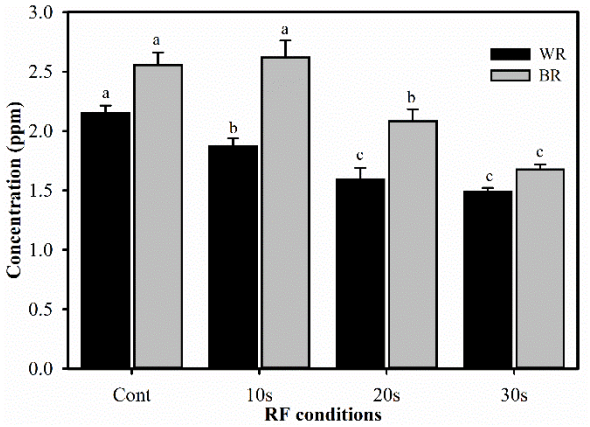


Figure 6: RF effect on Jasmine rice aroma.

The 2AP concentrations of control sample (without RF heating) white rice and brown rice were 2.15 and 2.56ppm, respectively. This indicated the concentration of Jasmine rice aroma in Brown rice was higher than white rice, this may due to the bran layer itself content 2 AP and it play a role as a protection of aroma compound. In the previous report [23, 40] found that the concentration of 2 AP in Brown rice remain higher than that in white rice. By observing the 2AP concentration after RF treatment with both varieties, significantly reduced due to the heat process and the high reduction sensitivity of aroma compound itself. Buttery et al [41] also reported consistently to the current finding that 2 AP compound is instable. This trend was consistent with the results obtained by Wongpornchai and colleagues [42], indicating that a lower 2AP concentration was found when using higher temperature and longer drying time. However, after 30s treatment both WR and BR remain 1.49±0.03 and 1.68±0.04ppm, respectively which is still higher than 2 AP odor threshold by 0.1ppb [41]

CONCLUSIONS

Properties of rice samples have been studied, radio frequency didn't have significant effect amylose content. However, Moisture content, % Chalky and crack, Texture, Viscosity, and 2AP concentration were affected significantly by treatment conditions. To maintain the good quality after heating, duration should be in between 20s and 30s.

ACKNOWLEDGEMENTS

First author would like to acknowledge Eureka Agro Machinery for accommodating this research project and Department of Food Science and Technology and Faculty of Agro Industry, Kasetsart University that offered Master Degree scholarship.

REFERENCES

[1] L.Zhou, B. Ling, A. Zheng, B. Zhang, S. Wang, Developing radio frequency technology for postharvest insect control in milled rice. J. Stored Prod. Res. 62 (2015) 22–31.
[2] S. Rajendran, V. Sriranjini, Plant products as fumigants for stored-product insect control. J. Stored Prod. Res. 44 (2008) 126–135.
[3] S. Wang, J. Tang, Radio frequency heating: a new potential means of post-harvest pest control in nuts and dry products. J. Zhejiang Univ. Sci. 5:10 (2004) 1169–1174.
[4] P.A. Follett, Effect of irradiation on Mexican leafroller (*Lepidoptera: Tortricidae*) development and reproduction. J. Econ. Entomol. 101 (2008) 710–715.
[5] L.G. Neven, L. Rehfield-Ray, Combined heat and controlled atmosphere quarantine treatments for control of western cherry fruit fly in sweet cherries. J. Econ. Entomol. 99 (2006) 658–663.
[6] M. Aluja, F. Diaz-Fleischer, J. Arredondo, J. Valle-Mora, J. Rull, Effect of cold storage on larval and adult *Anastrepha ludens* (*Diptera: Tephritidae*) viability in commercially ripe, artificially infested Persea Americana 'Hass'. J. Econ. Entomol. 103 (2010) 2000–2008.
[7] S. Jiao, J.A. Johnson, J. Tang, D.S. Mattinson, J.K. Fellman, T L. Davenport, S. Wang, Tolerance of codling moth, and apple quality associated with low pressure/low temperature treatments. Postharvest Biol. Technol. 85 (2013) 136–140.
[8] S. Zhao, C. Qiu, S. Xiong, X. Cheng, A thermal lethal model of rice weevils subjected to microwave irradiation. J. Stored Prod. Res. 43 (2007) 430–434.
[9] S. Wang, J. Tang, J.A. Johnson, R.P. Cavalieri, Heating uniformity and differential heating of insects in almonds associated with radio frequency energy. J. Stored Prod. Res. 55 (2013) 15–20.
[10] B. Alfaifi, J. Tang, Y. Jiao, S. Wang, B. Rasco, S.S. Jiao, S. Sablani, Radio frequency disinfestation treatments for dried fruit: model development and validation. J. Food Eng. 120 (2014) 268–276.
[11] D.N. Yadav, T. Anand, M. Sharma, R. Gupta, Microwave technology for disinfestation of cereals and pulses: an overview. J. Food Sci. and Tech. 51:12 (2014) 3568–3576.
[12] D. Wasan, A. Phomphai, R. Wanna, L. Wiset, J. Laohavanich, F. Ronsse, J.G. Pieters, Infrared heating as a disinfestation method against *Sitophylus oryzae* and its effect on textural and cooking properties of milled rice. Food Bioprocess Tech. 10:2 (2016) 284–295.
[13] L. Zhou, S. Wang, Verification of radio frequency heating uniformity and *Sitophilus oryzae* control in rough, brown, and milled rice. J. Stored Prod. Res. 65 (2016) 40–47.
[14] S.O. Nelson, W.K. Whitney, Radio frequency electric fields for stored grain insect control. Trans. ASAE. 962 (1960) 133-137.
[15] AOAC. 1995. Official methods of analysis (16th ed.). Washington, DC: Association of official Agricultural Chemists.
[16] A.G. Cnossen, M.J. Jiménez, T.J Siebenmorgen, Rice fissuring response to high drying and tempering temperatures. J. Food Eng. 59 (2003) 61-69.
[17] L. Lieve, D.B. Els, E.V. Greet, S.V. Wim, D. Veerle, D.M. Walter, A.D. Jan, Effect of milling on colour and nutritional properties of rice. Food Chem. 100 (2007) 1496–1503.
[18] B.O. Juliano, C.M. Perez, A.B. Blackeney, T. Castillo, N. Kongseree, B. Laignelet, T. Lapis, V.V.S. Murty, C.M. Paule, B.D. Webb, International cooperative testing on the amylose content of milled rice. Starch/Stärke. 33 (1981) 157-162.
[19] A.P. Resurreccion, Y. Tanaka, B.O. Juliano, Nutrient content and distribution in milling fractions of rice grain. J. Sci. Food Agr. 30 (1979) 475–481.
[20] A.A. Perdon, T.J. Siebenmorgen, R.W. Buescher, E.E. Gbur, Starch Retrogradation and Texture of Cooked Milled Rice during Storage. J. Food Sci. 64 (1999) 828-832.
[21] W. Jianyong, C. Jun, L. Wei, L. Chengmei, Z. Yejun, L. Dawen, L. Zhongqiang, G. Xiaojuan, Effects of aleurone layer on rice cooking: A histological investigation. Food Chem. 191 (2016) 28–35.

[22] S. Tirawanichakul, O. Bualuang, Y. Tirawanichakul. Study of drying kinetics and qualities of two parboiled rice varieties: Hot air convection and infrared irradiation. Songklanakarin J. Sci. Technol. 34:5 (2012) 557-568.
[23] T. Sriseadka, S. Wongpornchai, P. Kitsawatpaiboon, Rapid method for quantitative analysis of the aroma impact compound, 2-Acetyl-1-pyrroline, in fragrant rice using automated headspace gas chromatography. J. Agric. Food Chem. 54 (2006) 8183-8189.
[24] L. Zhou, S. Wang, Verification of radio frequency heating uniformity and *Sitophilus Oryzae* control in rough, brown, and milled rice. J. Stored Prod. Res. 65 (2016) 40-47.
[25] J. Wanitchang, P. Wanitchang, Development of the heated and moist air conditioner for rough rice. Rajamangala University‘s research report, Rajamangala University of Technology Tawan-ok (1995) 36 p.
[26] P. Janhang, N. Krittigamas, W. Lücke, S. Vearasilp, Using radio frequency heat treatment to control the insect *Rhyzopertha dominica* (F.) during storage in rice seed (*Oryza sativa* L.). In Proceeding of International Agricultural Research for Development. (2005) October 11-13. Stuttgart-Hohenheim, Germany.
[27] S. Gaurav, Digital color imaging handbook. CRC Press, 2002, ISBN: 0-8493-0900-x.
[28] R.S.N. Rao, S.O. Juliano, Effect of parboiling on some physicochemical properties of rice. J. Agric. Food Chem. 11 (1970) 289–294.
[29] M. R. Islam, N. Shimizu, T. Kimura, Energy requirement in parboiled and its relationship to some important quality indicators. J. Food Eng. 63:4 (2004) 433─439.
[30] H. Iyota, N. Nishimura, T. Onuma, T. Nomura, Drying of sliced raw potatoes in superheated steam and hot air. Drying Tech. 19 (2002) 1411–1424.
[31] C. Inprarsit, A. Noomhorm, Effect of drying air temperature and grain temperature of different type of dryer and operation on rice quality. Drying Technology. 19:2 (2001) 389–404.
[32] W. Rordprapat, A. Nathakaranakule, W. Tia, S. Soponronnarit, Comparative study of fluidized bed paddy drying using hot air and superheated steam. J. Food Eng. 71 (2005) 28–36.
[33] A.G. Cnossen, T.J. Siebenmorgen, The glass transition temperature concept in rice drying and tempering: effect on milling quality. Trans ASAE. 43:6 (2000) 1661–1667.
[34] C. Mestres, F. Ribeyre, B. Pons, V. Fallet, F. Matencio, Sensory texture of cooked rice is rather linked to chemical than to physical characteristics of raw grain. J. Cereal Sci. 53 (2011) 81─89.
[35] O. Rewthong, S. Soponronnarit, C. Taechapairoj, P. Tungtrakul, S. Prachayawarakorn, Effects of cooking, drying, and pretreatment methods on texture and starch digestibility of instant rice. J. Food Eng. 103 (2011) 258–264.
[36] Q.T. Le, S. Songsermpong, P. Rumpagaporn, A. Suwanagul, S. Wallapa, Microwave heating for accelerated aging of paddy and white rice. Aust. J. Crop Sci. 8:9 (2014) 1348-1358.
[37] A. A. Perdon, P.P. Marks, T.J. Siebenmorgen, N.B. Reid, Effect of rough rice storage conditions on the amylograph and cooking properties of medium-grain rice cv. Bengal. Cereal Chem. 74 (1997) 861-867
[38] S.Y.M. Indudhara, C.M Sowbhagya, K.R. Bhattacharya, Changes in physicochemical properties of rice with aging. J. Sci. Food Agri. 29 (1978) 627-639.
[39] A. Noomhorm, N. Kongseree, N. Apintanapon, Effect of aging on quality of glutinous rice crackers. Cereal Chem. 74 (1997) 864-867
[40] S. Mahatheeranont, S. Keawsa-ard, K. Dumri, Quantification of the rice aroma compound, 2-Acetyl-1-pyrroline, in uncooked Khao Dawk Mali 105 Brown rice. J. Agric. Food Chem. 49 (2001) 773-779.
[41] R.G. Buttery, L.C. Ling, B.O. Juliano, Chem. Ind. (1982) 958-959.
[42] S. Wongpornchai, K. Dumri, S. Jongkaewwattana, B. Siri, Effects of drying methods and storage time on the aroma and milling quality of rice (*Oryzae sativa* L.) cv.Khao Dawk Mali 105. Food Chem. 87 (2004) 407-414.

Development of Multimedia Package on Microstrip Antenna for Learning in Telecommunication Engineering

Pisit Sonla[a*], Anucha Chaichan[b], Pawana Choosiri[c] and Wannapa Manosueb[d]

Department of Electronic and Telecommunication Engineering, Faculty of Industrial Education, Rajamangala University of Technology Phra Nakhon, Bangkok 10800, Thailand

Keywords: Multimedia Package, Microstrip Antenna, Telecommunication Engineering

Abstract. The objective of the research was to develop and find an efficacy of multimedia package on microstrip antenna for learning in telecommunication engineering. The research tools consisted of of the content sheets, experimental set, PPT presentation and tests. The tools were firstly observed by 3 experts and evaluated by 15 student sample group who registered in microwave subject in industrial education program of Rajamangala University of Technology Phra Nakhon. The research results showed that 1) the multimedia package on microstrip antenna had an efficiency of 80.44/80.00 which was higher than the standard of 80/80 according to the hypothesis and 2) after learning, students' satisfactions were in a high level ($\bar{X} = 3.93, S.D. = 0.84$). In conclusion, the multimedia package on microstrip antenna can be used effectively for learning and teaching in telecommunication engineering.

Introduction

The education research and development of modern microwave circuits are much stronger within the microwave engineering [1]. However, teaching and learning for content of telecommunication engineering focus to calculate, to analysis and to design the microwave circuits such as microstrip line, filters, waveguide circuits and planar circuits. It is the basis of microstrip antenna. Thus, students should have knowledge of the calculation, analysis and design. It can apply their knowledge to design and construct microstrip antennas. Now, computer technology and communication have been developed very progressively and have been integrated into educational technology and encourage students to have more knowledge and skills.

After the surveying by using interview of teaching and learning conditions of microwave engineering found that microwave subject was the most essential and important subject with content have content is difficult to understand. The most instruction media have not several, have not enough for the students and uncomfortable by used.

In this paper we present the development of multimedia package on microstrip antenna for learning in telecommunication engineering. Multimedia package have been used to augment and supplant computers in classrooms because they are readily available, inexpensive and easy for educators to use. Multimedia package were effective classroom organizational tools for educators. Student can used an education tool for learning with this developed multimedia package.

Microstrip Patch Antenna

Microstrip antenna technology began its rapid development in the late 1970s. By the early 1980s basic microstrip antenna elements and arrays were fairly well establish in term of design and modeling [2]. In the last decades printed antennas have been largely studied due to their advantages over other radiating systems, which include: light weightiness', reduced size, low cast, conformability and the ease of integration with active devices [3]. From fig 1. microstrip patch antenna consists of a radiating patch on one side of a dielectric substrate and a ground plane on the other side. The patch is generally made of conducting material such as copper or gold and can take

any possible shape. The radiating patch and the feed lines are usually photo etched on the dielectric substrate. Microstrip patch antennas radiate primarily because of the fringing fields between the patch edge and the ground plane. It can be fed by a variety of methods [4].

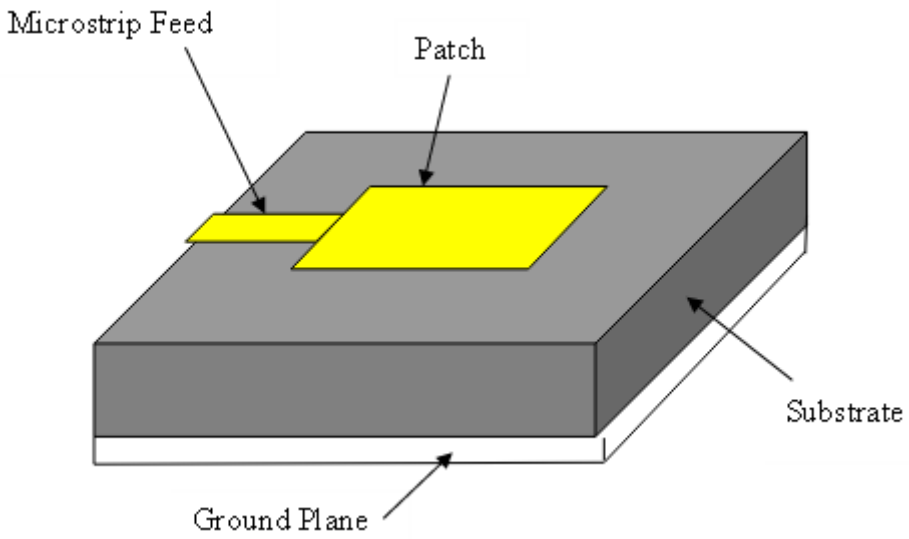


Fig 1. Structure of a microstrip patch antenna

Research Methodology

The research process consisted of analyzing the course curriculum of microwave subject, to determine the topic of teaching content and to define the behavioral objectives that provides a framework for the teaching [5]. However, presently the most instruction media have not several, have not enough for the students and uncomfortable by used. And then content is difficult to understand ect. In this research, we develop the research tools by analyzing the behavioral objectives that consist of the experimental set, the content sheets, PPT presentation and tests. The experimental sets of low pass filter circuit were developed using defined behavioral objectives. These consist of microstrip low pass filter using step-impedance, bandpass filter using open-loop microstrip resonator on double layer and parallel-coupled microstrip bandpass filter, as shown in Fig 2.

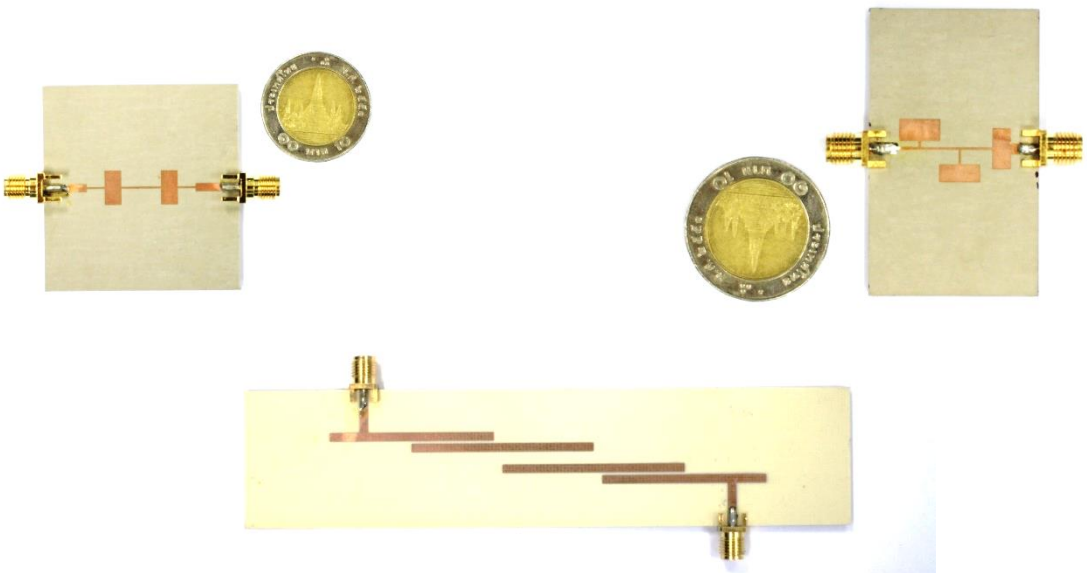


Fig 2. Circuit of design experimental set

Research Result

The research has been presented in three sections: a) the results of experimental set, b) the efficiency of the multimedia package on microstrip antenna and c) the student's satisfaction.

A) The results of experimental set

The experimental set consist of 3 sets, such as microstrip low pass filter using step-impedance, bandpass filter using open-loop microstrip resonator on double layer and parallel-coupled microstrip bandpass filter. Simulation based on theory to teach 3 lesson includes microstrip, resonance circuit, microwave filter circuit. We present an example of simulation design with theory microstrip low pass filter using step-impedance, as shown in Fig 3.

Fig 3 presents the results of microstrip low pass filter using step-impedance, we can calculate design of low pass filter circuit using chebyshev response. It was creating a calculated circuit diagram in an electromagnetic simulation program. The analyzed results are consistent with theory. [6]

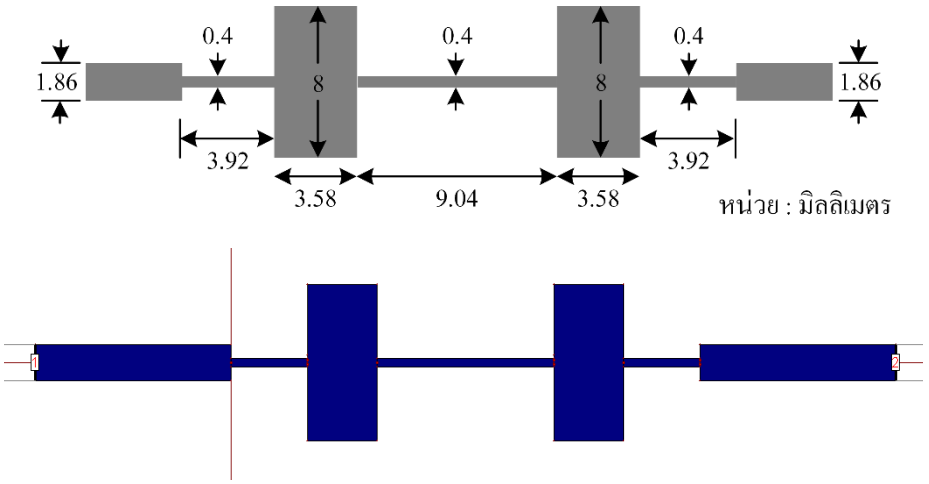


Fig 3. Microstrip low pass filter using step-impedance by simulate program IE3D

B) The efficiency of the multimedia package on microstrip antenna

The developed multimedia package for teaching in microwave course were evaluated by 3 experts whom have experience in the teaching on microwave engineering. The result found that the development of multimedia package was appropriate and quality can be used. The sampling was 15 student sample group who registered in microwave subject in industrial education program of Rajamangala University of Technology Phra Nakhon. The findings related to multimedia package had average score of total lesson test 80.44% and score of test 80.00%. So concluded that microstrip antenna had an efficiency of 80.44/80.00 which was higher than the standard of 80/80 according to the hypothesis, as shown in table 1.

Table 1. The results of efficiency of multimedia package

Score	N	$\bar{X}$	$S.D.$	Present
lesson test	15	12.07	0.10	80.44
test	15	24.00	0.17	80.00

C) The student's satisfaction

The developed of multimedia package was experimented by using 15 students who registered in microwave subject in industrial education program of Rajamangala University of Technology Phra Nakhon. The sample group was taught by using the developed of multimedia package in microwave

subject. After learning all lessons, we measured students' satisfaction of usage of developed multimedia package using questionnaire.

The findings after learning and teaching using the developed multimedia package are that the students have more knowledge and understanding of the course contents. Also the satisfaction of the students to developed multimedia package had mean value of 3.93 and S.D. equal to 0.84, as shown in table 2. Thus the developed multimedia package has good quality to use in the teaching of telecom-munication engineering of bachelor degree.

Table 2. The students' satisfaction of usage of developed multimedia package

List	$\bar{X}$	$S.D.$	Level Satisfaction
1. Learning and Teaching	3.85	0.86	High
2. PPT Presentation	3.94	0.86	High
3. Experimental set	3.98	0.83	High
4. Measurement and Evaluate	3.96	0.83	High
Average Total	**3.93**	**0.84**	**High**

Conclusions

This research has been presented the development of multimedia package on microstrip antenna for learning in telecommunication engineering of microwave subject which consists of experiment set and instruction tools. The conclusion of the findings is as following:

1) The developed experimental set can be used efficiently in the teaching of microwave subject. The measured results are correctly and consistent with the theory.

2) The developed multimedia package on microstrip antenna has content have easy to understand makes learning achievement better.

Overall the developed multimedia package on microstrip antenna were evaluated for efficiency of learning usage to accord engineering standard, therefore they may be used both theoretical and practical teaching.

Acknowledgment

I would like to thank faculty of industrial education, Rajamangala University of Technology Phra Nakhon fund in supporting this research study.

References

[1] S. Choocadee and S. Akatimagool. 2000. Waveguide Filter Design (WFD) Tool in the Teaching of Microwave Engineering. Technical Education Journal King Mongkut's University of Technology North Bangkok. 1(2). July-December 2000. pp. 11-17.

[2] D.M Pozar and D.H. Schaubert. 1995. Microstrip Antennas, the Analysis and Design of Microstrip Antennas and Arrays. IEEE Press. New York. USA.

[3] C.A. Balanis. 1998. Antenna Theory: Analysis and Design. John Wiley&Sons.

[4] K.R. Carver and J.W. Mink. 1981. Microstrip Antenna Technology. IEEE Transaction on Antennas Prapagation. Vol. AP-29 No.1. pp. 25-27.

[5] S. Khamkleang and S. Akatimagool. 2009. Microwave Planar Circuit Design Tool in the teaching of Microwave Engineerning. International Conference on Electrical Engineering/Electronics, Comupter, Telecommunications and Information Technology (ECTI-CON). pp.830-833.

[6] Rui Zhang, Raafat R. Mansour. 2009. Dual-Band Dielectric-Resonator Filters. IEEE Transaction on Microwave Theory and Techniques. Vol. 57 No. 7. pp.1760 – 1766.

Teaching Electric Circuit Laboratory with Arduino-Based Activity

Preecha Sakarung[1, a*]

[1]Section of Electrical Engineering, Faculty of Engineering,

Keywords: Electric Circuit Laboratory, Arduino, WinFACT

Abstract. In teaching electrical circuit laboratory, direct current power supplies, signal generators, multi-meters and oscilloscopes are fundamental but dated equipment which requires the replacement of Arduino microcontroller board to ease not only teaching in the university context but also self-study. The system consists of three parts: 1) Arduino board, 2) computer with WinFACT program, and 3) electrical devices connected on the breadboard. Furthermore, Graphical User Interface (GUI) on WinFACT program is rather effective in generating, measuring, and displaying signals in both direct current (DC) and modified direct current (MDC), which is made to resemble the alternate current, in which the positive sinusoidal signals previously achieved in DC are modified by subtracting with a constant value. In this paper, an application of Arduino for electric circuit laboratory course was reported in two aspects: direct current (DC) and alternate current (AC) with three cases of 1) resistance circuit in DC, 2) first-order circuit in DC, and 3) three-phase in AC. Regarding the experiment results, the error is lower than 5% compared to the electric circuit theory. This approach can be therefore implemented as the electrical circuit laboratory instruction due to its both satisfyingly accurate result and low price.

Introduction

The theory of electric circuit analysis provided the fundamental concepts and analytic techniques for electrical engineers. In fact, most electric circuit analyses aim mainly to solve the problems regarding voltage and current. There are many different software programs used for electric circuit theoretical proof such as LTspice and OrCAD-PSpice, which are simulation programs [1-2]. But in achieving the hand-on skills, it requires true understanding of the theories and familiarity with all electrical engineering tools, which can be found in the processes of wiring electrical component on solderless breadboards, or measuring voltage and current, but never experienced via any computer software. The university has therefore invested on electric circuit laboratory by purchasing many sets of lab equipment such as direct current power supplies, signal generators, multi-meters, and oscilloscopes in particular. However, there are some didactic product brands like LabVIEW-ELVIS [3] that provide all-in-one experiment platform, but with high cost. This paper then proposes a low-cost all-in-one electric circuit experiment platform using Arduino microcontroller and WinFACT program as the data acquisition system, and other electrical components and solderless breadboard are used in creating a variety of schematic electric circuits.

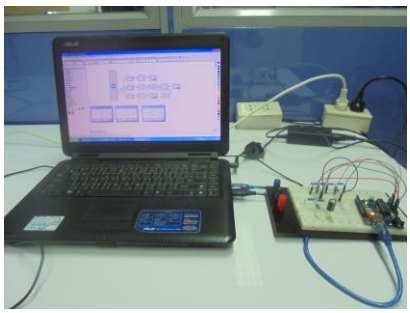


Fig. 1 the proposed all-in-one electric circuit experiment platform

Materials and Methods

Data Acquisition with Arduino and WinFACT. The data acquisition system can be used not only to collect data but also to generate arbitrary signals to stimulate the electric circuit.

Arduino microcontroller. Arduino is an open-source physical computing platform based on a microcontroller board [4]. It is a tool that allows computers to communicate with free-downloading software that runs on both the computer and the physical world. The open-source Arduino Software, ARDUINO IDE 1.8.2, is used to write and upload software code directly to the board. The Arduino UNO is a microcontroller board that is based on the ATmega328P. The board requires 5V and 16 MHz to function and it has 6 analog inputs with 14 digital input/output pins, in which 6 of them can be used as PWM outputs. Furthermore, the board will provide power supply at level 3.3 V and 5 V.

WinFACT program. WinFACT is a block diagram programming system that contains a set of tools for the analysis and simulation of the basic control systems. A lot of different interfaces enable the communication among the computer, peripherals devices, and the physical world via USB ports. The driver package retrieved from [5] consists of the BORIS driver (Arduino.dll), the DELPHI 6-source codes and the necessary firmware for the Arduino board (SERIAL.pde).

Three example cases. The following three cases demonstrate that the proposed scheme works well.

1) Resistance circuit in DC. The circuit consists of only two components which are DC sources and resistors. In Fig. 2, the circuit contains two DC voltage sources and five resistors. The three points to measure are A1, A2 and A3, where the voltages are V_1, V_2 and V_3 respectively. For the current measuring, i_1 and i_2 are the differences between the two voltage values, the first is at V_1 and V_2, and the latter is V_2 and V_3, then both are divided by the resistor value at 1000.

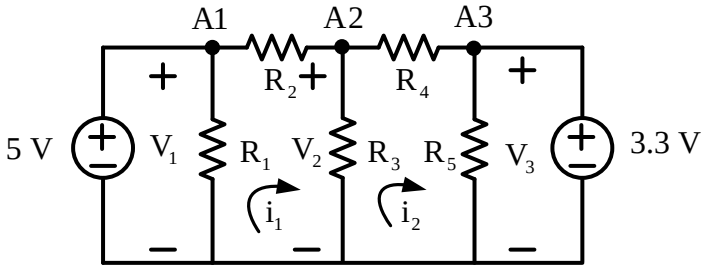


Fig. 2 the resistance circuit in DC

The value of each resistor is 1 kΩ. The equation (1) is gained by solving the given circuit in Fig. 2 with node analysis [6]. For the values of voltage and current, they are obtained respectively with the equations (2) and (3) below.

$$\begin{bmatrix} 1 & 0 & 0 \\ -0.001 & 0.003 & -0.001 \\ 0 & 0 & 1 \end{bmatrix} \begin{bmatrix} v_1 \\ v_2 \\ v_3 \end{bmatrix} = \begin{bmatrix} 5 \\ 0 \\ 3.25 \end{bmatrix} \quad (1)$$

$$\begin{bmatrix} v_1 \\ v_2 \\ v_3 \end{bmatrix} = \begin{bmatrix} 5.00 \\ 2.75 \\ 3.25 \end{bmatrix} \quad (2)$$

$$i_1 = \left(\frac{v_1 - v_2}{1000} \right) = 2.25 \text{ mA}$$

$$i_2 = \left(\frac{v_2 - v_3}{1000} \right) = -0.5 \text{ mA} \quad (3)$$

Based on Fig. 2, a block diagram with WinFACT is shown in Fig. 3. The voltage values at the three measuring points A1, A2 and A3 are 4.995, 2.752 and 3.245 volts, respectively. Also, the current values at the measuring points i_1 and i_2 are derived from the difference between the voltage values at A1 and A2 (2.243 mA) and at A2 and A3 (-0.4936 mA), respectively.

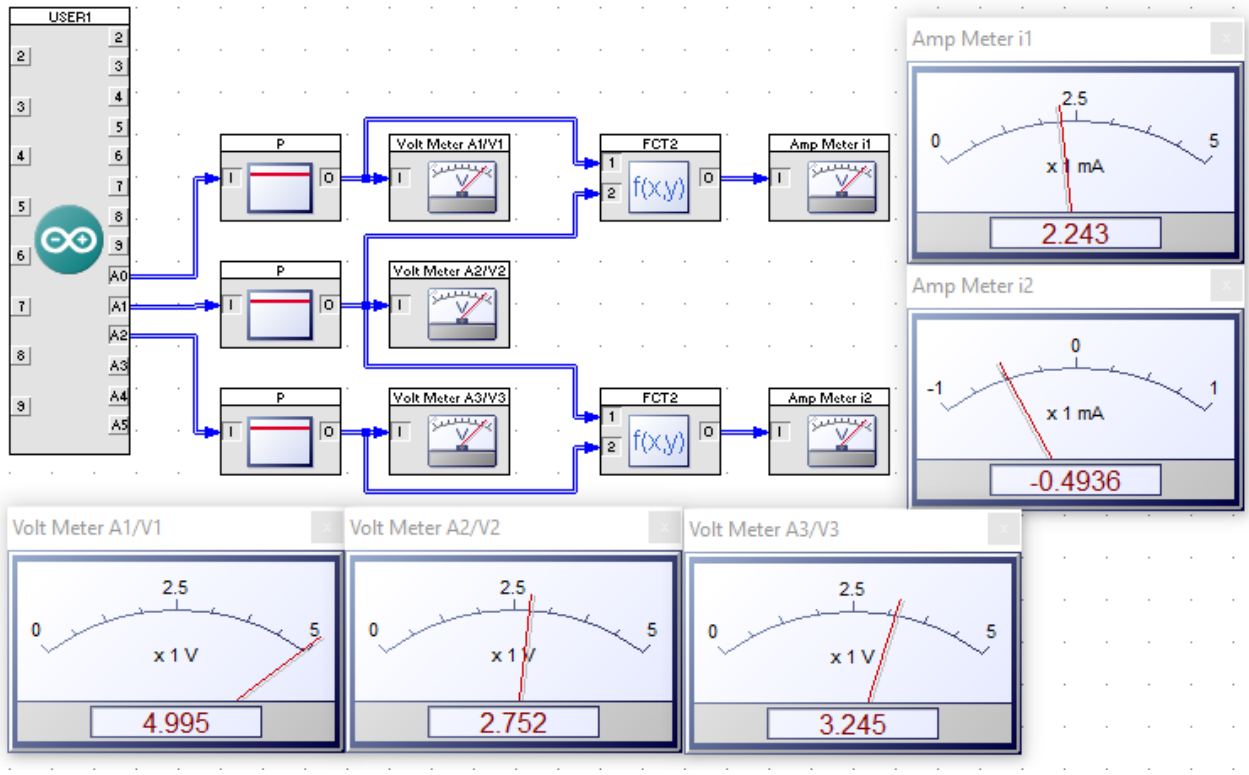


Fig. 3 the voltage and current measurements on WinFACT block diagram

2) First-order circuit in DC. There are two types of first-order circuits, which consist of either resistor-capacitor (RC) or resistor-inductor (RL). In this study, an example of RC circuit is shown in Fig. 4.

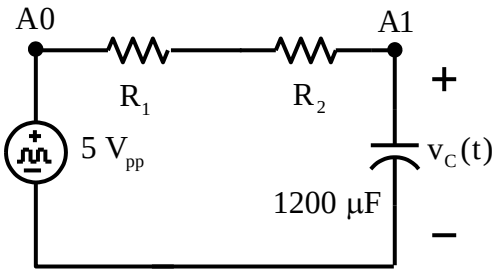


Fig. 4 the RC first-order circuit

The value of each resistor and the capacitor is 1 $k\Omega$, and 1200 μF. The two states of the circuit can be either discharging and charging, depending on the square-wave DC source. For discharging state, the equation (4) is used to find the transient response which results in the equation (5). For charging state, the complete response can be found by using the equation (6) and it shows in the equation (7).

$$V_C = V_O e^{\frac{-t}{(R_1+R_2)*C}} \qquad (4)$$

$$V_C = 5e^{-0.42t} \qquad (5)$$

$$V_C = 5 + Ae^{\frac{-t}{(R_1+R_2)*C}} \quad (6)$$

$$V_C = 5 - 5e^{-0.42t} \quad (7)$$

The time-response graph shown in Fig. 5 gives the information similar to what the oscilloscope does. Here, Arduino functions as the signal generator, which sends output signal in square wave format at pin 9 and it stimulates the circuit for desired responses. Since the analogue to digital converter is 10 bits, the values in the block diagrams A0 and A1 must be achieved by 5/1024. In this study, the transient and complete responses of RC circuit are shown below.

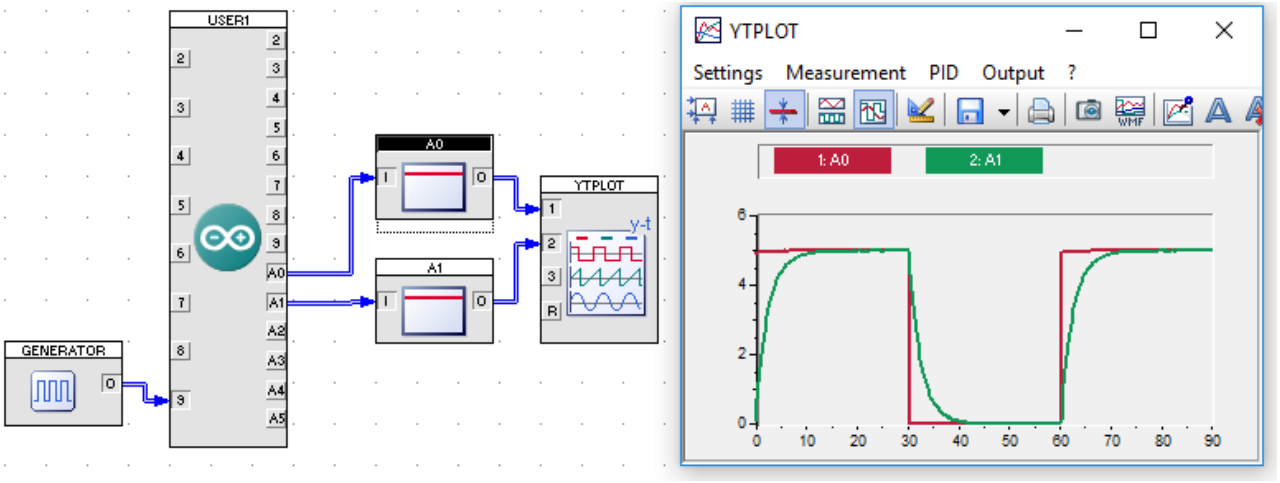


Fig. 5 the time response of first-order circuit with WinFACT

3) Three-phase circuit in AC. The single-phase AC source is created with Arduino in the form of PWM signals. The low-pass filter (RC circuit) helps PWM signals to resemble sinusoidal signals, which have the DC voltage levels between 0-5 volts. Therefore, the modified direct current (MDC), made to resemble the alternate current, can be obtained by modifying the positive sinusoidal signals previously achieved in DC by subtracting with a constant value. In this study, the three-phase AC source consists of three single-phase MDCs with time shift of 2.1 second each. Fig. 6 shows three-phase AC source with positive sequence connected to balanced RC load.

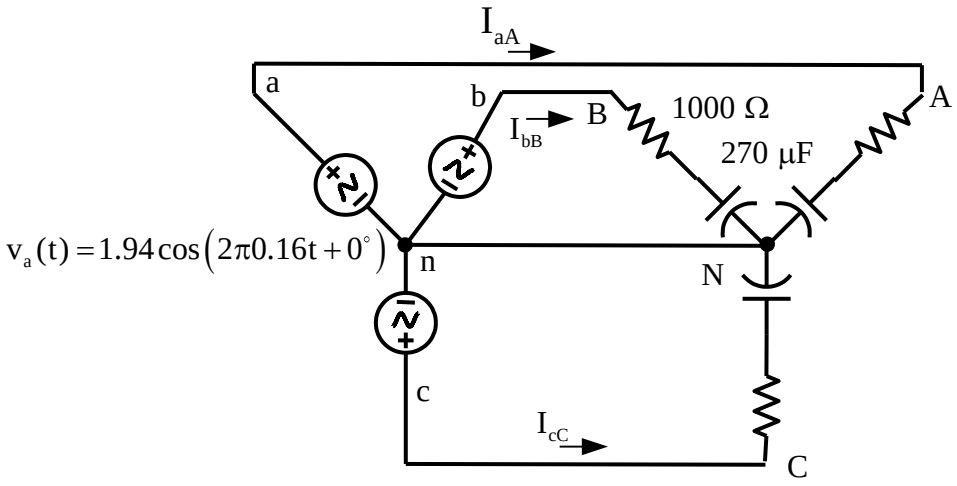


Fig. 6 the three-phase AC source and balanced RC load

In three-phase Y connections of both AC source and RC load, line voltages are gained when phase voltage is multiplied by $\sqrt{3}$, and line current equals to phase current. The equations (8-9) show the line current and the equations (10-11) show line voltage calculation.

$$I_{aA} = \left(\frac{1.94\angle 0^{\circ}}{1000 - 3684.14j} \right) \quad (8)$$

$$I_{aA} = 0.51 \text{ x } 10^{-3} \angle 74.81^{\circ} \quad (9)$$

$$V_{ab} = \sqrt{3}\left(1.94\angle\left(0^{\circ} + 30^{\circ}\right)\right) \quad (10)$$

$$V_{ab} = 3.36\angle 30^{\circ} \quad (11)$$

In Fig. 7, it shows how Arduino pins 3, 6 and 9 generate MDC signals for three-phase AC circuits a, b and c, respectively. The line voltage values are gained from the difference of two phase voltage values, depending on phase sequence.

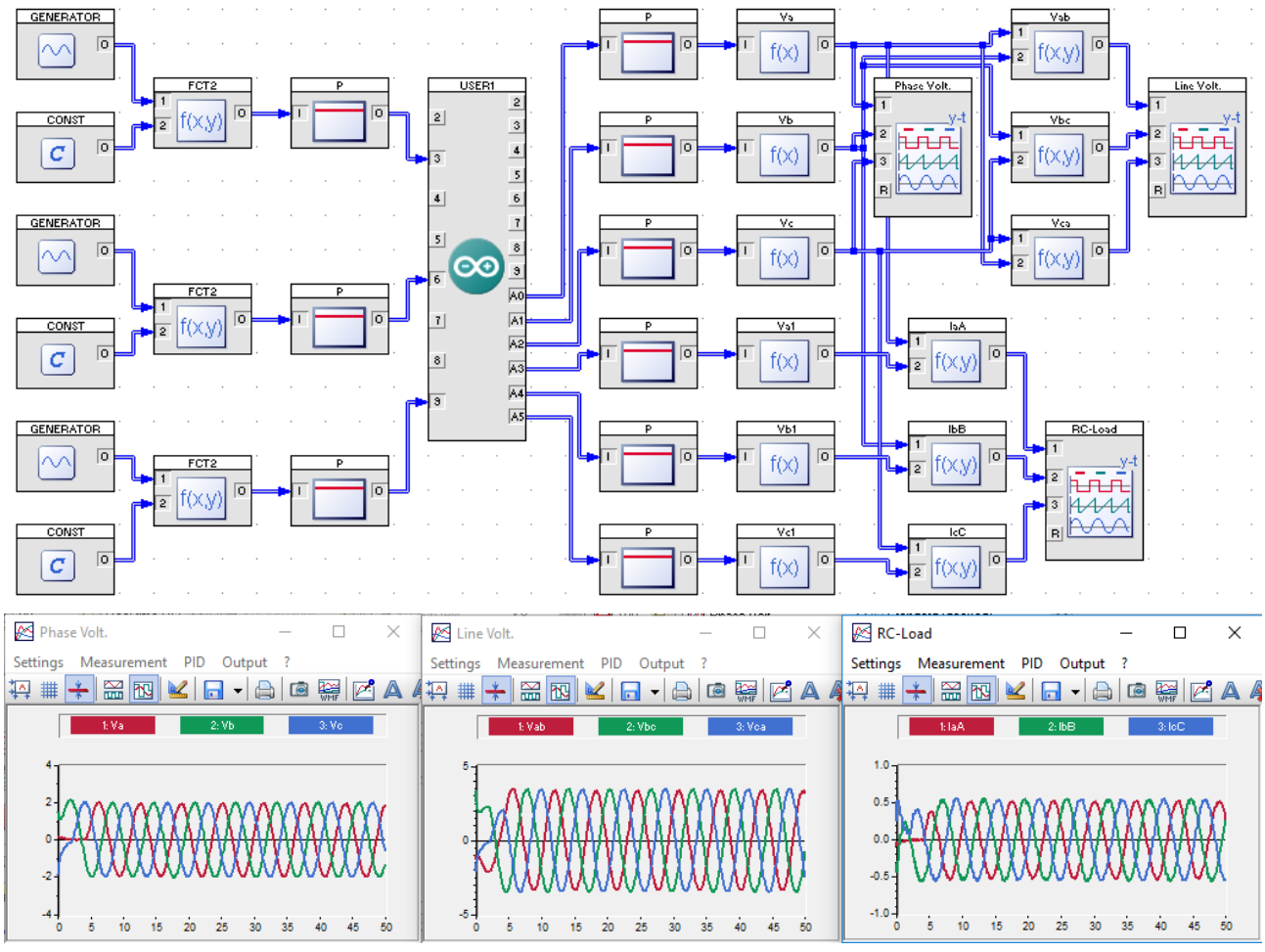


Fig. 7 the time response of three-phase circuit with WinFACT

Table 1. The comparative results between theories and experiments

cases	calculation (theories)	measurement (experiments)	errors
1	$v_1 = 5.0$ V	$v_1 = 4.995$ V	0.10 %
	$v_2 = 2.75$ V	$v_2 = 2.752$ V	0.07 %
	$v_3 = 3.25$ V	$v_3 = 3.245$ V	0.15 %
	$i_1 = 2.25$ mA	$i_1 = 2.243$ mA	0.31 %
	$i_2 = -0.5$ mA	$i_2 = -0.4936$ mA	1.28 %
2	$V_{C(t=36)} = 0.4$ V	$V_{C(t=36)} = 0.42$ V	4.76 %
	$V_{C(t=63)} = 3.58$ V	$V_{C(t=63)} = 3.66$ V	2.18 %
3	$V_{ab} = 3.36\angle 30^\circ$ V	$V_{ab} = 3.36\angle 31^\circ$ V	3.20 %
	$I_{aA} = 0.51\angle 74.81^\circ$ mA	$I_{aA} = 0.50\angle 74.3^\circ$ mA	1.96 %

Results and discussions

The DC voltage sources, square waves, and sinusoidal waves can be created by using Arduino and WinFACT. In Fig. 3, it shows the measured voltage and current values in both numeric and graphic displays. The graphical time-response values (oscilloscope-like) are also shown in Fig. 5 and Fig. 7, in which the step size of calculation for simulations is 0.28 and 0.1 second respectively. In Table 1., the comparative results show that error values are at lower than 5% each. For time-response graphs, the results that time axis shows depending on the step size of calculation; if it is too small, then time axis will show unreal time. Trial and error method is used to adjust the step size for better results which are close to the real time, which is when the time response on WinFACT is close to the one gained from oscilloscope.

Summary

In this paper, an application of Arduino and WinFACT for electric circuit laboratory course was reported in two aspects of the direct current (DC) and the alternate current (AC) with three cases of 1) resistance circuit in DC, 2) first-order circuit in DC, and 3) three-phase in AC. Regarding the experiment results, the error found is lower than 5% compared to the electric circuit theory. This approach can be therefore implemented as the electrical circuit laboratory instruction due to its both satisfyingly accurate result and low price.

References

[1] URL: http://www.linear.com/designtools/software/, access on 2/1/2017.

[2] URL: http://www.orcad.com/resources/orcad-downloads, access on 12/2/2017.

[3] URL: http://sine.ni.com/psp/app/doc/p/id/psp-458/lang/en, access on 24/2/2017.

[4] The Arduino Playground., URL: http://playground .arduino.cc/, access on 1/3/2017.

[5] Download Area., URL: http://www.kahlert.com /web/english/e_download.php/, access on 2/3/2017.

[6] W. Hayt, J. Kemmerly and S. Durbin, Engineering Circuit Analysis, 8th edition., McGraw-Hill, New York, (2012).

Excitation Energy Transfer in Light-Harvesting Complex II Detection Probe Model Using Micro-optical Device

Sarayut Pantian[1,a*], Napalai Seesod[2]

School of Physics, Faculty of Science,
Thepsatri Rajabhat University , Lopburi, 15000, Thailand

Keywords: Coherence dynamics, Nonlinear optical device, Photosynthetic light harvesting system, Rabi frequency.

Abstract

I have simulated the frequencies of photosynthetic pigment dynamics using Rabi oscillation Mode under nonlinear micro-ring resonator. The Rabi oscillation Mode is consequently generated by the photosynthetic pigments and also AlGaAs material interacting with the electromagnetic field in time-domain. The simulation results have shown that the coupling intensities affect the output intensity oscillation and also directly affect the Rabi frequency. The probability to find exciton in the excited state of the device with reducing frequencies at resonance is also calculated, in which the probabilities similarly with the results from spectroscopy technique. These results are obtained for resonance states. The Rabi frequency and oscillation in the range of terahertz is obtained. This range can be useful for photosynthetic antenna and sensing applications.

Introduction

Light harvesting in photosynthetic pigment involves the absorption, store and conversion of photon energy from light source. The following processes start with the pigment is absorbed photon energy, followed by transfer that energy and convert to chemical form suitable for processes relates a diversity of physical and chemical mechanism [1,2]. The pigments absorb light drive to the excited state and relaxed their energy after transfer the energy. The famous model is used to explain the behavior of the photosynthesis process known as The Fenna-Matthews-Olson (FMO) pigment protein complex. This model represents photosynthetic excitation energy transfers (EET), and has been widely studied experimentally and theoretically [3-7]. The most topics were studied focus on electronic coherence in photosynthetic light harvesting system that worked with some bacteria under physiological condition. Meanwhile, the researching on plants have a small number. Because of that bacteria had been lived during experiment while the pigment of plants cannot live for long. Therefore, the photosynthetic pigment of plants probe becomes an interesting topic for research.

Recently, Spectroscopy technique has been visualized two-dimensional electronic vibration of Chlorophyll a and b [8-12]. The experiments represent the presence of long-lived quantum coherence in photosynthetic pigment protein complexes spanning bacterial and plant species with a variety of functions and compositions. Its ubiquitous presence and wavelike energy transfer implicate quantum coherence as key to the high efficiency achieved by photosynthesis [6]. The theoretical and experimental studies show us the time evolution of population of FMO complex is generated the specific frequency (THz) [13]. These frequencies are recognized as the basic property that can be used to describe the interaction between atom and the electromagnetic field, which is the dynamical system of transient atom from ground state to another state with the frequency known as Rabi frequency. In principle, the interaction between the electromagnetic fields within the medium material is a small-distance-scale with short-time-scale interaction, particularly, in the resonance regime and the associated device structures require quantum mechanical description of the electronic state available in the medium. This system can be created in different ways, one of which is added the interaction Hamiltonian term in the Hamiltonian system and considered the solutions that are time-dependent functions or frequency-dependent functions depending on the Fourier transformation, where finally, Rabi frequency and oscillation can be determined.

In this paper, the Rabi oscillation of particle (photon) generates and propagates within a micro-ring resonator, a small scale optical device has been widely studied and investigated in many applications [14-18], is described by the set of Hamiltonian, which is in the form of field and its interaction of transient the electronic vibration of light harvesting complex II from local state to the excited state, which is formed by the average two level atoms. It is related to the Rabi frequency in time-domain for finding the probability of the electronic vibration of light harvesting complex II in the excited state at each period. In simulation, a system is applied to the nonlinear micro-ring circuit, which is the Alumilium Gallium Arsenide (AlGaAs) [19-22], where the practical device parameters are used in the simulation. It is not the silicon device. Therefore, AlGaAs has the nonlinear optical properties at the various wavelengths and its ultrafast nonlinearity higher than silicon, which is suitable for high-speed passive-waveguide usage. From the obtained simulation results, we have discussed the potential of using Rabi frequency and oscillation within a nonlinear micro-ring circuit for the photosynthetic antenna and sensing applications.

The advantage of the technique is that the wave-particle duality can lead to wide range of applications, which will be the important issue for future researches and investigations.

Theoretical model and Methods

A simple physical model of EET dynamics for the pigment protein complex containing N pigments, that describes each pigment as coupled two-level systems ($S_0 \rightarrow S_1$) interacting with electromagnetic field. The total Hamiltonian of the EET dynamics consists of three parts [13,22],

$$H_{\text{tot}} = H_{\text{ex}} + H_{\text{rad}} + H_{\text{ex - rad}} . \tag{1}$$

The exciton part describes the transition of electronic states at the equilibrium position of nuclei,

$$H_{\text{ex}} = \sum_{j=1}^{N} |j\rangle \varepsilon_j \langle j| + \sum_{k \neq j} |j\rangle J_{jk} \langle k| , \tag{2}$$

where $|j\rangle$ represents the state of the j th pigment where it is in S_1 state and all others are in their S_0 states. ε_j is the transition energy of the pigment excited state (site energy), and J_{jk} is the electronic coupling between the j th and the k th pigments with the atomic transition frequency ω_0 . In this paper, we only consider fluctuation in ε_j (dynamic diagonal disorder) and assumed a constant J . The unperturbed Hamiltonian for the radiation field that effects on the pigment protein complex, can be written as

$$H_{\text{rad}} = \hbar\omega a^{\dagger} a , \tag{3}$$

where a and $a^{\dagger}$ are the creation and annihilation operators for a photon energy, $\hbar\omega$. The interaction Hamiltonian is given by

$$H_{\text{ex-rad}} = -d \bullet E(t) \tag{4}$$

Eq. (4), which is described the coupling of the electromagnetic field to atom and related to the electric dipole moment operator, d . At this point we consider a spatially uniform which is well known as the dipole approximation. The pigment protein complex behaviors are described by the general state that can be expressed as a linear combination of them. Here, the general state is the evolution of time and perturbed by electromagnetic field. The dynamic in coherence state is described by the modified Heisenberg equation of motion for the matrix operator ρ is given by

$$\frac{\partial \rho}{\partial t} = -\frac{i}{\hbar}\left[H_{\text{tot}}, \rho\right] . \tag{5}$$

The coherent light probes using micro-optical ring resonator systems are proposed. The particle oscillation is propagating within the micro-ring system, in which the two-level atom system is established [22], which can be used to connect with the photosynthesis process. In fact, two level system of photosynthesis system are the paradox states of each other. The systematic of measurement system is shown in Fig. 1.

Results and Discussion

The Rabi oscillation probe can be generated by using the system as shown in Fig. 2, which is optical micro-ring system than can be fabricated to form the experimental instrument. However, this is a simulation work that uses all practical device parameters, where in this study the device materials are silicon oxide and Alumilium Gallium Arsenide (AlGaAs). In simulation, the above calculation was carried out for an AlGaAs ring resonator with radius $R_1 = R_2 = 5$ μm, device length L = 10 μm**,** linear refractive index, n_0=3.34**,** a two-photon absorption constant, $\beta = 0.5$ cm $(GW)^{-1}$, linear loss coefficient, $\alpha = 5$ $dB(cm)^{-1}$. Free carrier life time, $\tau = 1$ ns. Nonlinear refractive index $n_2 = 1.5\times10^{-4}$ cm^2 $(GW)^{-1}$. Fractional power remaining in the straight waveguide after the coupler, i.e. coupling coefficients are $r_1 = r_2 = r_3 = 0.2$.

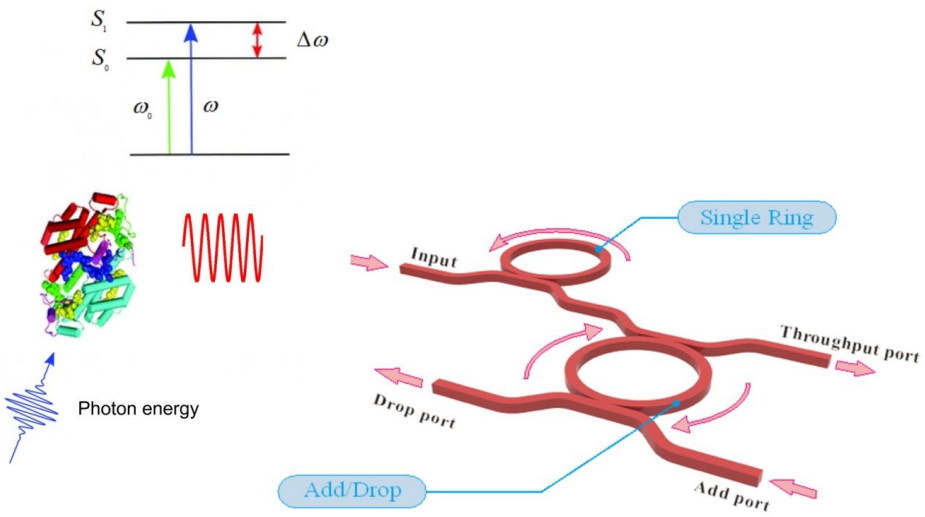


Figure 1. The proposed Rabi oscillation system using a micro-optical device which detection probes the signal from the photosynthetic relaxation energy in coherence state [23].

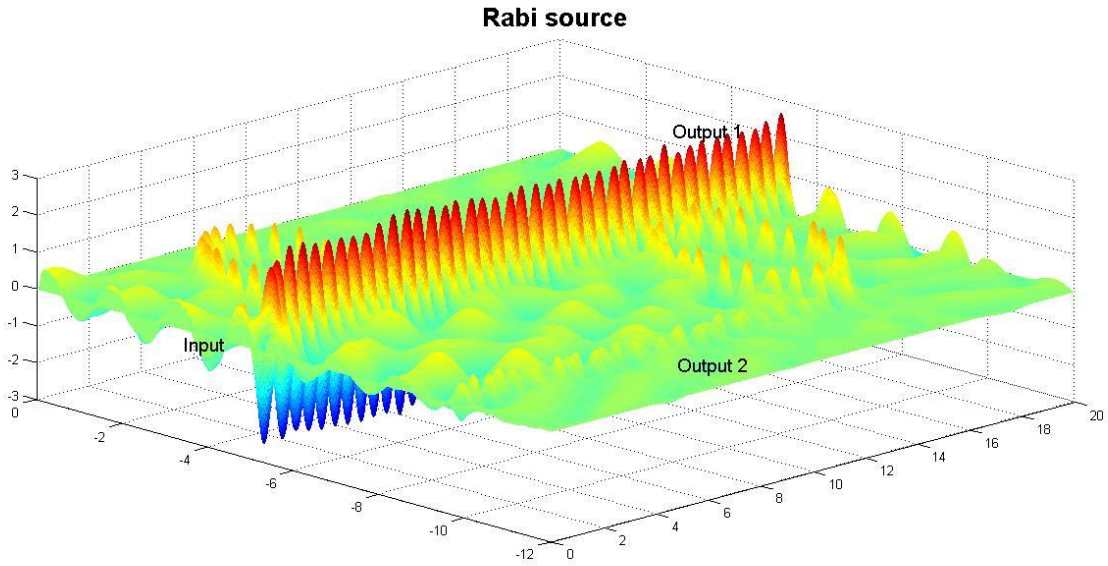


Figure 2. The oscillation is generated and the photon probe is obtained by using the MATLAB program

In simulation, Rabi frequency and time evaluation of the population of FMO complex were calculated by following equations. Then, input that electromagnetism frequency into the system at input port, the light probe is generated and accelerated within the system, which is shown in Fig. 2. The monitoring signals can be detected via the Through (Output 1) and Drop ports (Output 2), respectively. The change in phase of light (particle) can be introduced the change in device output intensities, which can be used to monitor and measure the required physical parameters, especially, within electronic states, where the link parameters can be seen and interpreted via the drop and through ports. Fig. 2 shows the Rabi probability oscillation result, which is obtained by MATLAB program.

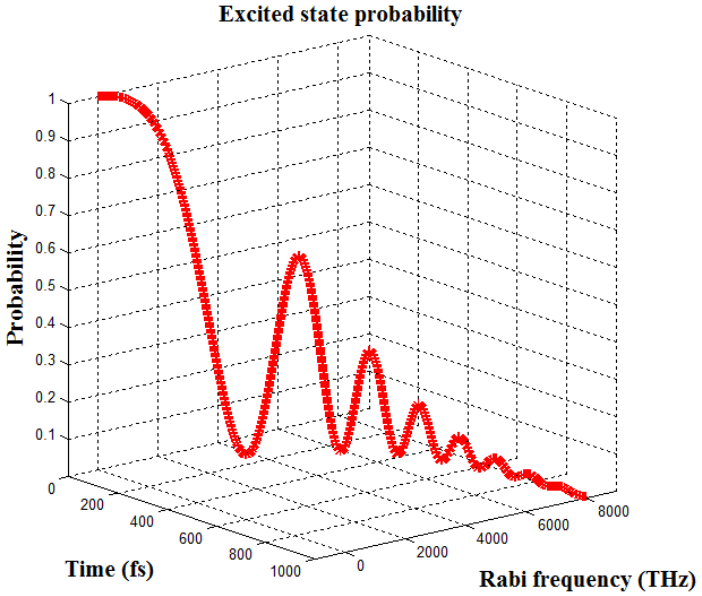


Figure 3. Rabi oscillation's probability result at Drop port signal with frequency in THz region.

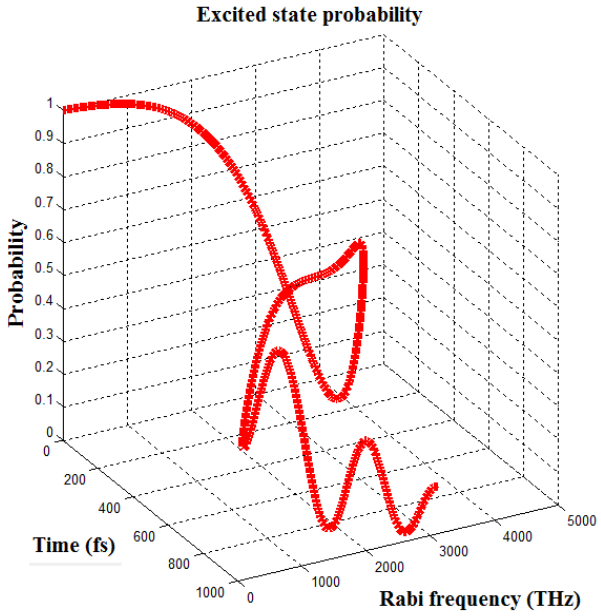


Figure 4. Rabi oscillation's probability result at Through port signal with frequency in THz region.

Fig. 3 and 4 show the Rabi oscillation's probability related with time evolution of population in excited state of FMO complex and Rabi frequency at THz scale. The change of Rabi frequencies in the THz scale can be configured the site energy states different for advantage to observe the behaviors of FMO complex or its disorder. Fig. 5 shows time evolution of population in excited state of FMO complex's scale is equivalent to the phonon relaxation time, estimate by [13,24], drop port is find result with the following references. Some populations are equal 0, we can interpreted that the FMO complex is in the local state, S_0, then it exciting again until the receive energy from photon had been depleted. Fig. 6 shows the linear relationship of the signals can be obtained, which is suitable for sensor applications, where the lower input power can provide output linearity, alternatively, the nonlinear behaviors such as chaos, bifurcation and bistability can be occurred. The results are obtained the bistable characteristics and probability density to finding atom in the ring resonator. The transmission intensity ratio between output port and input port with difference coupling coefficients are shown the bistable characteristics at throughput port and drop port as well.

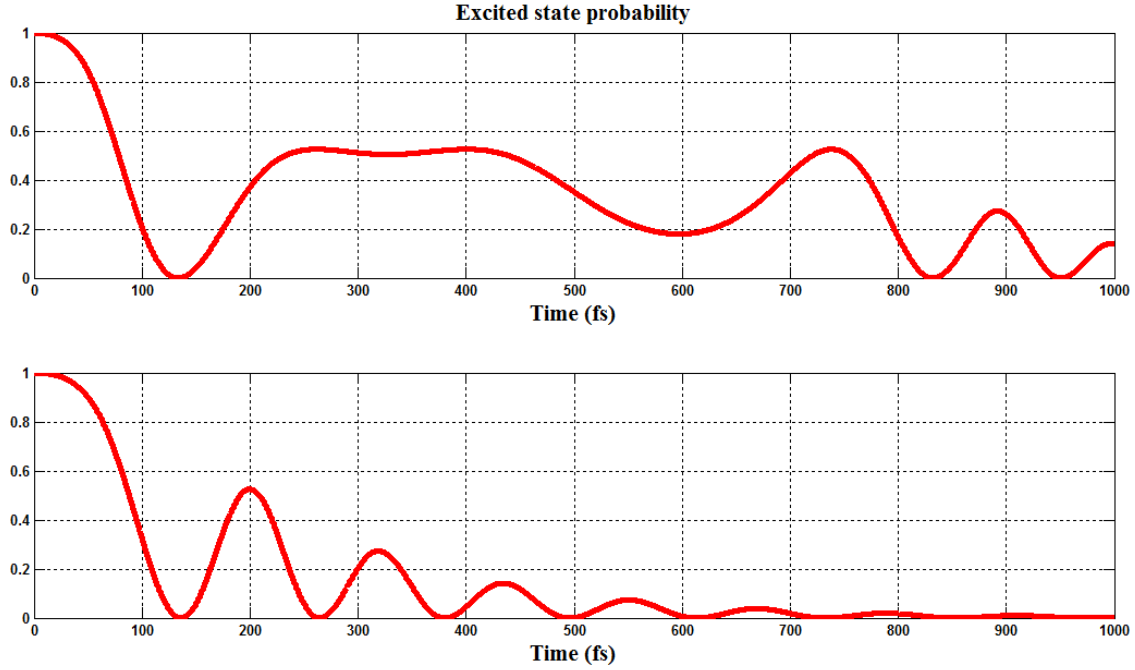


Figure 5. Time evolution of the population in excited state of FMO complex, where top and bottom side are the signal from Through port and Drop port, respectively.

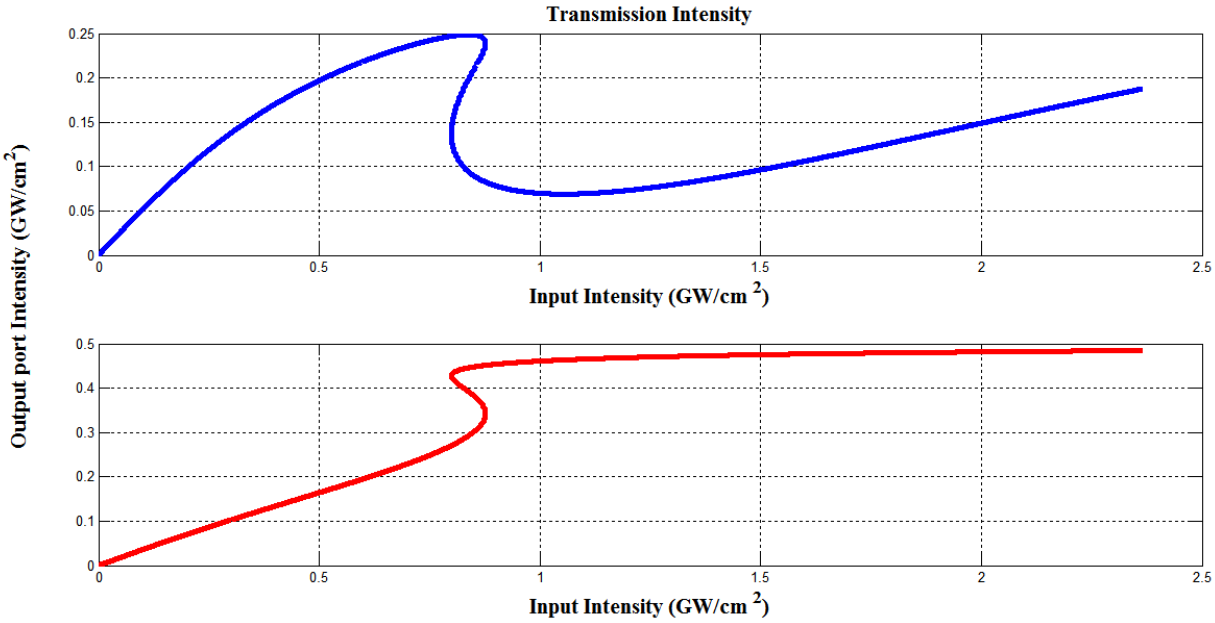


Figure 6. The bistable properties with coupling coefficient is equal to 0.2 that shows the photon nonlinearity behavior.

Conclusion

I have proposed the techniques of detection probe vibration energy relaxation of the light harvesting complex II. The EET dynamic is estimated by assuming that the effect on FMO complex has only coulomb potential between the molecules, the phonon effect had been vanished here. The exciton dynamic was setup from the Heisenberg equation and Rabi frequency also calculated, two-level system $S_0 \rightarrow S_1$, with the time evolution. The following frequencies are the source of micro-ring resonator. The results show us the time evolution of population in excited state of FMO complex are decreasing along the time, both of output port. Therefore, drop port's signal is agreed with the experimental results, the time evolution of population in the THz scale, which can be useful for required information for photosynthetic pigment complex II investigation and related applications.

Acknowledgment

I would also like to give the acknowledgment for the laboratory and research facilities from the King Mongkut's Institute of Technology Ladkrabang (**KMITL**), Bangkok 10520, Thailand.

References

[1] Hall, D. O. and Rao, K.K., Photosynthesis, 6th edn, Cambridge, UK: Cambridge University Press, (1999).
[2] Blankenship. R. E., Molecular mechanisms of photosynthesis, Oxford, UK: Blackwell Science, (2002).
[3] Fassioli F et al, Photosynthetic light harvesting: excitons and coherence, J. R. Soc. Interface, Vol 11: 20130901, (2013).
[4] Lim J et al, Vibronic origin of long-lived coherence in an artificial molecular light harvesting, Nature Communications, Vol 6: 7755, (2015).
[5] Ranger T et al, How the molecular structure determines the flow of excitation energy in plant light-harvesting complex II, Journal of Plant Physiology, Vol 168, (2011) 1497-1509.
[6] Tessa R. Calhoun and Graham R. Fleming, Quantum coherence in photosynthetic complexes, Phy. Status Solidi B, Vol 248, No.4 (2011), 833-838.
[7] Jahan M. Dawlaty et al, Microscopic quantum coherence in a photosynthetic light harvesting antenna, Phil. Trans. R. Soc. A, Vol 370, (2012), 3672-3691.
[8] Duan H G et al, Two-dimensional electronic spectroscopy of light-harvesting complex II at ambient temperature: a joint experimental and theoretical study, J. Phys. Chem. B, (2015), 119, 12017.
[9] Lewis N H et al, A method for the direct measurement of electronic site populations in a molecular aggregate using two-dimensional electronic vibrational spectroscopy, J. Phys. Chem. B, (2015) 143, 124203.
[10] Oliver T A, Lewis N H and Fleming G R, Correlating the motion of electrons and nuclei with two-dimensional electronic vibrational spectroscopy, Proc. Natl. Acad. Sci. U.S.A, (2014), 111, 10061-10066.
[11] Dong H et al, Determining the static electronic and vibrational energy correlations via two-dimensional electronic vibrational spectroscopy, J. Chem. Phys, (2015), 142, 174201.
[12] Lewis N H et al, Measuring correlated electronic and vibrational spectral dynamics using line shapes in two-dimensional electronic vibrational spectroscopy, J. Chem. Phys, (2015), 142, 174202.
[13] Ishizaki A and Fleming G R, Theoretical examination of quantum coherence in a photosynthetic system at physiological temperature, PNAS, Vol 106, No. 41, (2009), 17255-17260.
[14] T A Ibrahim, R Grover, L C Kuo, S Kanakaraju, L C Calhoun, and P T Ho, All-optical AND/NAND logic gates using semiconductor Microresonators, IEEE Photonics Technology Letters, (2003), Vol. 15 1422–1424.
[15] C Y Chao, W Fung, and L Jay Guo, Polymer microring resonators for biochemical sensing applications, IEEE Journal of Selected Topics in Quantum Electronics, (2006), Vol. 12 134–142.
[16] M Forst, J Niehusmann, T Plotzing, J Bolten, T Wahlbrink, C Moormann, and H Kurz, High-speed all-optical switching in ionimplanted silicon-on-insulator microring resonators, Optics Letters, (2007), Vol. 32 2046–2048.
[17] Yupapin P and N Pornsuwancharoen, Proposed nonlinear microring resonator arrangement for stopping and storing light, IEEE Photonics Technology Letters, (2009) , Vol.21, 404-406.
[18] Thammawongsa N, S Tunsiri, M A Jalil, J Ali and P Yupapin, Storing and harvesting atoms/molecules On-Chip: Challenges and Applications, J. Biosensors & Bioelectronics, (2012), Vol.3 14-115.
[19] Zainol F D, N Thammawongsa, S Mitatha, J Ali, P Yupapin, Nerve communication model by bio-cells and optical dipole coupling effects, Artificial Cells, Nanomedicine, and Biotechnology, (2013), 1-8.
[20] Sarapat N, K. Kulsirirat and P Yupapin, Tissue culture with 3D monitoring by distributed ring circuits, Journal of Bisensors and Bioelectronics, (2013), Vol. 4(2), 9-22.
[21] Khunnam W, N Yongram, N Sarapat, P Yupapin, Quantum matter generated by trapped particles, ScienceJet, (2013), Vol 2, 37-39.
[22] Yupapin P, Pantian S and Ali J, Novel design Rabi oscillation system for human quantum life detection probe, Life Sci J, (2014), 11(2) 235-243.
[23] Zyga L, Study supports role of quantum effects in photosynthesis, The Journal of Physical Chemistry Letters, (2012), Vol.3, 272-277.
[24] Rozzi C A et al, Quantum coherence controls the charge separation in a prototypical artificial light harvesting system, Nature communication, (2013), 4:1602.

The Comparison of the Alternative Fuel Properties at Low Temperatures

Somjai Peanprasit[1,a], Nataporn Chindaprasert[1,b], Chonlakarn Wongkhorsub[2,c]
Padet Sankasem[3,d] and Padipan Tinprabath[1,e,*]

[1]Department of Mechanical Engineering, Faculty of Engineering, Rajamangala University of Technology Phra Nakhon, Bangkok 10800, Thailand

[2]College of Industrial Technology, King Mongkut's University of Technology North Bangkok, Bangkok 10800, Thailand

[3]Mahachai Institute of Automotive Technology, Samutsakhon 74000, Thailand

Keywords: Properties of fuel, diesel fuel, pyrolysis fuel, biodiesel fuel, low temperature

Abstract

This paper is studied about the characteristic of 3 kinds of fuel which are pyrolysis oil, Bio-diesel oil, and diesel oil in standard temperature and low temperature in order to study the basic characters of the oils before using in the vehicle. The characteristics of the oil have to follow the emission standard of the car (EURO 6) which released on 2016. In the EURO 6 standard, the emission test has to be done at the temperature -7^{o}C. Therefore, the temperature -7^{o}C is an interesting point to study the characteristic of oils as it is an essential function to predict the emission of the vehicle. The pyrolysis oil and the biodiesel oil are concerned as the alternative energy which is potentially subsidiary oil for diesel engine and they made for recycle material. The tested oils, which are pyrolysis oil, Bio-diesel oil, and diesel oil, are tested by tester ASTM D341 standard. The machine can be tested under the temperature condition -56 till 105 ^{o}C . the result shown that the density of the oil is increasing normally. However, the viscosity of the diesel and bio-dielsel are palabplically increased whereas the reducing of temperature is unaffected to the pyrolysis oil which is relevant to Riazi and Tinprabah's equation to predict characteristic of oil.

1.Introduction

Nowaday, the biodiesel and other alternative oil are mix with diesel 2-20% to reduce using petroleum [1,2]. As this reason, the bio material and waste material are used to produce the oil as they can increase energy reliability, environmental friendly and greenhouse gas reduction. This bio-oil is normally has high cetane number but low sulfur and aromatics[3]. The disadvantage of the biodiesel is high viscosity, high cloud point, high pour point but low heating value. the used in engine are produce high NOx which leads low engine power and high cost [1]. Therefore, it is necessary to study fuel characteristics which are; density, viscosity at the point that low temperature effects the emission occurs in engine especially at the starting period of the engine. The European Union has established the emission standards in 2016 which called Euro VI [4]. This standard will be the latest standard applied to cover the starting engine at low temperature problem which are advance spraying characteristics and emission at the low temperature [4]. At temperature-7^{o}C, the viscosity of the oil is increased rapidly[4]. Applying bio-diesel or pyrolysis oil 5% blended is unaffected to the spray nozzle of the engine [4,5]. The emission is tended to behave follow the Euro standard. However, it is essential to study the characteristics of bio-diesel and pyrolysis oil in terms of physics fuel properties at low and high temperature [4,5,6,7] which influence the flow and spraying of fuel in nozzle head include the prediction of spray characteristics. The spray length, spray angle and fuel atomization are the spray character that influence the engine performance and emission from engine. The objective

of the research is to study the characteristic of the fuel at the temperature -7°C regarding the EURO 6 standard. The research are applied with pyrolysis oil, diesel oil, and bio-diesel oil from canola oil at the testing temperature condition -8°C to 40°C. The concerned characteristics are density and viscosity.

2.Theory

2.1 Density of fuels

The density of the fuel at ambient temperature and low temperature can be analyzed by the equation of Riazi [8] as shown in equation (1).

$$\rho_T = 0.99.SG\text{-}10^{-3}.(2.34\text{-}1.898SG).(T\text{-}288.7) \quad (1)$$

Whereas; SG is specific gravity and T is temperature (K)

2.2 Viscosity of fuels

The viscosity of the fuel at the temperature range -4 °C to 100°C can be predicted by following Riazi's equation [8]

$$Log_{10}(v_T) = A.(/311T)^B.T - a \quad (2)$$
$$A = Log_{10}(v_{(T=311)}) + a \quad (3)$$
$$B = b.\ Log_{10}(v_{(T=311)}) + c \quad (4)$$

Whereas, T is temperature (K) $v_{(T=311)}$ is kinetic viscosity at 311 K or 38°C, and a = 0.8696, b = 0.2801and c = 1.8616

For temperature 4-°C to -10°C can be calculated by following Riazi's equation [8] and developed by Tinprabath , et al [4] as shown;

$$Log_{10}(v_T) = A.(269/T)^B.T - a \quad (5)$$
$$A = Log_{10}(v_{(T=269)}) + a \quad (6)$$
$$B = b.\ Log_{10}(v_{(T=269)}) + c \quad (7)$$

Whereas, T is temperature (K), $v_{(T=269)}$ is kinetic viscosity at 269 K or -4°C, a = 0.8639-, b = 87.656-and c = 124.30

3.Experimental apparatus

3.1 Viscosmeter

The testing apparatus for fuel characteristics is Anton Paar@ Stabinger Viscosmeter (model SVM 3000/G2) of Laboratoire PRISME, from Orleans University, France. The apparatus is able to measure both density and viscosity by using ASTM D341 testing standard, the temperature can be measured from -56 to 105°C, and the accuracy of the density is +/- 0.35%

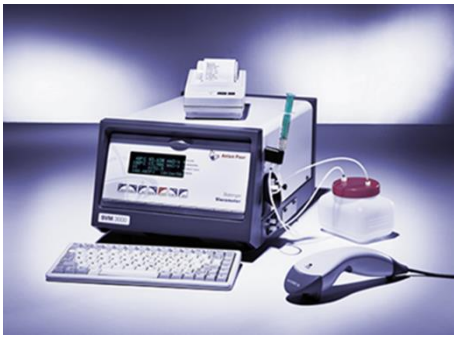


Fig.1 Anton Paar@ Stabinger Viscosmeter (model SVM 3000/G2) [9]

3.2 Fuels

The research are applied with pyrolysis oil, diesel oil, and bio-diesel oil from canola oil at the testing temperature condition -8°C to 40 °C.

Fig.2 Experimental test fuels

3. Results and discussion

3.1 Density

The testing result of the density is shown in figure 3. It is found that the density of the fuel is decreased when the temperature of the fuel increased; on the other hand, the density is increased when the temperature is decreased. The bio-diesel fuel is the highest density while the pyrolysis is the lowest density. The diesel oil behave in between the two fuels. The figure shown that the densities of three fuel are increased while the temperature is decreased related to the equation (1) in every types of fuel at the 0.3% accuracy.

3.2 Viscosity

The testing result shown in figure 4 is the relation of the temperature and the kinetic viscosity of the fuel. It is found that all the experimented fuel at this range of temperature behave similarly which the viscosities of the fuel are decreased when the temperature is increased whereas, the viscosities are increased while the temperature is decreased. Especially at the temperature -4^{o}C, the viscosity of diesel and bio-diesel are dramatically increased whereas the increasing rate of the viscosity for pyrolysis oil is in normal rate. It could be count as the advantage character of the pyrolysis oil at the low temperature. At this point the behavior of all the fuel can be predicted by the equation (2) and (5) with 3% accuracy.

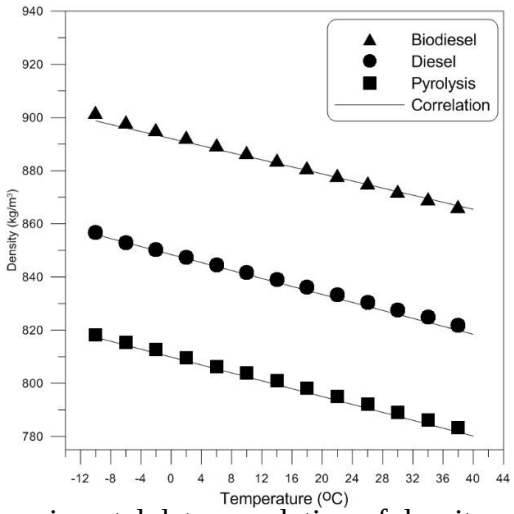


Fig.3 Experimental data correlation of density

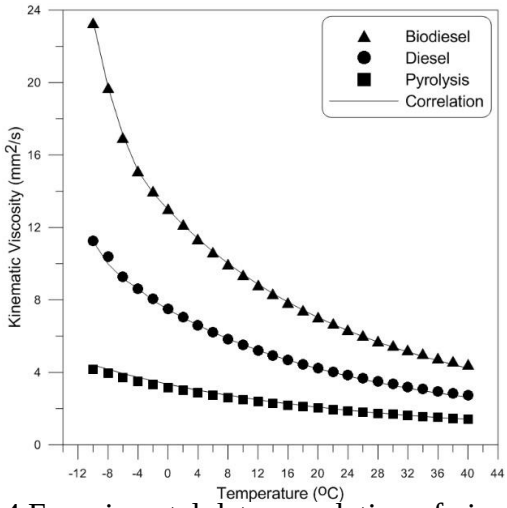


Fig.4 Experimental data correlation of viscosity

Table 1 Fuel matrix in standard conditions

	Pirolysis		**Biodiesel (B100)**		**Diesel**	
T, ^{O}C	Density (kg/m^3)	Viscosity (mm^2/s)	Density (kg/m^3)	Viscosity (mm^2/s)	Density (kg/m^3)	Viscosity (mm^2/s)
-10	818.2	4.16	901.90	23.31	856.80	11.27
-8	816.8	3.95	900.15	19.72	855.05	10.38
-6	815.4	3.71	898.40	16.97	853.00	9.28
-4	814.1	3.52	897.00	15.12	851.60	8.63
-2	812.7	3.33	895.50	14.00	850.20	8.04
0	811.1	3.17	894.10	13.03	848.80	7.51
2	809.6	3.01	892.60	12.15	847.40	7.04
4	808.3	2.87	891.20	11.35	845.95	6.60
6	806.3	2.74	889.75	10.63	844.50	6.21
8	805.4	2.62	888.30	9.97	843.20	5.85
10	803.8	2.50	886.85	9.38	841.70	5.52
12	802.5	2.39	885.40	8.83	840.30	5.22
14	801	2.29	883.95	8.33	838.90	4.94
16	799.4	2.20	882.50	7.87	837.50	4.68
18	798.1	2.11	881.05	7.44	836.10	4.44
20	796.5	2.03	879.60	7.05	834.70	4.23
22	795	1.95	878.15	6.69	833.30	4.02
24	793.4	1.88	876.75	6.36	831.90	3.84
26	792.2	1.81	875.30	6.05	830.45	3.66
28	791.1	1.73	873.85	5.74	829.10	3.50
30	789	1.68	872.35	5.50	827.65	3.35
32	787.8	1.62	870.95	5.23	826.30	3.21
34	786.2	1.56	869.45	5.02	824.90	3.07
36	784.8	1.51	868.05	4.80	823.50	2.95
38	783.3	1.46	866.50	4.60	821.85	2.83
40	781.5	1.42	865.00	4.44	820.50	2.74

3.3 Relative density and viscosity

The testing results are shown in figure 5 and table 1. The correlation of density and viscosity plotted as parabolic shape which shown that the density and viscosity tend to increase while the temperature decrease, the trend is dramatically shown in diesel and bio-diesel which the pyrolysis oil behave generally. The reason of this issue could related with the pour point and the CFPP of the diesel and bio-diesel which are lower than pyrolysis oil.

4. Conclusions

The study of characteristic of pyrolysis oil, diesel and bio-diesel oil at the room temperature and low temperature is shown that the decrease of the temperature is unaffected with the density of the fuel but affected with the kinetic viscosity of the diesel and bio-diesel. The kinetic viscosity property of the pyrolysis oil behaves ordinarily with the low temperature. Therefore, it can concerned that the pyrolysis oil can be blended with diesel or bio-diesel oil in order to apply in vehicle in the future.

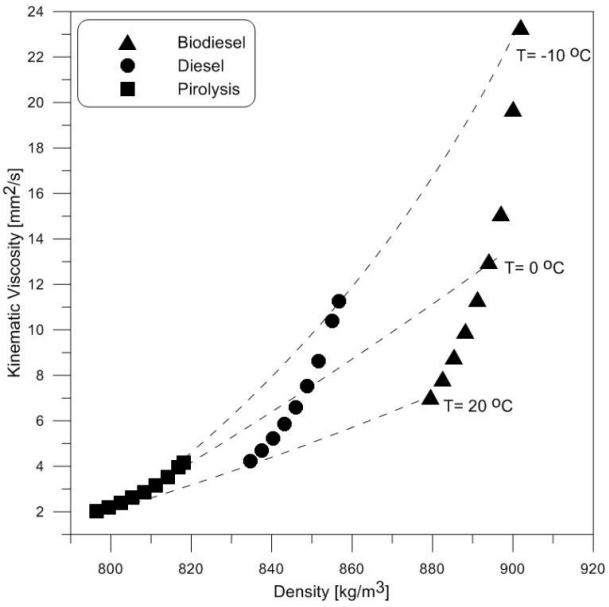


Fig.5 Experimental data correlation of viscosity

Acknowledgements

This work was cofinanced by Rajamangala University of Technology Phra Nakhon (RMUTP), Thailand for financial support. The authors would like to thank you very much. The authors would like to thank you to Combustion Technology and Alternative Energy Research, King Mongkut's University of Technology North Bangkok for the source of Pyrolysis oil and Camille Hespel and Ob Nilaphai, Laboratoire PRISME, Orleans University, France for testing the fuel characteristic.

References

[1] J. K. Author, No S.Y., How vegetable oils and their derivatives affect spray characteristics in CI engines - A review, Atomization and Sprays 2011;21(1),pp. 87-105.

[2] Perdiguero J., Jiménez JL., Sell or not sell biodiesel: Local competition and government measures, Renewable and Sustainable Energy Reviews 2011;15, pp. 1525-1532.

[3] Demirbas A., Progress and recent trends in biodiesel fuels, Energy Conversion and Management 2009;50, pp.14-34.

[4] Tinprabath P, Hespel C, Chanchaona S, Foucher F., Influence of biodiesel and diesel fuel blends on the injection rate under cold conditions, Fuel 2015;144, pp.80-89.

[5] Tinprabath P, Hespel C, Chanchaona S, Foucher F., Impact of cold conditions on diesel injection processes of biodiesel blends, Renewable Energy 2016 ;96 pp. 270-280.

[6] National Biodiesel Board. Low blends of biodiesel and cold weather operability,[cit.2014-03-20] <http://www.biodiesel.org>.

[7] Sarin A., Biodiesel production and propretes, Cambridge: RSC Publishing; 2012.

[8] Riazi M.R., Characterization and properties of petroleum fractions, ASTM manual series: MNL50, 2005.

[9] Anton Paar GmbH, Instruction manual SVM 3000 stabinger viscometer, Austria, 2005.

STRATEGIC GREENHOUSE GAS REDUCTION POLICY PLANNING COMPARING BIO-MASS AND CLEAN COAL POWER PLANT USING CARBON FOOTPRINT FOR ORGANIZATION (CFO) METHODOLOGY

Natworapol Rachsiriwatcharabul*,Suwat Paengteerasukkamai, Prapaphorn Ployyod

Department of Sustainable Industrial Management Engineering, Faculty of Engineering,
Rajamangala University of Technology Phra Nakhon, Bangkok 10800, Thailand

Keywords: greenhouse gas reduction**,** policy-planning**,** power plant

Abstract Intention of this article is to ilustrate the diferences of strategic policy plannings for greenhouse gas reduction comparing bio-mass and clean coal power plant using carbon footprint for organization (CFO) methodology. First, CFO was introduced and implemented at both type of power plants. Top five emission source of each power plant will be identified, ranked, and analyzed, Then greenhouse gas reduction policy will be conducted. Finally, result of this strategic policy planing of each power plant's type will then be compared. The final comparasion result will then be concluded. Benefits of this articale is to illustrate strategic policy planing using CFO methodology as well as final comclusion will support greenhouse gas reduction policy planning and implementating for pilot power plant themselves at micro level, and government at macro level

Introduction

The current global climate change trend has led many countries around the world, including Thailand, to prepare themselves for the mitigation and adaptation of global warming impact [1]

Before, Thailand was a country that was not being forced to reduce greenhouse gas emissions. However after 2020, all countries in the world will have to propose greenhouse gas emission reduction targets. Including Thailand will need to be prepared to reduce greenhouse gas emissions properly. To be ready to accept the agreement of the 21st Conference of the Parties (COP) meeting, which has sent each country's operational goals so called Intended Nationally Determined Contributions (INDCs). For Thailand, our prime minister plate to reduce greenhouse gas emissions by 20-25 percent between by 2030

Based on Thailand greenhouse gas emissions data from 2000 – 2012,a summary of the greenhouse gas emissions trend shows that total greenhouse gas emissions of Thailand, including removal sources has a magnitude between 169.81 - 227.73 Mt CO_2eq and when calculating the amount of greenhouse gas emissions excluding removal source from forestry and the land use during the past 12 years, Thailand has a greenhouse gas (GHG) emission value between 257.63 - 350.68 Mt CO_2eq. Based on sectoral rankings of greenhouse gas emissions, the first is energy sector, secondly, agriculture, forestry and land use (AFOLU), industrial process and product use (IPPU) and waste management respectively. [2]

Since energy sector is the most emitting of greenhouse gases compared to others, not only in Thailand but the world total, strategic greenhouse gas reduction policy planning has to focus on the

energy sector. Especially when most of the energy used in the world comes from fossil fuels (coal, oil and natural gas), which are main contribution to global warming. As a result, both public and private entities have focused on alternative energy production. Biomass power plant is a type of biomass power plant that uses waste from biomass fuels such as residues from agricultural. Theses residue from processed agricultural products are used as a fuel for electricity and steam generation, More over biomass industries is also help in enhancing economics status quo for local people. In addition, energy production from biomass fuels with appropriate technology is polluted and created less greenhouse gas conditions.[3] At present, biomass power generation is playing an increasing role in the industrial sector.

However, the use of renewable energy and reduction of fossil fuel consumption is still a problem, and the barriers to the combustion of fuels and raw materials used in generating electricity are not maximized. It also affects the management costs and emissions to the atmosphere. The purpose of this research is to assess the amount of greenhouse gas emissions from the biomass power generation sector from the power plants. Two types of power plants; coal and biomass is used to collect primary data in the study. The results is to compare greenhouse gas reduction policy of this two type of power plant in which will lead us to proper planning for global warming mitigation.

Research Methodology

Assessment of greenhouse gas emissions using Carbon Footprint for Organization (CFO) of Thailand Greenhouse gas Organization (TGO) is conducted. Two case studies, including coal power plants and biomass power plants is used in this study. The first part evaluates Direct Emissions from various activities including the use of fuel for combustion of machinery, fuel for corporate vehicles, use of raw materials and chemicals in wastewater treatment leakage of refrigerant from air conditioning system including methane emissions from the used of septic tank is calculated and the second part assesses indirect greenhouse gas emissions from energy consumption, so called indirect emissions from energy used.. The process of calculating the carbon footprint of an organization consists of 6 steps, as shown in Figure 1.

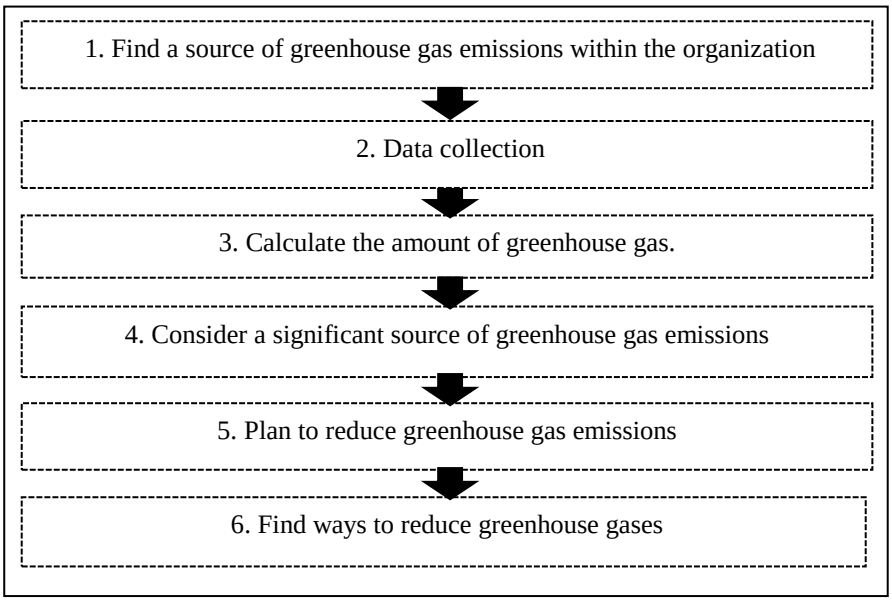


Figure 1. 6 Steps to Carbon Footprint Assessment [4]

Data collection

Carbon Footprint for Organization (CFO) calculated from coal and biomass power plants has been analyzed. These primary data collection from case studies is collected from January to December 2016 for a period of 12 months. Various types of fuel consumption data in the production process and within the organization including wastewater treatment, information amount of refrigerant in air conditioner system, including the greenhouse gas emission factor used to calculate carbon footprint for organization. This can be detailed in Table 1 and Table 2.

Tables 1 Shows fuel consumption raw materials and resources in coal power plants, for a period of 12 months, and emission factors for calculation.

No.	Source of Greenhouse gas emissions	Unit	Lifecycle Inventory (LCI)
1	Fuel Oil Consumption (Diesel Burner Boiler)	Litr	941,200.08
2	Coal Consumption (Burner Boiler)	Kg	4,001,335,906.01
3	Diesel Fire pump	Litr	900.00
4	LPG Canteen	Kg	3,360.00
5	Diesel vehicle	Litr	297,925.86
6	Gasoline vehicle	Litr	25,436.91
7	Sewage Waste	kg	2,635.29
8	Waste Water	kg	8,445.69
9	Carbon dioxide (CO_2 purging)	Kg	400.00
10	Methane emissions from employee toilets	Kg	8.76
11	Fire extinguishers	Kg	4.54
12	Refrigerants R 134A	Kg	0.09
13	Refrigerants R 410 A	Kg	2.50
14	CO_2eq emissions in the Degas process for the production of Demin water	Kg	88.12
15	Acetylene	Kg	243.69
16	CH4 emissions from gas to coal.	Kg	848,209.95
17	Electricity	Kwh	4,815,501.00

Tables 2 Shows fuel consumption raw materials and resources in biomass power plants, for a period of 12 months, and emission factors for calculation.

No.	Source of Greenhouse gas emissions	Unit	Lifecycle Inventory (LCI)
1	Diesel generator	Litr	400.00
2	LPG for Maintenances	Kg	864.00
3	Diesel Burner Boiler	Litr	7,405.00
4	Diesel backhoe loader	Litr	18,900.00
5	Methane emissions from employee toilets	Kg	424.86
6	Electricity	Kwh	85,870.00

Table 1 and Table 2 illustrates total amount of fuel used in the organization's activities. including raw materials, and other resources for coal-based and biomass-based fuels for a period of

1 2 months from January to December 2 0 1 6 . These two tables can be used to calculate carbon footprint of organizations to assess greenhouse gas emissions.. This study explores the extent of the organization boundary using operational control in accordance with the factory operation license documents. The total emission acceptance tolerance is defined as materiality in which in this case is set to a rate of 5 percent.

Evaluate for the greenhouse gas emissions

How to assess greenhouse gas emissions by calculating carbon footprint. Case study of coal power plants and fuel power plants. Biomass refers to the assessment model in accordance with the organizational guidelines of the Greenhouse Gas Management Organization The Greenhouse Gas Emission Factor (GHG) is the secondary data from the organization's Carbon Footprint Assessment Schedule, Volume 4, Issue 2 April 2015, as shown in Table 3, and the amount of emissions. Greenhouse gas in tons of carbon dioxide equivalent is equivalent to the following greenhouse gas calculation formula.

$$\text{GHG emissions} = \Sigma (\text{Activity}_i \times \text{GHG EF}_i). \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(1)$$

GHG emissions = Greenhouse gas emissions
Activity_i = Activity data
GHG EF_i = Greenhouse gas emission factor$_i$

Tables 3 Emission factors for calculation

No.	Source of Greenhouse gas emissions	Emission Factor (CO_2Eq)	Emission Factor Reference
1	Diesel (Stationary combustion)	2.7080	IPCC Vol.2 table 2.2, DEDE
2	Coal	2.2855	Calculated the heating value of the coal used.
3	LPG (Stationary combustion)	3.1133	IPCC Vol.2 table 2.2, DEDE
4	Diesel (Mobile combustion)	2.7446	IPCC Vol.2 table 2.2, DEDE
5	Gasoline (Mobile combustion)	2.2376	IPCC Vol.2 table 2.2, DEDE
6	Carbon dioxide	1.0000	Thailand greenhouse gas organization guide book
7	Methane emissions	25.0000	Thailand greenhouse gas organization guide book
8	Refrigerants R 134A	1,430.0000	(R134 A) List of refrigerants
9	Refrigerants R 410 A	2,088.0000	(R410 A) List of refrigerants
10	Acetylene	2.2804	Thailand greenhouse gas organization guide book
11	Electricity	0.5813	Thailand greenhouse gas organization guide book

Research result

Based on carbon footprint for organization assessment of coal power plants and biomass power plants, taking into account the data collected over the 12 months period of each types of power plant, the results of the greenhouse gas emissions of the coal power plants, as shown in Table 4, and biomass power plant is shown in Table 5.

Tables 4 Greenhouse gas emission assessment results from coal power plants case study

No.	Source of Greenhouse gas emissions	Greenhouse gas emissions ($tonCO_2eq$)	Ratio of Greenhouse gas emissions (%)
1	Fuel Oil Consumption (Diesel Burner Boiler)	2,548.74	0.03
2	Coal Consumption (Burner Boiler)	9,145,176.77	99.69
3	Diesel Fire pump	2.44	0.00
4	LPG Canteen	10.46	0.00
5	Diesel vehicle	817.69	0.01
6	Gasoline vehicle	56.92	0.00
7	Sewage Waste	65.88	0.00
8	Waste Water	211.14	0.00
9	Carbon dioxide (CO_2 purging)	0.40	0.00
10	Methane emissions from employee toilets	0.22	0.00
11	Fire extinguishers	0.00	0.00
12	Refrigerants R 134A	0.12	0.00
13	Refrigerants R 410 A	5.22	0.00
14	CO_2 emissions in the Degas process for the production of Demin water	0.09	0.00
15	Acetylene	0.56	0.00
16	CH4 emissions from gas to coal.	21,205.25	0.23
17	Electricity	2,799.25	0.03
	Total	**9,173,718,833.72**	**100.00**

Based on the assessment of greenhouse gas emissions of coal power plants, Table 4 shows that direct and indirect emissions of greenhouse gases Has an equal sum 9,173,718,833.72 $tonCO_2eq$. The major sources of greenhouse gas emissions from coal power plants, which are the main raw materials used in production, are 9,145,176.77 $tonCO_2eq$, accounting for 99.69 percent of all greenhouse gas emissions.

Tables 5 Greenhouse gas emission assessment results from biomass power plants case study

No.	Source of Greenhouse gas emissions	Greenhouse gas emissions ($tonCO_2eq$)	Ratio of Greenhouse gas emissions (%)
1	Diesel generator	1.08	0.80
2	LPG for Maintenances	2.69	1.97
3	Diesel Burner Boiler	20.05	14.72

No.	Source of Greenhouse gas emissions	Greenhouse gas emissions ($tonCO_2eq$)	Ratio of Greenhouse gas emissions (%)
4	Diesel backhoe loader	51.87	38.08
5	Methane emissions from employee toilets	10.62	7.80
6	Electricity	49.92	36.64
	Total	**136.24**	**100.00**

From Table 5, greenhouse gas emissions from power plants, biomass Greenhouse gas emission totals 136.24 ton CO_2eq. The major sources of greenhouse gas emissions for this type of power plant are from The use of diesel fuel in the incinerator is 20.05 ton CO_2 eq, representing a greenhouse gas emission factor of 20.05 percent, and the use of diesel in logistic and biomass transfer systems is equal. 51.87 tons CO_2eq accounted for 38.08 percent.

Discussions

Based on the assessment results of greenhouse gas emissions of the coal power plants, 99.69% is derived from coal as raw material in the power generation process. Considering the way to improve these types of greenhouse gas emissions, we have to consider the most important sources of emissions. That is, coal raw materials used in the power generation process. Executives of the power plant will need to provide a supplier of raw materials that can deliver higher value thermal coal. More effective This will reduce the amount of coal used in the production process. The amount of electricity produced increased. Production costs decrease The study was conducted in the case of a coal power plant case study. In case of finding a way to reduce the amount of coal using 15% by the time of collection of coal power plant data, this case can produce all the electricity. 11,298,024.60 MW / hour From this greenhouse gas reduction approach. The total amount of greenhouse gases can be predicted as shown in Table 6.

Tables 6 Compare the effect of reducing the Coal used in the process to reduce greenhouse gas emissions.

Source of Greenhouse gas emissions	**Coal used in the process (Tons)**	**Productivity (Mwh)**	**Total greenhouse gas emissions (Tons CO_2eq)**	**Intensity (Tons CO_2eq /Mwh)**
Coal Consumption (Before)	4,001,335.91	11,298,024.60	9,172,900.83	0.81190
Coal Consumption (After)	3,401,135.52		7,801,942.32	0.69056

From Table 6, it was found that the improvement of the coal power generation process by reducing the amount of coal by 15% would result in a reduction in the overall greenhouse gas emissions of the power plant. The equivalent of 7,801,942.32 tons of carbon dioxide equivalent. It also affects the emission of greenhouse gas emissions per unit from the 0.81190 Tons. CO_2eq / Mwh to 0.69056 Tons CO_2eq / Mwh or decrease to 43.93 percent.

From Table 5, the researchers have proposed reduction of greenhouse gas emissions of power plants, biomass fuels By considering the reduction of diesel fuel use in the fueling point of the production process and the amount of diesel fuel used in the conveying and moving of biomass fuels, it is reduced by 15%. The greenhouse gas emissions before and after are shown in Table 7.

Tables 7 Compare the effect of reducing the Diesel used in the burner boiler and backhoe loader to reduce greenhouse gas emissions.

Source of Greenhouse gas emissions	Volume in the process (Tons)	Productivity (Mwh)	Total greenhouse gas emissions (Tons CO_2eq)	Intensity (Tons CO_2eq /Mwh)
Diesel burner boiler (before)	7.40	322,824.91	136.23	0.00042
Diesel backhoe loader (before)	18.90			
Diesel burner boiler (after)	6.29		125.45	0.00039
Diesel backhoe loader (after)	16.06			

Table 7 found ways to reduce the amount of fuel as a source of greenhouse gas emissions, mainly consisting of Diesel burner boiler and Diesel backhoe loader for less. In 15 percent can reduce greenhouse gas emissions from energy production. electricity from biomass is equal to 136.23 Tons CO_2eq fell 125.45 Tons CO_2eq affect the rate of greenhouse gas emissions per unit of electric power, bio-fuels. The rest of the 0.0039 Tons CO_2eq / Mwh or decrease in the ratio of 7.15 percent.

When the plant produces electricity from coal electric power plants and biomass compared to the amount of greenhouse gas emissions before and after the improvements shown in Figure 2.

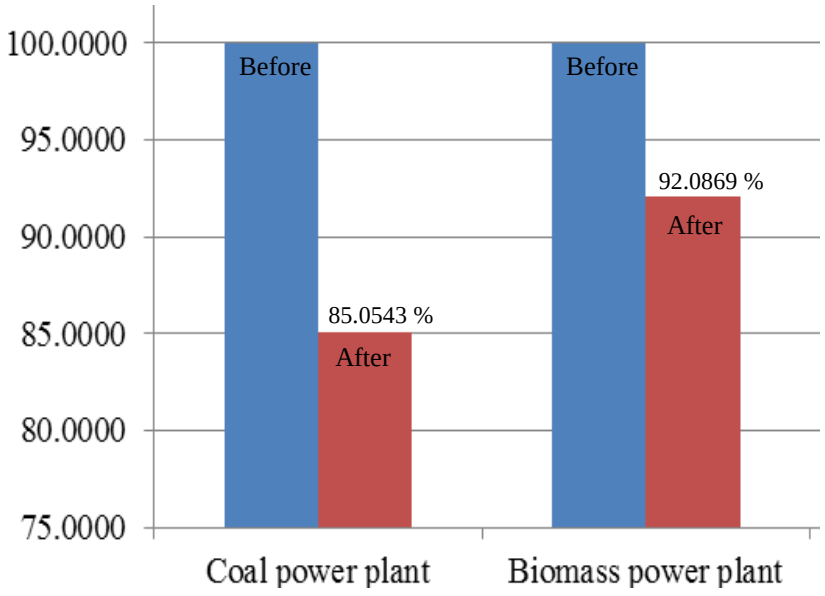


Figure 2 compares the percentages of greenhouse gas emissions of power plants.

Figure 2 shows a comparison of the percentage of greenhouse gas emissions from coal power plants and power plants, biomass fuels. The main activities are the most greenhouse gas emissions. Compared to the total ratio of activities within the organization. The percentage of greenhouse gas emission reductions was reduced to 85.0543 or a decrease of 14.9457, due to the decrease in the ratio resulting from the use of the most used coal in the production process. The net heat of coal used by the power plant. The potential for greenhouse gas emissions is quite high. As a result, the

greenhouse gas emissions from coal-based materials are as high as 99.69% . Therefore, the improvement approach by reducing the amount of coal used in the 15% ratio will significantly reduce the amount of greenhouse gas emissions. important

The emission of greenhouse gases from power plants, biomass fuels, uses the consideration of greenhouse gas reduction initiatives from the reduction of diesel fuel used in spotting, power generation, and diesel fuel used in logistics. and unloading biomass within the organization, the ratio fell to 15 percent can reduce the percentage of greenhouse gases. The remaining parts of 92.0869, representing a decrease of 7.9131.

Since the power plant case study of the two plants that are characterized by the use of raw materials and processing activities are different. The events are a source of greenhouse gas emissions are radically different. This is clearly seen. Biomass power plants, the emissions of greenhouse gases less. Major sources of greenhouse gas emissions do not come from the main raw materials used in energy production, as biomass fuels are depleted from agricultural residues. In the process of considering the acquisition of raw materials, it is classified as Biogenic. The potential value of greenhouse gas emission is zero. There is no greenhouse gas emissions. The amount of greenhouse gas emissions from the calculation of carbon footprint is not on the main raw materials like coal in a coal power plant. However, greenhouse gas emissions are being disseminated to other activities such as the combustion of various types of fuels, both in machinery and vehicles in the logistics system of the organization.

Summary

Based on the assessment of the greenhouse gas emissions of the power plants, two case studies are composed of coal power plants for power generation and biomass-based power plants using composite materials. Biogenic, such as sugarcane, corn, etc., with carbon footprint calculation tools. Lahore corporate guidelines for assessing greenhouse gas emissions of Greenhouse Gas Management Organization. (Public Organization) found that one coal power plant for a period of 12 months had a greenhouse gas emission equivalent of 9,172,900.83 tons of carbon dioxide equivalent. Considering the greenhouse gas reduction approach from the main activities, the greenhouse gas emission reduction could be reduced to 7,801,942.32 tons of carbon dioxide equivalent. And one greenhouse gas emissions from a biomass fuel plant are equivalent to 136.2383 tons of carbon dioxide equivalent, and considering greenhouse gas emission reduction from both diesel and machinery activities. logistics system Can be equal to 125.45 tonnes of carbon dioxide equivalent. According to the study, the difference between coal power plants and biomass fuels can be seen. The different activities. The event will be used for comparison to the same emission source is not clear. Including the number of power plants, this case study only two, so it is recommended to those who continue to study. There should be more cases in this case. And should be the same type of power plant, which will make the study clearly compare the source of greenhouse gas emissions from the main activities. And it can be used as a strategy for planning and proposing a policy to drive the plant to be environmentally sustainable in the future.

References

[1] Health Policy Foundation, Greenhouse Gas Emissions Reduction: Opportunities and Challenges, Thailand, 2014.

[2] Information on http://www. tgoweb.logicdevsite.com

[3] Information on http://www. Phattarapon-04.blogspot.com

[4] Information on http:// www.ftpi.or.th

THE STUDY AND DEVELOPMENT OF CONTEMPORARY PRODUCT DECORATION FOR REAL WOOD IDENTITY IN THAILAND ARCHITECTURE.

Kornpong Thongsri*, Saranyou Sawangmark, Noppadol Karwwisart, Rugiwan OunsongKarm, Parinan Bannchern

Faculty of Architecture and Design, Rajamangala University of Technology Phra Nakhon, Bangkok 10800, Thailand

Keywords: Decorative Products, Contemporary, Architecture Thailand

1. Introduction

1.1 The history and origin of the problem.

The rapid growth of economy, society and the cause of social change affect the way of living and well-being of people in the community. Society, culture, lifestyle, occupation, income and local wisdom have passed from ones to another generation for a long time. These change lifestyle of farming career, professional handicrafts, local architecture including with traditional wisdom, such the local wisdom of technician in community in Prachinburi. Woven rattan mat crafts has been unique and famous for ages in this province. Knowledge and wisdom in this area has been maintaining and carrying on for a long period. This matters crafts could be obliterated by future trends in technology development. Without education and priority conventional wisdom, it might be lost in the end. If the community can improve cognitive development, based on the act, we can develop and create new forms. We can also raise local products and local knowledge to give the importance. The development guideline initiative of His Majesty the King is the use of "understanding, access and development". This can be applied to all areas along HRM the King's principles.

"Understand" means understanding in life, tradition, social needs and problems. It can be understood, by discuss, advice and exchange information.

"Access" means a feeling of psychological problem, suffering, happiness, and local wisdom of community of people who live in the area by giving them accustom the idea, strongly confidence and faith.

"Development" means the improvement of quality of life, sentiment, well-being, career and working procedure.

Although, Thai ancient art is beautiful and fascinated, it is not necessary to repeated and imitated all things. Each generation of art depends on the age and environments. Today, it is different from those ancient times. The restoration of Thai antique in fine arts is to support the application and maintain national ancient pattern by using applied art which points to the usefulness, beauty, and recognizes to the depth and lively. Moreover, the development of Thai modern art has related with the west modern art for a long time in history in which philosophy, belief pattern, content and methodology and influence the variety process environment of Thai art. West art trends influences Thai art in case of being more freedom from Buddhism in the past. But local arts has not educational system, it consists with belief, live and usage. To make concept and emphasize knowledge are from the experiment and usage along with the environment of society such giving Thai value and wisdom knowledge.

Actually, social foundation or culture is the way of living and Thai wisdom. It is important content of culture. Local wisdom demonstrates knowledge and ability in solving a life problem of people in each society. The content of local wisdom is interested by both Thai and aboard. The way to support, maintain and recognize culture becomes an important awareness.

Generally, the purpose of carpentry architecture and woven rattan mat are to facilitate the way of living, ceremony, belief, tradition and religion. The regulation of use influences creative work because it influences other components such as material selection, shape, framework and pattern. Technology becomes important factor to response human needs in variety patterns such as a mat for sitting and sleeping, a bag, sandals, a file, and saucer. However, these are not several enough.

The study and design new product needs to maintain the some old patterns, forms and local wisdom. Therefore, research needs most are to study the definition of shape, feature, local uniqueness, inspiration, beauty, culture preservation and local wisdom.

1.2 The objective of the research

1) To study the procedure and concept of art on Thai carpentry and the feature of rattan mats weave to design contemporary furniture.
2) To study the quality and usefulness.
3) The study the satisfaction of contemporary Thai art furniture.

2. Study

The study is to design and develop home décor and contemporary carpentry products to identity the architecture and woven rattan mats in Prachinburi by using Thai contemporary art concept to support the beauty value and show the uniqueness and local wisdom.

1) To study information, survey data, process of carpentry and woven rattan mat pattern in Prachinburi.

2) To study furniture models, art concept of Thai contemporary.
3) To study the principle of visual art and beauty.
4) To design furniture, concept art of Thai contemporary.
5) To test the satisfaction of furniture.

The sample and population were 100 tourists in the village of Nong Ngong, Prachinburi.

3. Results and Discussion

This study found that rattan mat has woven for a long time in Prachinburi. From previous generation to the next generations, craftsmen has learnt and transferred their knowledge about rattan planting, choosing material from each places and the fine and soft of rattan textile. Their procedure of rattan weave is unique, the textile is sticky and tender. They learn about the place to plant such planting. If plant near the brackish water coast, the material will be soft, tender and suitable to weave. On the other hand, planting in other places, material is not as good enough as the previous inform.

The process of rattan weave consists of material preparation; harvest, dry and weave. The base pattern is block strips, and alternate line and color bring beautiful pattern. This community like to use natural color rattan to weave mat. Generally, the place of this weave is under the house or space area. 2 people are craftsmen, one sits at the back of the woven equipment and the other sits beside to put the rattan line. The first one has to notice the right position of the line and notice if rattan line is torn or turn over. The line and equipment has to be in the right position. The length of the mat depends on the length of each string.

The concept of carpentry and rattan mat weave inspired by natural shapes, patterns and the influence of natural environment such a lotus. Lotus is considered a high value flower, pure and special. And it is always used to pray. Its shape is curve. When we sketch this into Lai Thai, it looks gentle and warm.

There are 3 kinds of furniture design in this research which depends on the different uses and show feature concept of lotus shape and consists of pattern1-chaise (sofa), pattern2-single seat and pattern3- table decoration.

The analysis of the benefits of the use of the furniture style of carpentry and woven rattan mats was;

- Pattern 1 chaise (Sofa) an average test was 4.49, at a good level.
- Pattern 2 single chairs (Single Seat) an average test was test was 4.45, at a good level.
- Pattern 3 display table (Table decoration) an average test was 4.30, at a good level.

Satisfaction beauty of furniture shape, style of carpentry and woven rattan mats that conducted Thai culture as much as possible was;.

- Pattern 1 chaise (Sofa) an average test was 4.75, very good.
- Pattern 2 single chairs (Single Seat) an average test was 4.37, a good level.
- Pattern 3 display table (Table decoration) an average test was 4.45, at a good level.

4. Summary

The research of contemporary home decorative products-the carpentry architecture identification in the central region of Thailand was to develop and design furniture of Thai contemporary art. Also, this study benefits furniture design.

4.1 Research result

The study of contemporary home decorative products-the carpentry architecture and woven rattan mat identification in the region of Prachinburi, Thailand has a long history. It has directly accumulation by people in that society. Environment factors bring local wisdom, local culture and become rattan mat weave. Their weave processes have the uniqueness that plant rattan in their area. The quality of soil influences the quality of rattan-sticky and soft. The process of weave uses 2 craftsmen harmonize the length of the line, pattern, design and equipment along the area.

There are 3 patterns of weave consist of carpentry. And the concept design along with Thai art contemporary by using natural shape of lotus and shows Thai unique. Although, the heritage from previous generation to next ones such Lotus pattern that expresses the special flower to pray and show the purity, the same inspiration and concept in this study purpose for different usages.

The result found that; Pattern1-chaise (sofa), is the most suitable to design and develop furniture the surface material is not to smooth. It is good to design because a little bulge of rattan suitable for a back massage and feel relax for users. For the use, the standard size of matt matches the human body, it suitable for 2 people. Moreover, for the design, it can show Thai art contemporary because the shape of product curve is like lotus. It looks soft, warm and tender.

4.2 Suggestion

1) Base on the findings, the commercial aspect, this product is useful. But the cost is quite high. It should be considered. Survey prospect should be considered because the researchers produce costly product and require skillful craftsmen.
2) The present careers has changed rapidly cause the old craftsmen and entrepreneur’s problems, it should be supported in this career.
3) Government agencies should give priority to woven rattan mat career to promote product and distribution continuously.

5. Acknowledgments

This research was funded and support by expenditures budget of the year 2015 from the Faculty of Architecture and Design, Rajamangala University of Technology Phra Nakhon that point to the importance of doing research. Thank you to those who contributed and those who provide information and answer questions in this research.

6. Reference

N.n. Parknam. 1999. **Evolution of Art**. Bangkok: Publishing Love and Laundry Express.

Marichat Ourarenan. 2002. **Trends of Contemporary Art**. Bangkok: Chulalongkorn University Press.

Suchat Taotong. 2001. **Eastern culture and local wisdom**. London: Publisher Odeon Store.

Wirat Phitpiboon. 1984. **The contemporary design. Bangkok**: Chulalongkorn University Press.

Textiles and Clothing Sustainability

DESIGN AND DEVELOPMENT OF BANANA FIBER TEXTILES WITH GRAPHIC PRINTING AND DECORATION NANO INNOVATION TO THE ECONOMIC COMMUNITIES COMMERCIAL

Praparnporn Theeramongkol [1*], Bussara Sioyraya[2],
Autcha Hattayananont[3], Kingkarn Pijukkana[4], Darunrat Pigunthong[5]

[1,2,3] *Department of Fashion Clothing and Textile Design, Faculty of Home Economic Technology, Rajamangala University of Technology Phra Nakhon, Bangkok 10800, Thailand*

[4,5] *Faculty of Architecture and Design, Rajamangala University of Technology Phra Nakhon, Bangkok 10800, Thailand*

Abstract

This research is Design and development of banana fiber textiles with graphic printing and decoration Nano innovation to the Economic Communities Commercial. The results were found that Innovative nano-fiber cloth decorated banana fabric can be antibacterial at 99.95% and *staphylococcus* bacteria *klebsiella pneumonia* at 99.93% and water reflection / water level of 80 percent water, which can be absorbed into the cloth and wet it a bit. The mostly respondents were All have been satisfied at maximum average and highest level of satisfaction that with an average 4.91 percent to 98.24 satisfaction is at the highest level. Execution Technology, The workshop schedule for the two days workshop on 17 - 18 September 2559 at the Chedi district office in Suphanburi province, 25 people. Most of the participants were satisfied with the level, in Graphics and decorative nanotechnology innovation, most every question.

Keywords: Products Design Graphic Printing Fabric From Banana Nano Innovation Communities Commercial

1. preface

Project Research, Design and development of banana fiber textiles with graphic printing and decoration Nano innovation to the Economic Communities Commercial. Researchers have the intention of bringing the fabric from this banana fiber into the design and development of a product that is more special than the typical design. Modern graphic designs are printed with specialized printers in a modern graphic format. By designing and developing graphic styles to be current fashionable. To promote the image and make the appearance of the natural fibers to be more attractive colors. This is a great way to promote your products to a wide range of buyers every day (Thai Graphic Designers Association (ThaiGa)) In this research we focus exclusively on graphic design on the product only to be in line with the topic and purpose of the research, in order to link with the design of the product. In this research, industry and researchers had the idea to integrate nanotechnology into the mix in order to solve the woven fabric to mold or moisture that often occurs with fabrics from natural fabric and to add value to their products as well.

2. Research Methodology

2.1 To study of Characteristics' physical of Banana fabric, Nano technology and Graphic design on products
2.2 To study graphic design and printing process on fabrics from Banana fabric
2.3 To produce prototype products
2.4 Data analysis of user evaluation of the products
2.5 Transfer knowledge the community and evaluate the satisfaction of the participants

3. methods

3.1 Investigation in the shopping area and store and check samples of women's bag products available on the market.

3.2 Graphic Design Process and Printing Process on Banana fabric.

In this research, the researchers designed graphic patterns for printing on Banana fabric with the idea of re-arranging the lines together and a pastel color. It is a flurry of lines and feel movement from the curve. Make it look and feel comfortable. According to the color trends of 2017, which can bring different shades. To apply to various opportunities. As designed and color coded in the design process. This graphic pattern can be viewed frequently. When printed on fabric and developed into a product. The result is that the product looks stylish, appropriate to the gender and age of the user. It was include product opportunities.

Fig. 1 PANTONE Color trends for 2017
sourcing: http://www.fashiontrendsetter.com/

3.3 Printed pattern design and drafting prototyping pattern using color trends 2017 (167c, 419c, 7716c, 7475c, 4685c and 7527c). Sketching of the underlying graphic pattern using the 2017color trend Sublimation or Transfer printing requires sublimation ink, which is a synthetic ink. It is well absorbed in polyester fabric such as TC (polyester) and TK (polyester 100%). The sublimation ink can be absorbed by a natural fabric for printing.

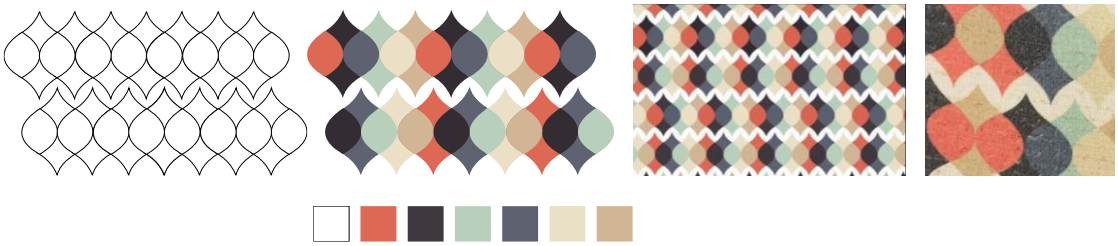


Fig. 2 Graphic designs for printing and prototyping pattern using color trends 2017 on banana fabric

3.4 Bag design process from Banana fabric from the investigation in the shopping area and store and check samples. The data was collected and designed, and then selected by the experts, the possible way to enter the production process. And there are five ways to add value to banana fibers: four graphic of women's handbags (shoulder bag, Backpack, Shoulder Bag and handbags) and 1 suitcase

Bag Design Form 1 **Bag Design Form 2** **Bag Design Form 3** **Bag Design Form 4**

Bag Design Form 5

Fig. 8 Five styles bag design sketching selected

3.5 Finishing Process Banana fabric with Nano innovation

In this research, we used Banana fabric and then printed graphics to nanotechnology to help prevent bacteria. (Anti-Bacterial) and water reflection before being produced as a ladies handbag.

3.6 Data analysis

The researcher collected the data to verify the completeness. Accuracy of data and coding information in mean and percentages as follows:

Part 1 analyze the general data of the respondents.

Part 2 analyze the data from the participated satisfaction assessment on training courses in designing and developing banana fiber fabrics with graphic printing and Nano innovation for towards Economic Community Development

4. Result

4.1 Create Prototype Products

From drafts designed and selected. It is used as a prototype product. Using Banana fabric printed graphics decorated with Nano innovation.

4.1.1 Fabric for prototype production

Innovative Nano-fiber on Banana fabric can be anti-bacterial at 99.95% and *staphylococcus* bacteria *klebsiella* pneumonia at 99.93% and water reflection / water level of 80 percent water, which can be absorbed into the cloth and wet it a bit.

Fig. 10 Banana fabric decorated with Nano innovation

4.1.2 Five prototype of Banana fabrics printing women's bags (shoulder bag Backpack Shoulder Bag and handbags) and 1 suitcase

Fig. 11 Shoulder bag

Fig. 12 Backpack

Fig. 13 Handbags / Shoulder bags

Fig. 14 Clutch bag

Fig. 15 Suitcase

4.2 Consumer Satisfaction Survey of Banana Fabric's user

4.2.1 The mostly respondents were female, aged 21-25 years, earn less than 15,000 baht. All have been satisfied at maximum average and highest level of satisfaction that with an average 4.91, percentage on 98.24 as shown in Chart 1.

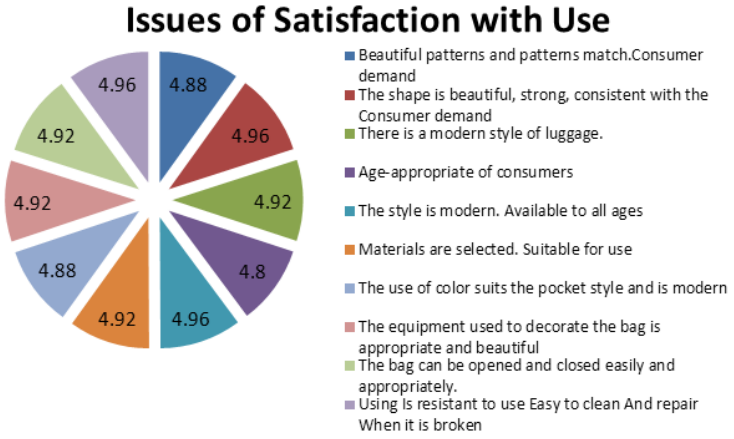


Chart 1 Satisfaction of Transfer to Community

4.2.2 For transfer knowledge the design of products from Banana Fabric with graphic printing and Nano technology fabrics towards economic community development. The workshop schedule of execution Technology for the two days. There are 25 participants including community enterprise and between 17 - 18 September 2016 at the Chedi district office in Suphanburi province. Various women's groups and interested parties within the surrounding area. Most of the participants were satisfied with the level in every question.

Fig. 11 Slipknot bag sketch for Transfer to Community

Fig. 12 Lectures provide knowledge about research
and let participants participate in the printing fabric and the sewing Slipknot bag practical training

Fig. 13 Image of the participants of the project and their products

Table 1 Average and Satisfaction Levels

Question	x	Level of satisfaction
Objective of the project		
1.1 Accordingly demand of trainees	4.92	high
1.2 Accordingly demand of the community group	4.92	high
1.3 Accordingly the purpose of objectives	4.92	high
1.4 The content is correct	4.80	high
1.5 Modern knowledge, suitable for the situation	4.96	high
Service Process		
2.1 Publicity of project management thoroughly	4.76	high
2.2 Timeline and training location	4.76	high
2.3 The content of the training is appropriate	4.76	high
2.4 The organizer is well prepared and provided the service	4.76	high
The lecturer		
3.1 Qualities and personality are appropriate	4.72	high
3.2 Expertise / knowledge of the content of the training	4.72	high
3.3 Ability to transfer knowledge	4.72	high
3.4 The technique of knowledge transfer is interesting	4.96	high
3.5 Training time management	4.92	high
3.6 Clarity in answering questions	4.92	high
Facility		
4.1 Sufficiency of general facilities	4.92	high
4.2 The service and facilities of the project team are very good	4.96	high
Average all	**4.85**	
percentage	**96.94**	

5. Conclusion and Discussion

5.1.1 Based on this research, the innovative banana fabric was coated with nanoparticles. To antibacterial (Anti-bacterial) and water repellent (water repellent) that inhibits the bacteria *Staphylococcus* at 99.95% and *klebsiella pneumonia* at 99.93% and the water / water reflector can reflect water at 80 percent. The test area is wet, the water can be seep into the fabric a little and the fabric is slightly damp. Moreover, It can absorb water and moisture well.

5.1.2 Based on the study, Design and development of products and prototype products from Banana fabric with graphic design using the 2017 color trend and printing and Nano innovation. Then, consumers' satisfaction toward the use of Banana fabric was evaluated, the results showed that most agreed with the design. Whether it is a beautiful shape, strong, consistent with the needs of consumers. The bags which their pattern is modern and stylish, suitable for ages of consumers. The material is selected suitable for using. The color is appropriate the bag style and device that is decorated bag is appropriate and beautiful. The bag can be opened closed easily, easy to clean and repair when it is broken

5.1.3 Technology transfer activities on September 17 - 18, 2016. The training was divided into 5 groups. When the training was completed, the participants' satisfaction with training was assessed. Using questionnaires to evaluate. The results was separated 2 parts. Part 1, the most of the participants were female and aged 31-40 years old. Have a career as a farmer and most of them do not have income or earn 5,001-10,000 baht. Part 2 asks the satisfaction of the trainees. It is a question that asks about satisfaction with the curriculum. It is divided into 3 aspects: the objectives of the project. Curriculum structure and the content of the course. It was found that most of the samples were most satisfied.

6. Acknowledgments

Research on the design and development of banana fiber fabrics with graphical printing and Nano innovation towards Economic Community Development. It can be successfully with the kind support of the National Research Council of Thailand, which has endorsed the evaluation of research proposals, offer the fiscal year 2016. **This research was supported in year's budget by Rajamangala University of Technology Phra Nakhon and we're great to THANK YOU Rajamangala University of Technology Phra Nakhon** for providing the opportunity and funding of this research to the research's team. Include participants in technology transfer activities from Chedi district, U Thong district, Suphanburi province. Thank you to all of our research team who are dedicated to working hard and working to resolve all the problems and obstacles that arise during the research process. The benefits of this research are to strengthen the community. It stimulates the economy as a whole. The results of this research may be Creating a community product is another way.

7. reference

Bussara Soiraya. **Development of industrial banana fiber products and the development of prototype yarns and fiber yarns.** Full research report fiscal year 2550.

H.J. Zwaga and T. Boersema.1983, **Evaluation of a set of graphic symbols.**

Yi-Lin Lo and Chang-Franw Lee/ Melaka, 7 – 10 December 2010, **A Study of Text and Icon on Drug Bag Design.**

Thai Graphic Designers Association (ThaiGa)

Aspelund, Kari. **The Design Procress.** New York : Fairchild Publications, Inc. 2006. Verlag/Bangert Publications, 1988.

Black, Peter H. **"Seeking the Ideal From : Product Design and Consumer Response".** Journal of Marketing, 1995.

De Chiara, J.S., Pancro, J.L., and Zelnik, M.T. **Time-Saver Standards for Interior Design and Planning.** Singapore : Mc Graw-Hill Inc, 1992.

Heskett, John. **Industrial Design. London** : Thames and Hudson Ltd, 1993.

Lulius Panero & Martin Zelnik. **Human Dimension & Interior Space.** New york : An imprint of Watson - Guptill Publications, 1979.

YARN DEVELOPMENT FROM RICE STRAW TO COMMERCIAL

Autcha Hattayananont [1*], Praparnporn Theeramongkol[2], Bussara Sioyraya[3]

[1]*Department of Fashion Clothing and Textile Design, Faculty of Home Economics, Rajamangala University of Technology Phra Nakhon, Bangkok 10300, Thailand*

Abstract

This research studied the process of seperating fibers from rice straw. To study the physical properties of fibers from rice straw and the process of manufacturing yarn from rice straw fibers. It was analyzed by physical textile testing laboratory. It has been developed as woven and develop into commercial products. The process of separating fibers from dried rice straw and to select the rice straw fiber length at least 30 cm. to subtract and peel off the joints of the lower end by hand. Then, Rice straw fiber was in the physical laboratory, it is cellulose fibers, tensile strength (Newton) at 28.18, elongation (percent) is at 2.01. There are metals plenty of CADMIUM and LEAD at 0.1 mg/kg, CHROMIUM (TOTAL) / (VI) and COPPER at 0.5 mg/kg. The fiber cross-section is oval and clearly. Its length is smooth and transparent when viewed from the side to see the clarity of the fiber. Rice straw fiber weave in plain structure. Warp yarn uses a special brown, cream and brown flat yarns. There are 2 warps yarn and using natural color without blench. When woven as a fabric to develop for 4 products prototype : table lamps, tablecloths in Japanese style, potted plants and Accessories Box.

Keywords: yarn development**,** rice straw**,** commercial

1. Preface

Current, environmental issues around the world are experiencing an ongoing basis. In particular, Waste is increasing day by day. The cause of residual waste disposal, both types of waste are biodegradable and degradation. Some types can be recycled and can reused to benefit another. As above, almost problems can be found them in industrial sector especially, the textile industry: spinning yarn and fabric production and includes the apparel and every products from any enterprises: industrial enterprises, community enterprises and communities. In order to reduce the amount of textile materials waste from textile manufacturing, which cause pollution, reduce residual waste and also to enhance the knowledge. Moreover, contributed to develop of textile products, both in public sector, private sector and communities groups involved. Researchers realize and recognize alternatives to develop a new special yarn which using textile materials waste to re-produce a new value and to be add-valued.

2. Research Methodology

2.1 To study the sorting process of fibers from rice straw
2.2 To study the physical properties of fibers from rice straw
2.3 To study the fibers production of rice straw yarn

3. Methods

3.1 Study and find raw materials. After harvesting grain with a grain truck. The farmer will leave straw and rice stubble that may still be a green color. To lay them down on the floor, the sunlight burn for a few days, then the straw and rice to collect the fiber to continue to separate the fiber.

Fig. 1 showing harvests by harrow and rice harvested from harvest.

3.2 Physical testing of properties of rice straw, We use a rice strew which was sunlight few days of sun-dried rice, and go through the physical testing. When the test was carried out, the rice cobs were prepared by equilibrating in a laboratory at 20 ° C ± 2 ° C with a tolerance of 65 ± 2 for 8 hours (Bussara et al., 2007).

3.2.1 The cross section of the fiber testing

3.2.1.1 Bring 1 yarn of rice strew with rayon yarn.

3.2.1.2 Put the yarn through a small hole punched. To cut off the excess of the steel plate both front and rear. The only fiber in the hole in the plate.

3.2.1.3 Bring the fiber to the microscope. (OLYMPUS (BX41) The magnification of the microscope is 100 times. When you see the cross section of the fiber is clear, take a digital picture.

3.2.2 Fiber strength Tested with a test machine. Tensile Testing Machine (Instron Model 5566) It has a test speed of 300 millimeters per minute. Use a test run of 100 millimeters and a temperature condition. 21 ± 1 ° C and a relative humidity of 65 ± 2%

3.2.3 Analysis of Heavy Metals by Tester UV-VIS Spectrometer and INDUCTIVELY COUPLED PLASMA-OPTICAL EMISSION Spectrometer To find the amount of heavy metals in the fiber as follow CADMIUM CHROMIUM (TOTAL) , (VI) COPPER and LEAD

3.4.4 Cross section and longitudinal section of fiber from rice paddy Using the test machine.
LIGHT MICROSCOPE (OLYMPUS BX41) here is the magnification of the microscope. Cross Section 200 x Longitudinal Region 40 x

3.3 Weaving process

Type of weaving machine is table weaving machine. Its size is 80x60 cm. square, made of wood and consisting of plastic seals. It can split the yarn into two sets, width of fabric can be woven up to 60 centimeters. The characteristic of this yarn used in weaving is flat polyester yarns and difference color.

Fig. 2 Table weaving machine and flat polyester yarns

4. Result

4.1 To study the sorting process of rice stew fiber from rice straw

In the experiment, the researcher brought the sun-dried rice straw for 2-3 days. The fiber is sorted by choose the specific fiber that is the stem of the rice and not less than 30 centimeters. Use the handle of the rice straw on the top and bottom of the bottom of the trunk. Break the joints and pull the shell of the rice straw. Be careful truck not to be broken. Place the strips of peeled rice in a horizontal position, tie them together to prepare for woven into a cloth. The Important thing is should be covered them by wet cloth before weaving.

Fig. 3 Image of peeling rice straw

4.2 Test results of physical properties of rice fiber.

Study and physical properties of fiber from rice straw. Physical properties of rice straw at the textile testing center, Textile Industry Development Institute, the results are as follows.

4.2.1 Based on the test results of ASTM D 276: 2000, the type of fiber is CELLULOSIC FIBER or cellulose fiber.

4.2.2 Based on the test results of the Tensile Testing Machine (Instron Model 5566) with a test speed of 300 millimeters per minute.Use a test run of 100 millimeters and a temperature condition. 21 ± 1 ° C and a relative humidity of 65 ± 2%. It was found that Fiber strength of the rice straw had the highest tensile strength (Newton) at 28.18 and the elongation at 2.01.

4.2.3 Analysis of heavy metals by UV-VIS Spectrometer and INDUCTIVELY COUPLED PLASMA-OPTICAL EMISSION Spectrometer to determine the amount of heavy metals in the fiber as follow

4.2.3.1 CADMIUM found that rice cobs contained a quantity of CADMIUM of 0.1 mg / kg

4.2.3

CHROMIUM (TOTAL), (VI) at 0.5 mg / kg

4.2.3.3 COPPER found that rice cassava had a COPPER content of 0.5 mg / kg.
4.2.3.4 LEAD found that rice cassava had a LEAD content of 0.1 mg / kg
It is all heavy metal are not over the standard maximum

4.2.4 Cross section and longitudinal section of fiber from rice straw, using the MICROSCOPE LIGHT TESTER (OLYMPUS BX41) has the magnification of the microscope. Cross sectional area 200 x longitudinal region 40 x

4.2.4.1 Cross-sectional area of rice fiber Oval shape, there is a lumen in the center of fiber. It can be seen clearly. The longitudinal section have a smooth fiber surface, which is translucent. It can clearly see the lumen in the middle of the fiber with the following details:

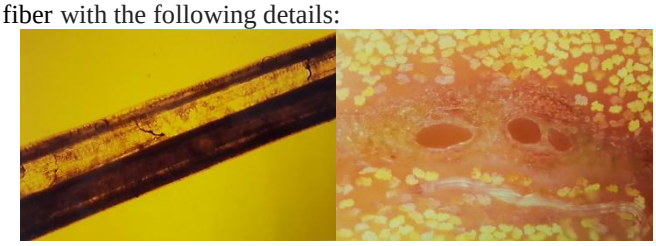


Fig. 4 Cross-sectional area of rice fiber and Longitudinal section of rice fiber

4.3 Weaving cloth from rice straw fiber

4.3.1 The weaving machine used is a table weaving machine. The weaving pattern is basic weaving structure. Warp yarn used as flat poly yarn (Cream and dark brown), the yarn is made of 2 types of selected rice straw fibers, namely natural yarn and natural rice staple. The type of ashes used is the type of ash, the color after the dye is found that the color is slightly less sticky. Because of the surface of the rice cob fibers are polished and smooth.

Yarn, rice, natural color Yarn of rice stubble dyed with dried okra

Fig. 5 Woven fabrics using yarn dyed from natural yarn and rice yarn dyed with dried okra

4.4 Product Development from rice straw (Prototype)

4.4.1 table lamp

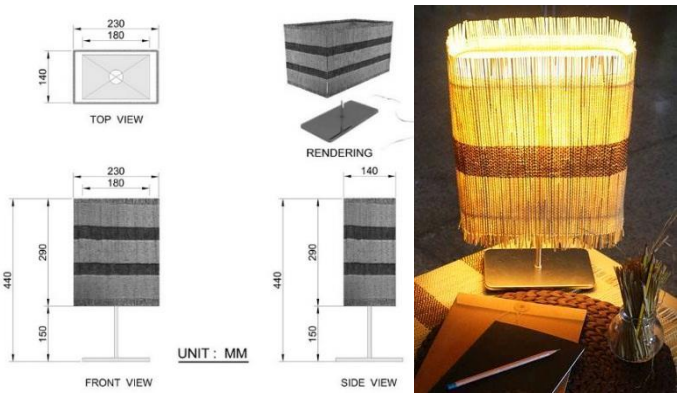


Fig. 6 Table Lamps prototype product

4.4.2 Japanese tablecloth

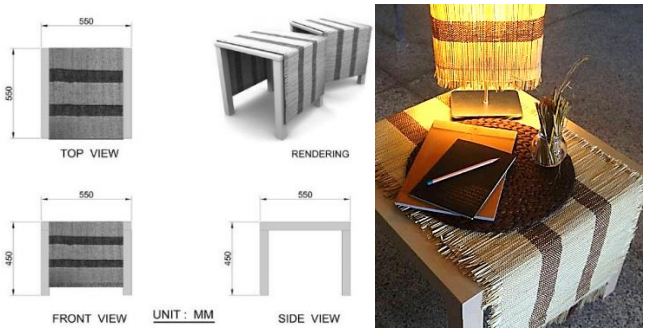


Fig. 7 Japanese tablecloth prototype product

4.4.3 Flowerpot

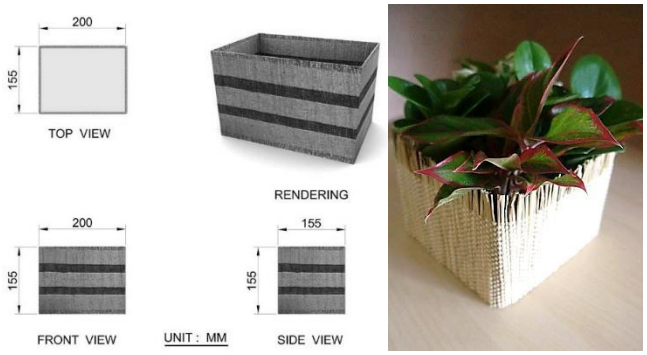


Fig. 8 Flowerpot prototype products

4.4.4 Multi-purpose box

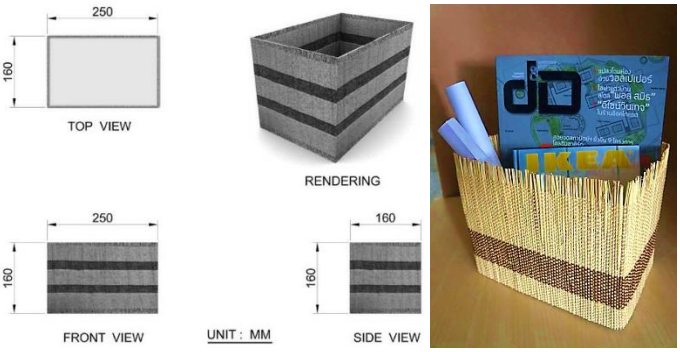


Fig. 9 Multi-purpose box prototype product

5. Conclusion and Discussion

5.1 Conclusion of the research

5.1.1 The sorting process of the fiber from the rice straw in the experiment of using the rice straw on the sun dyed for 2-3 days, then the fiber is selected by selecting the fiber of the trunk of the rice. Choose from a trunk of not less than 30 centimeters long, broken at the joints of the trunk, pull the rice husk out. Bring rice straw to peel. Place them flat and tied together. Keep it in unpolluted weather to prevent fungus and broken fibers. When we weave the fabric, we should be covered the rice straw yarn by wet cloth.

5.1.2 Test results of physical properties of rice fiber Study and physical properties of fiber from rice paddy. Physical properties of rice cassava at the textile testing center. Textile Industry Development Institute The result is a rice stubble. Organized in cellulose fibers. The maximum tensile strength (Newton) was 28.18 and the elongation was 2.01. Based on the heavy metal analysis by UV-VIS Spectrometer and INDUCTIVELY COUPLED PLASMA-OPTICAL EMISSION Spectrometer, CHROMIUM (TOTAL), (VI) showed that rice husk contained

CHROMIUM (TOTAL), (VI) at 0.5 mg / kg COPPER. COPPER was 0.5 mg / kg and LEAD showed that rice straw had a LEAD content of 0.1 mg / k G, where the amount of such substances is not considered to be harmful to the customers. Cross-sectional and longitudinal of fiber from rice straw. Using the MICROSCOPE LIGHT TESTER (OLYMPUS BX41) has the magnification of the microscope. Cross sectional area 200 x longitudinal region 40 x by the effect of cross sectional area of rice fiber. Oval shape. The fiber in the middle of the fiber is clearly visible, and the longitudinal section of the fiber is found to have a smooth, translucent fiber surface. It can clearly see the lumen in the middle of the fiber.

5.1.3 Woven fabrics of rice fiber, weaving and the type of weaving machine. The pattern of weaving is a weaving, which is the strongest woven structure. Warp yarn used as a special flat yarn (brown, cream and dark brown) and weft yarn. We use the rice straw fiber with the same yarn as the warp yarn. There are two types of yarn that are made from rice fiber. Using the natural color of the rice cob It is not dyed or dyed in anyway. When woven into a cloth, it was made into 4 prototype products as table lamps, Japanese tablecloths, flowerpots and multi-purpose boxes.

5.2 Suggestions for future research findings and research.

5.2.1 There should be an experiment on dyeing. Whether it is synthetic dyeing or other natural dyeing. Along with other adherence assays to see the effects of pigment staining on rice fiber.

5.2.2 In the next research should be changed to more commercial form. By pushing the manufacturing process into the industrial system. To try to get more production in the future.

5.2.3 The next research should be developed in the form of a woven structure, such as weaving create more beautiful products.

6. Acknowledgments

Research Yarn Development from Rice straw to commercial. It can be successfully with the kind support of the National Research Council of Thailand, which has endorsed the evaluation of research proposals. **This research was supported in year's budget by Rajamangala University of Technology Phra Nakhon and we're great to THANK YOU Rajamangala University of Technology Phra Nakhon** for providing the opportunity and funding of this research to the research's team. Include participants in technology transfer activities from Chedi district, U Thong district, Suphanburi province. Thank you to all of our research team who are dedicated to working hard and working to resolve all the problems and obstacles that arise during the research process. The benefits of this research are to strengthen the community. It stimulates the economy as a whole. The results of this research may be Creating a community product is another way.

7. reference

Bussara Soiraya. **Development of industrial banana fiber products and the development of prototype yarns and fiber yarns.** Full research report fiscal year 2007.

Corbman, B.P. 1983. **Textile Fiber to fabric**. 6 th ed. Singapore : Mc graw-hill.

BunpordTekajarin. 1996**. Yarn production Department of Textile Industry Promotion**. Ministry of industry**,** Bangkok.

Innovation technology, financial markets, and economic growth

Effect of Child Labour Abuse in Multinational Company on Consumer Behaviour of Teenagers and Early Adults

Orachun Jansritanyagose[1,a*], Yonten[1,b], Yangchen Pema[1,c],
Benyapa Wirunhapinyo[1,d], Kwanruetai Boonyasana[2,e]

[1]Department of International Business, Faculty of Business Administration,
Rajamangala University of Technology Phra Nakhon, Bangkok 10300, Thailand
[2]Department of Finance, Faculty of Business Administration,
Rajamangala University of Technology Phra Nakhon, Bangkok 10300, Thailand

Keywords: child labour abuse, consumer behaviours, multinational company

Abstract Child labour abuse is one of the inequalities of society in relation to exploitation either through the company controlling a cost or the labour force working unconditionally. This research investigates the relationship between a company's rumoured use of child labour and consumer behaviour. A mix-methodologies approach involves observation and analysis while the population for this study comprises 19 to 24-year-old consumers of cereal product, who lived in Bangkok in 2016. Quantitative data is provided by way of a questionnaire to 100 subjects, and qualitative data through in-depth interviews of 10 subjects. The chi-square test result of $p < 0.01$ indicates two variables are strongly significant, with the young generation in question still consuming the cereal product as usual even after hearing the rumour, showing that teenagers and early adults are not concerned about this issue before purchasing. What concerns them more are product taste appeal, affordable price, belief in product, and if child labour is involved legally, it is seen as providing an opportunity of a career path for children. Therefore, although the issue of child labour abuse might harm a firm's reputation, there is room for understanding of the relationship between consumer and company product.

Introduction

In 2015, the United Nations International Children's Emergency Fund (UNICEF), based on information from the International Labour Organization (ILO) and the World Bank, stated that 168 million children worldwide, from 5 to 17 years of age, were trapped in child labour situations subject to unfair income, lack of workplace security, no social protection, inappropriate work attributes and working under conditions of slavery [1]. The Compassion International in 2017 further stated, "Forced labour is thought to generate around $150 billion a year in illegal profits" [2]. A big company could extract a commercial advantage with regard to this issue, and rumour to this effect has been associated with the Nestlé company. This company produces a diversity of goods for consumers around the world especially food products whose popularity can be impacted by consumer behaviour [3].

Nowadays, the acceleration of globalization has affected human behaviour. Human interaction can involve either exploitation or benefit through fast moving technologies and the impact of the digital revolution on business trends. Such change in technology can lead to disruptive conse-

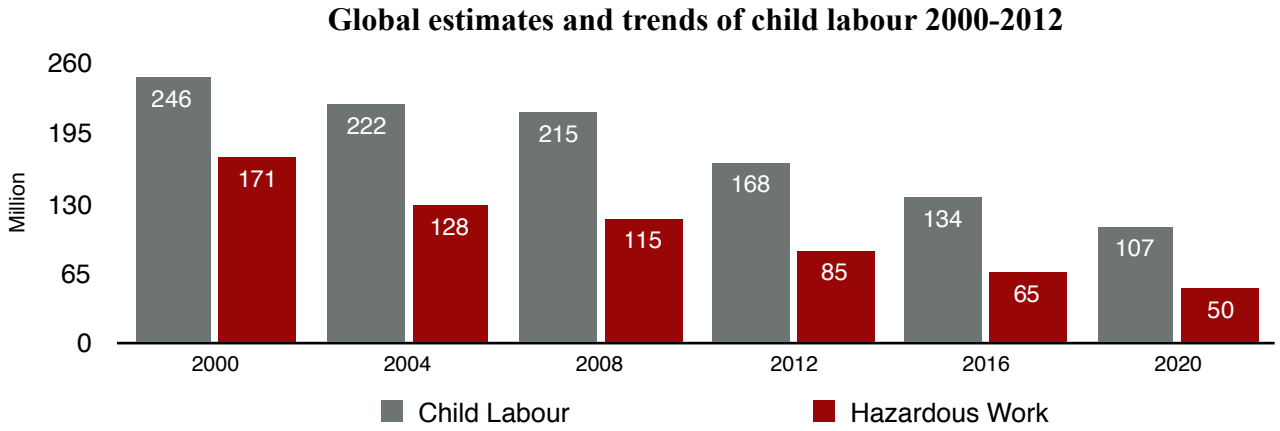
quences such as bankruptcy for businesses from time to time [4]. Some businesses fail because of an inability to cope with harsh situations or adjust to upcoming trends. Important categories for human survival and interaction include availability of fresh water, effective medicine, adequate shelter and food.

With regard to food, the current trend is towards healthy consumption and away from fast foods. As a result, businesses produce healthy food as an option for the consumer although fast food remains popular among teenagers, early adults and workers who lack time for food preparation and cooking. Hence, many people in this generation would prefer cereal for breakfast because it is faster than cooking and they still obtain nutrition from the grain they consume. There exists a huge potential for cereal companies such as Nestlé and Kellogg's who have access to worldwide markets.

Sometimes these businesses involving food manufacture encounter rumours regarding production. Such a rumour involved the Nestlé company with regard to the use of child labour in the production of cocoa. Humphrey Hawksley in 2012 mentioned via the British Broadcasting Corporation (BBC) news service that "cocoa is the raw product that makes chocolate in a global industry worth more than $90bn (£58bn) a year" [5]. Nestlé is making a lot of money from this industry and tries to generate corporate social responsibility within the cocoa supply chain process and set policy guidelines within the company [6].

Accordingly, the company provides support for farms, new schools, examining supply, monitoring and remediation and transparency within the cocoa business. Assistance from UNICEF is by way of "Roadmap for Achieving the Elimination of the Worst Forms of Child Labour by 2016 [1]" which could help eliminate child labour abuse through support of cultural differences and provision of resources such as household income, hospital services or access to nurseries, quality education and child protection [1].

Figure 1: Child labour trend



Even though child labour is declining, at the current rate there will still be more than 100 million children in the labour force by 2020 [1]. The situation could be improved by implementation of the UNICEF roadmap and other organizations working together. The roadmap is called "Roadmap for Achieving the Elimination of the Worst Forms of Child Labour by 2016" [8].

Methods and Procedures

- The Chi-square test and significance

Qualtrics (2011) states that the chi-square method is the basic test used for significance where cross-tabulation variables are included. The statistical data analysis for the chi-square test involves a study of the relationship and gives a value of significance (Hartman, 2014). The test evaluates how likely it is that any observed difference between sets arises by chance.

A P-value indicates the level of significance with a reading greater than 0.1 meaning the result is not significant and the relationship can be attributed to chance [7].

If the P-value is between 0.1 and 0.05, it means that the result is significant at the 10 percent level or P-value is less than 0.1. If the P-value is between 0.05 and 0.01, it means the result is significant at the 5 percent level or P-value is less than 0.05. If the P-value is below 0.01, it means the result is significant at the 1 percent level or P-value is less than 0.01. The level of significance indicates the strength of the relationship under analysis (Pallant, 2010; Qualtrics, 2011). P-value readings above 0.05 are regarded as indications of non-significance in this study [7].

Table 1: The Chi-square test results from data collected

Chi-square Tests

	Value	df	Asymptotic Significant (2-sided)
Pearson Chi-square	26.493[a]	9	0.002
Likelihood Ratio	29.984	9	0.000
Linear-by-Linear Association	5.674	1	0.017
N of Valid Cases	100		

a. 9 cells (56.3%) have expected count less than 5. The minimum expected count is 1.10.

- Cross-tabulation Analysis

This research examines the relationship between goods produced through possible child labour abuse and frequency of buying after hearing the rumour. The chi-square test results indicate a strongly significant level of relationship between the two ($p = 0.002$). The chi-square table in results should include a value and associated level of significance (p) (Asymp. Sig. (2-sided). The significant value should be smaller than 0.05 (Pallant, 2010; Qualtrics, 2011) [7].

Empirical Results

The questionnaires were completed by 100 respondents, from 19 to 24 years of age, 53% of whom were male and 47% female. Results show a significant relationship between child labour abuse and consumption in relation to the rumour for cereal production in multinational companies. (see table 2).

However, this study employs mix-methodological research focusing on quantitative data through questionnaire and qualitative data through in-depth interview. The results here show that the customers still purchase and consume the product regardless of the rumour that child labour is involved. Reasons given include product taste appeal, affordable price, and belief in the product.

- Hypothesis

H_O: There is no relationship between child labour abuse and consumption in relation to the rumour for cereal production in multinational companies.

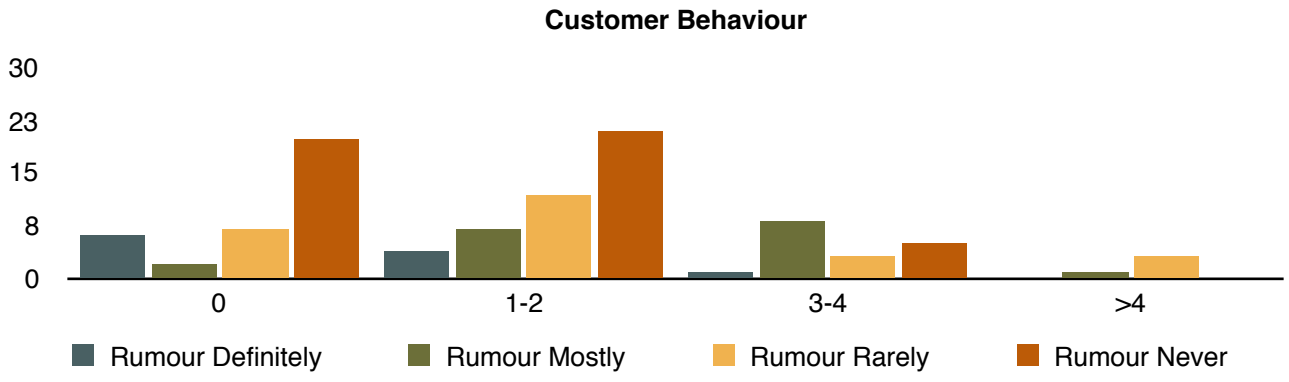
H_1: There is a relationship between these variables.

Table 2: Test of significant relationship between child labour abuse and consumption in relation to the rumour.

Effect of Child Labour Abuse		Consumption in Relation to the Rumour				X^2	X^2
		0	1-2	3-4	more than 4	value	prob
Rumor	Definitely	6	4	1	0	26.49	0.002
		17.14%	9.09%	5.88%	0%		
	Mostly	2	7	8	1		
		5.71%	15.91%	47.06%	25%		
	Rarely	7	12	3	3		
		20%	27.27%	17.65%	75%		
	Never	20	21	5	0		
		57.14%	47.73%	29.41%	0%		
Total		35	44	17	4		
		100.0%	100.0%	100.0%	100.0%		

Table 2 shows the chi-square p-value (0.002) is less than our α value (0.01). Therefore, H_O is rejected indicating that there is a strongly significant relationship between child labour abuse and consumption in relation to the rumour for cereal production in multinational companies. Most of the consumers (26.49%) who selected to consume for their first choice, agree that there is an effect of child labour for their consumption.

Figure 2: The customer behaviour after heart the rumour in chart



According to the figure above, there are two charts where the numbers are getting close to each other. The higher one shows the consumer will purchase and consume as usual per week. The second higher one shows they will not consume and purchase the product.

We see that the results show a relationship at $p < 0.01$ level ($x2 = 26.493$, $df = 9$, $p = 0.002$) which means the results are strongly significant at the highest level, 1% level of significance (Pallant, 2010; Qualtrics, 2011). The results of the analysis show a Pearson chi-square value of 0.002, which indicates significance at the highest level (Pallant, 2010; Qualtrics, 2011). From this, it is

seen that 44% of consumers would still purchase a product after hearing the rumour of child labour abuse, while 35% never would. Even though the percentage numbers for each group are close, this study shows that, while the rumour is important to consumer behaviour, perspective on production is viewed differently.

In addition, this study includes in-depth interviews to examine consumer perspective and how consumer behaviour might change after hearing the rumour regarding child labour abuse. Results here show that consumers are not concerned about the rumour and continue buying the cereal product regardless. Reasons put forward include product taste appeal, affordable price, belief in the product, and if child labour is involved legally, it is viewed as providing an opportunity for a career path for children.

Conclusion

This paper examines that there is a strongly significant relationship between the rumour of child labour abuse and consumer behaviour after hearing the rumour. A mix-methodologies approach is used to gather quantitative and qualitative data on consumption of multinational companies cereal product. The populations for this study comprise residents of Bangkok in 2016 of ages 19 to 24 years, with 100 subjects partaking in a questionnaire, and 10 subjects partaking in in-depth interviews. Results show that, after hearing the rumour of the company being involved in child labour abuse for cocoa production in a developing country, people consume as usual.

Acknowledgment

The authors express their thanks to Rajamangala University of Technology Phra Nakhon. In addition, the authors would like to thank Mr. Robin Neill for support and suggestions.

References

[1] UNICEF. Child protection from violence, exploitation and abuse. (2015). Information on https://www.unicef.org/protection/57929_child_labour.html

[2] The Compassion International, Child Labour Facts. (2017). Information on https://www.compassion.com/poverty/child-labour-quick-facts.htm

[3] J. Mccarthy, The child labour lawsuit against Nestlé: what you need to know. (2016). Information on https://www.globalcitizen.org/en/content/the-child-labour-lawsuit-against-nestle-what-you-ne/

[4] C. M. Christensen, The Innovator's Dilemma: When New Technologies Cause Great Firms to Fail. Boston, MA: Harvard Business School Press, 1997

[5] H. Hawksley, Nestle 'failing' on child labour abuse, says FLA report. (2012). Information on http://www.bbc.com/news/world-africa-18644870

[6] Nestlé, Nestlé cocoa plan delivers more sustainable cocoa. (2012). Information on https://www.nestle.co.th/en/media/pressreleases/cocoa_plan_delivers_more_sustainable_cocoa

[7] S. Follin & V. Fransson, The impact of gender and age on customer loyalty – a quantitative study of Swedish customers' experiences of a loyalty program. Linnæus University (2013) 38-58

[8] International Labour Organization. (2013). Global estimates and trends of child labour 2000-2012. Information on http://www.ilo.org/wcmsp5/groups/public/@ed_norm/@ipec/documents/publication/wcms_221881.pdf

An Impact of EU's Generalized System of Preference Withdrawal on Thai Frozen Shrimp Export

Sorapat Sirorab[1,a*] Nuttapon Photchanaprasert[2,b]

[1, 2]The department of agricultural and resource economics, Faculty of economics, Kasetsart University Bang khen campus, Thailand, 10900

Keywords: Generalized System of Preference (GSP), Frozen Shrimp, Competitiveness.

Abstract. As a recent result of EU's Generalized System of Preference (GSP) withdrawal on Thailand, the tariff preference for Thai frozen shrimp export to the European Union had been deleted and consequently the duty of the frozen shrimp nearly tripled from 4.2 to 12 percent in 2015. This paper attempts to investigate the impact of EU's GSP withdrawal on Thai frozen shrimp export using SMART model to measure the total effects of the tariff increase - trade contraction and trade diversion. Additionally, this paper also applied the constant market share (CMS) model to investigate the change in Thailand's competitiveness during pre and post suspension of EU's GSP. The results present that the cancellation of tariff preference had a significantly negative impact on volume of Thai frozen shrimp export to EU market, derived from the trade contraction and trade diversion towards major competitors. Additionally, this cancellation also noticeably made Thailand lost its competitiveness to other trade rivals in EU market as well. Therefore, a remedial measure should be provided to Thai frozen shrimp exporters, and improvement of production cost management and quality standard as well as negotiation of FTA agreement between Thailand and EU should be expedited in order to promote Thai frozen shrimp export and retrieve Thailand's competitiveness back.

Introduction

Thailand has been a major player in shrimp production with 30-year experiences, and has rapidly become a major producer and exporter in the world. Generally, shrimp cultivation (farming) with intensive system in Thailand are operated in several parts of the country with approximately 23,900 individual farms, spreading across around 80,000 hectares which are capable of annually producing shrimp around 496,988 metric tons. There are around 180 shrimp processors which have ability to export its products to overseas, consisting of 66 medium-sized and 124 large-sized processors, with over 700,000 workforces [1]. In 2014, shrimp products were ranked 6th for the agricultural commodity generating the highest value of Thai agricultural exports, and the profitable form of shrimp which has export potential is frozen shrimp product [2].

In term of international trade, the major export destinations of Thai frozen shrimp consist of United States, Japan, the European Union 15[1] and Canada. The European Union15 is the largest importing market for frozen shrimp, and strategically the third biggest export market for Thai frozen shrimp annually generating Thailand's revenues around 132 million euros, sharing around 13 percent of total Thai frozen shrimp export to global market during 2008 – 2014 on average [3]. Historically, the European Union has previously granted Thailand with preferential market access

[1] The volume of Thai frozen shrimp export to EU 15 is approximately accounted for 98 percent of export to EU 27 [3].

as EU's generalized system of preferences (GSP)[2] since 2006. As a result, the tariff of around 6200 products exported by Thailand was separately reduced depending on degree of sensitivity. For Thai frozen shrimp, it was listed as a semi-sensitive product eligible for tariff privilege leading to tariff reduction from 12[3] % to 4.2[4]%. Next, the preference has undeniably finished due to establishment of EU's new GSP's regime in regulation 978/2012 [5], and Thailand met the criterion of EU's GSP graduation mechanism. Thailand had been classified by World Bank as a high or upper-middle income country for three consecutive years during 2011 – 2013[5] [6], and was consequently judged to be excluded from the EU's GSP as well as tariff preference cancelled, becoming effective at the beginning of 2015. The cancellation of tariff preference has been concerned and expected to have a considerable impact on Thai frozen shrimp export to the European Union as the duty of the frozen shrimp increased and nearly tripled from 4.2 to 12 %. Moreover, Thai frozen shrimp has been the most important fishery product exported to EU with high comparative advantage[6] and has also intensively relied on the tariff privilege from the program.

According to the change in duty, past studies according to theory of custom union reveals that the change in tariff can lead to change in price of product export and consequently to change in trade, depending on the derivation of trade creation and trade diversion [7]. In the analysis of tariff reduction for an importing country as seen in [8, 9, 10, 11], trade creation can take place when the reduction of tariff allows trade partner imports to replace high-cost domestic production, improving in welfare. On other sides, Trade diversion can emerge when the removal of tariff causes trade to divert from other countries to the partner country on the fact that the other countries are the low cost source of imports. On the other hand, in the case of Thailand facing with suspension of tariff preference (a tariff increase), the termination of tariff preference could cause the European Union to swift to purchase more frozen shrimp produced in their domestic sector instead of importing from Thailand (negative trade creation or trade contraction)[7] and simultaneously could causes the European Union to divert their import to the Thailand's major competitors such as Ecuador, India, Vietnam, Indonesia and Bangladesh which still benefit from EU's GSP (trade diversion). In addition, the increase in the tariff might made Thailand lost its competitiveness to other trade rival in EU market due to the trade effects and more expensive-priced frozen shrimp derived from the change in tariff advantage[8].

The main objectives of this paper consist of two parts. Firstly, this paper attempts to investigate the impact of tariff increase, derived from the EU's generalized system of preference withdrawal on Thai frozen shrimp export, analyzing trade contraction and trade diversion effects. Secondly, this

[2] EU's generalized system of preferences (GSP) has been recognized as important features of the European Union's commercial policy for many decades. The system provides developing countries with easier market access to the European Union generally in term of tariff exemption with the major goals to stimulate developing countries' exports and economic development without requirement in reciprocation from the countries concerned. The details were described in [4],

[3] Frozen shrimp was classified as a semi sensitive product, and the modulated preferential duty for imported frozen shrimp is, hence, 65% duty reduction.

[4] As Thailand is the member of World Trade Organization (WTO), it was granted a special favor or an exemption of tariff rate under WTO agreement, called "Most-Favored-Nation" (MFN) and consequently was able to export its frozen shrimp to the countries under WTO agreement with duty imposition at 12%.

[5] Upper – Middle income country is a country with an average gross national income per capita of US$3,976 to US$12,275. This was classified by the World Bank and calculated by using the atlas method.

[6] The comparative advantage of Thai frozen shrimp export to EU during 2006 – 2012 exceeds one, accounting for 2.449 on averages

[7] In the analysis of tariff increase for an exporting country as this study, the trade creation would be negative aspect so called "trade contraction", while trade diversion is still called the same.

[8] Without the EU's GSP, Thai frozen shrimp exporters has to pay 7.8 percent more in term of duty

paper also aims to investigates the change in competitiveness of Thai frozen shrimp export in EU market pre and post EU's GSP withdrawal in order to provide the related stakeholders and government with useful information and policy recommendations for promoting Thai frozen shrimp export in the future.

The balance of the paper is organized as follows. The following section introduces methodology, followed by the results as well as summary and policy recommendations, showed and discussed, respectively.

Methodology

This section is comprised of 3 parts. First part describes the data collection and data source. Second and Third parts specify the SMART and CMS models applied to investigate the effect of tariff increase and the change in competitiveness, respectively.

Data collection and Data source

This study used quarterly time series data from January 2007 to December 2014 accounting for 32 observations and yearly time series data from 2014 – 2015. The data on prices and quantities of frozen shrimp were collected from Trade Map database and other data was collected from office of agricultural economics in Thailand, Thai Customs Department, EURO STAT and Bank of Thailand including related researches.

SMART Model-Partial Equilibrium Analysis

In order to investigate the impact of EU's generalized system of preference withdrawal on Thai frozen shrimp export, this study applied the SMART model to do partial equilibrium analysis, measuring the total effects of tariff increase. The SMART model was introduced by UNCTAD so as to estimate various impacts of commercial policy changes. It also consists of analytical modules that support trade policy analysis, covering the effects of trade liberalization and tariff changes. In addition, the advantages of using the SMART model are that firstly it is able to decompose the total trade effects into trade creation and trade diversion which are consistent with the economic foundation, Secondly, it has an ability to analyze the effects of trade policy reforms in the presence of imperfect substitutes which is more appropriate than the homogenous good model[9] developed by [12]. Thirdly, it is able to measure the trade diversion effect in absence of penetration data[10], which differs from the model invented by [8]

(1) Trade contraction or Negative trade creation

Trade contraction is defined as the decrease in EU's demand for Thai frozen shrimp as a result of increasing in price of frozen shrimp exported by Thailand due to abolishment of tariff preference. In other words, EU would turn to purchase the frozen shrimp produced in domestic sector instead of importing from Thailand. Thus, the expression for trade contraction can be written as Eq. 1[11]

[9] The homogenous good model is not consistent with the reality because the same products exported by difference sources typically are heterogeneous and have some discrepancies such as quality, attributes, product design, advertising or even technologies used in production.

[10] The import data from each EU's GSP beneficiary in apparent domestic consumption.

[11] Theoretical model and provenance of the equation in detail are illustrated in [13].

$$TC_c = B_1 m_c \frac{t_c^{MFN} - t_c^{GSP}}{(1+t_c^{GSP})} \left(\frac{1}{1-B_1/\mu_c}\right) \quad (1)$$

Where TC_c is trade contraction in 2015, B_1 price elasticity of EU15's import demand for Thai frozen shrimp. μ_c is export supply elasticity, assumed perfectly elastic[12]. m_c is quantity of Thai frozen shrimp export to EU15 in 2014, t_c^{MFN} and t_c^{GSP} are most favored nation and EU's GSP tariff rate imposing on Thai frozen shrimp, respectively.

The price elasticity of EU15's import demand for Thai frozen shrimp B_1 was estimated by using economics model with Ordinary Least Square (OLS) technique as illustrated in Eq. 2

$$\ln m_{th} = B_0 + B_1 \ln P_{th} + B_2 \ln P_{ecua} + B_3 \ln Y + B_4 \ln EX + B_5 \ln S + \mu \quad (2)$$

Where B_0 is constant. B_1 is price elasticity of EU15's import demand for Thai frozen shrimp. B_2 - B_5 are parameters. m_{th} is quantity of Thai frozen shrimp export to EU15. P_{th} is EU15's import price of frozen shrimp imported from Thailand. P_{ecua} is EU15's import price of frozen shrimp imported from Ecuador. Y is value of EU15's real GDP per capita in US dollars. EX is exchange rate of Thai baht relative to EURO. S is Thai shrimp production index. μ is error term.

(2) Trade diversion

In the case of tariff preference cut, if the tariff increase does not apply to Thailand's other trade rivals, accordingly EU imports of Thai frozen shrimp would decrease owing to the substitution of import from Thailand, becoming relatively more expensive, to its trade rivals. In other words, EU would turn to import the frozen shrimp imported from other GSP beneficiaries instead of importing from Thailand. Thus, the expression for trade diversion can be written as Eq. 3[13]

$$TD_i = \frac{m_i \cdot m_c}{m_{i+} m_c} \frac{t_c^{MFN} - t_c^{GSP}}{(1+t_c^{GSP})} C_i \left[\frac{(m_{i+} m_c)\,\mu_c \mu_i}{(m_{i+} m_c)\,\mu_c \mu_i - m_c \mu_c - \mu_c m_i}\right] \quad (3)$$

Where TD_i is trade diversion toward country i in 2015. C_i elasticity of substitution. μ_c and μ_i are export supply elasticity of Thailand and its competitors, assumed both are perfectly elastic. m_c and m_i are quantity of Thai and its competitors frozen shrimp export to EU15 in 2014.

$i = \{1,2,\ldots..,5\}$, whereas

1 = Ecuador

2 = India

3 = Indonesia

4 = Vietnam

5 = Bangladesh

[12] The infinite elasticity implies that the export supply curves are horizontal and the world prices are kept exogenous. Additionally, it makes sense to set the assumption of infinite export supply elasticity as Thailand and the European Union are minor players in world frozen shrimp market and none of them is able to determine the world price of frozen shrimp [10, 11].

[13] Theoretical model and provenance of the equation in detail are illustrated in [13].

The elasticities of substitution between Thailand and each competitor C_i were estimated by using economics model with Ordinary Least Square (OLS) technique as illustrated in Eq. 4

$$\ln \frac{m_{th}}{m_{ti}} = C_0 + C_i \ln \frac{P_{th}}{P_i} + C_2 \ln Y + \mu \quad (4)$$

Where m_{ti} is quantity of each competitor frozen shrimp exported to EU15. C_0 is constant. C_i elasticity of substitution. C_2 is parameter. P_{th} is EU15's import price of frozen shrimp imported from Thailand. P_i is EU15's import price of frozen shrimp imported from each competitor.

Constant market share model

To investigate the change in competitiveness of Thai frozen shrimp export in EU15 market pre and post EU's GSP withdrawal, this study applied constant market share model using the annual data in 2014 as the base year and 2015 as final year. Constant market shares analysis (CMS) is a model to analyze the export competitiveness of a country during two periods. It is comprised of a technique that can decompose the change in export of a country into a series of component and also provide with identification of the contribution of each component to define the result.

In order to overcome the some application problems in traditional CMS model noted by [14], this study employed CMS model developed by [15] and also applied the model in case of one commodity/market analysis, where there are no commodity composition and structural interaction effects, as seen in [16, 17, 18]. Additionally, this study also used quantitative data instead of value data in order for an avoidance of the effects of exchange rate and price or unit value on analysis[14]. Therefore, the one commodity/market CMS model with quantitative term can be specified in Eq. 5 and Eq. 6

$$\Delta q_j = \underbrace{S_j^0 \Delta Q_j}_{\text{Structural effect}} + \underbrace{\Delta S_j Q_j^0}_{\text{Residue effect}} + \underbrace{\Delta S_j Q_j^0}_{\text{second-order effect}} \quad (5)$$

$$\Delta q_j = \underbrace{S^0 \Delta Q_j}_{\text{Growth effect}} + \underbrace{[S_j^0 \Delta Q_j - S^0 \Delta Q_j]}_{\text{Market distribution effect}}$$

$$+ \underbrace{\Delta S\, Q_j^0}_{\text{Pure residue effect}} + \underbrace{[\Delta S_j Q_j^0 - \Delta S\, Q_j^0]}_{\text{Static structural residual effect}} \quad (6)$$

$$+ \underbrace{[(\frac{Q^1}{Q^0} - 1)\, \Delta S_j Q_j^0]}_{\text{Pure second-order effect}} + \underbrace{[\Delta S_j \Delta Q_j - [(\frac{Q^1}{Q^0} - 1)\, \Delta S_j Q_j^0]}_{\text{dynamic structural residual effect}}$$

Where q_j is the quantity of Thai frozen shrimp export to the European Union15 j. S is the market share of Thai frozen shrimp export in the global market. S_j is the market share of Thai frozen shrimp export in the European Union15 j. Q_j is total quantity of world's frozen shrimp export to the European Union15 j. Q is total quantity of world's frozen shrimp export. Δ represents the change during the two periods. Superscript 0 and 1 denote the base year 2014 and final year 2015.

[14] The change in quantity of export has already absorbed the effect of price change. Therefore, if value data are used, the unit value can disturb interpretation of the CMS's estimation.

As illustration in Eq. 5, it is first level decomposition. The structural effect indicates Thailand's export growth caused by change in general market effect. The residual effect indicates Thailand's export growth caused by change in Thailand's general competitiveness in EU market. The second-order effect indicate the change in export due to the interaction between the change in the Thai frozen shrimp export and the change in demand for frozen shrimp's both in the world and EU market. Meanwhile, according to Eq. 6, it is second level decomposition derived from the first level. The structural effect is split into growth and market effects. The growth effect measure the part of the export growth of Thai frozen shrimp export caused by the general increase in global exports. The market distribution effect indicates whether Thailand has concentrated its export of frozen shrimp on EU market whose demand is relatively more rapidly growing than global market. The residue effect is split into pure residue and static structural residual effects. The pure residue effect measure the change in Thai frozen shrimp export due to a change in its competitiveness in global market. Whereas, the static structural residual effects reflects the changes in Thai frozen shrimp export due to a change in its competitiveness in EU market. Finally, the second-order effect is also split into pure second-order and dynamic structural residual effects. The pure second-order indicates whether the change in Thai frozen shrimp export is adaptable to the demand change in the world, the while dynamic structural residual effect indicates that in EU market.

RESULTS

Impact of EU's GSP withdrawal on Thai frozen shrimp export

Due to the EU's GSP suspension on Thailand, the tariff preference for Thai frozen shrimp export to the European Union was deleted and the duty of the product consequently tripled from 4.2 to 12 percent. The prices of Thai frozen shrimps in EU15 were marked up by percentage change in tariff rate, and Thai frozen shrimp exporters suffered with a decrease in volume of export. Table 1 illustrates that the increase in tariff due to the EU's GSP withdrawal has a negative impact on volume of Thai frozen shrimp export and the volume of export in 2015 decreased by - 6,202 metric tons, accounting for -68.01 percent of volume of export in 2014 or -64,227,912 euros[15] or -2,441,302,935 baths[16] in term of value. The decrease was derived from trade contraction and trade diversion effects. The increase in duty leaded to trade contraction causing EU15's consumer to swift to purchase more on frozen shrimp produced in EU instead of frozen shrimp imported from Thailand, accounting for -977 metric tons or -15.76 percent of total effects. Simultaneously, this also leaded to trade diversion causing the European Union 15 to divert their imports to Thailand's trade rivals still eligible for preferential tariff such as Ecuador, India, Vietnam, and Bangladesh, accounting for -5,225 metric tons or -84.24 percent of total effects. Among the Thailand's competitors, the European Union has mostly diverted its import to Bangladesh, followed by Vietnam, Ecuador and India accounting for -1,728, -1,674, -943 and -880 metric tons, respectively. Additionally, it is somewhat surprised that the elasticity of substitution and amount of trade diversion towards Bangladesh are higher than Ecuador, India, and Vietnam, even though the species of shrimp exported by Thailand and Bangladesh are noticeably different[17]. This might be due to the

[15] Averaged unit valued of Thai frozen shrimp export to EU15 in 2015 was 10,356 euros per metric ton
[16] 1 euro = 38.01 baths (the exchange rate on average in 2015)
[17] The species of shrimp, mostly cultured and exported by Thailand is white leg shrimp, while Bangladesh is tiger prawns [19].

degree of concentration of Bangladesh's frozen shrimp export on the same market destinations in EU market such as United Kingdom, Germany and France [3]. On the other hand, there was no trade diversion towards to Indonesia derived from insignificant in elasticity of substitution implying that Thai frozen shrimp and Indonesia's frozen shrimp are not substituted each other. This would be due to the fact that Thai shrimp is more reliable than Indonesia shrimps in term of quality standards since Indonesian shrimps have been usually detected to be infected by viruses and contaminated by prohibited antibiotics [20, 21].

Table 1. Results of estimated elasticities and the impact of tariff increase on Thai frozen shrimp export in 2015

[Unit : metric tons]

Countries	Base year export: 2014	B_1	C_i	Trade contraction	Trade diversion towards
Thailand	9,107	-1.42*	-	-977	-
Ecuador	92,907	-	-1.52**	-	-943
India	79,270	-	-1.44**	-	-880
Bangladesh	36,351	-	-3.17*	-	-1728
Vietnam	46,234	-	-2.94*	-	-1674
Indonesia	6,514	-	-1.11	-	0
Total trade contraction				-977	
Total trade diversion					-5225
Total trade effects		- 977 + - 5225 = - 6,202			

Note: 1. B_1 is price elasticity of EU's import demand for Thai frozen shrimp
2. C_i is elasticities of substitution
3. *,**,*** Significant at the 99%,95% ,90% level.
4. Ecuador's status as EU's GSP beneficiary was extended in 2015 because FTA agreements between EU and Ecuador have been already done and it is in the process. Thereby, Ecuador is still enjoying the tariff preference.

The change in competitiveness of Thai frozen shrimp export

Table 2 presents the result of CMS analysis of Thai frozen shrimp export to the European Union during 2014 – 2015 in accordance with the first level decomposition established in empirical model. According to the table, it can be seen that the actual volume of Thai frozen shrimp export to the European Union during the periods decreased by 6236 metric tons. This was caused by the negative structural effect, negative residue effect and positive second order effect, quailing to -243, -6,157 and 164 metric tons, accounting for -3.89, -98.73 and 2.63 percent of actual export change,

respectively.

More specifically, at the second level decomposition, the observations can be made as follows. Firstly, the growth effect is negative and equivalent to -10.38 **%** while the market effect is positive and equivalent to 6.48 **%**. This implies that during 2014 –2015, the demand for frozen shrimp in world market was shrinking while that in EU market was expanding and Thailand has concentrated in its frozen shrimp export to EU market slightly growing faster than world market.

Secondly, the pure residue is negative and equivalent to -5.98% indicating that Thailand has lost its competitive edge for frozen shrimp export in global market. This was attributed to the fact that Thailand has seen its export share fall in EU, Japan and Canada markets. Meanwhile, statistic residue is negative and equivalent to -92.75% indicating that during 2014 – 2015, Thailand had significantly lost its competitiveness for frozen shrimp export in EU markets. Additionally, when comparing both static structural residue effect and pure residue effect, it can be seen that the negative static structural residue effect is noticeably larger than negative pure residue. This implies that there was a significant situation, occurring and remarkably hampering the competitiveness of Thai frozen shrimp export in EU market. This was attributed to the impact of tariff privilege cut due to EU's GSP suspension on Thai frozen shrimp export, becoming effective at the beginning of 2015.

Finally, the pure second-order effect is positive and equivalent to 2.95%, implying that Thai frozen shrimp export during 2014 – 2015 was adaptable to the change in the world's demand. On the other hand, the dynamic structural residue effect is negative and equivalent to -0.30%, indicating that Thai frozen shrimp export was inadaptable to the change in the EU's demand. In other words, this implies that Thailand was unable to increase its share in EU market while the EU's demand for frozen shrimp was more rapidly increasing than the world's.

Summary and Policy recommendations

As a result of EU's Generalized System of Preference (GSP) withdrawal on Thailand, the preferential tariff for Thai frozen shrimp export to EU15 had been cancelled. Thai frozen shrimp export to EU15 had to pay higher tariff rate at MFN rate, while her major competitors such as Ecuador, India, Vietnam, Indonesia and Bangladesh have still paid for tariff rate at GSP rate which is lower than MFN rate. The prices of Thai frozen shrimp in EU15 were marked up by percentage change in tariff rate and Thai frozen shrimp exporters suffered with a decrease in volume of export derived from trade contraction and trade diversion. In addition, the cancellation obviously caused Thailand to lose its competitiveness to its trade rivals in EU market. This becomes a challenge for Thai shrimp export industry how to stand or maintain its share in the market without EU's GSP.

To promote Thai frozen shrimp export and rise up the competitiveness, the following recommendations should be actualized. Firstly, the government should find a remedial measure for suffered exporters as they suffered from the decline in export. Secondly, because the elasticity of EU's import demand and elasticities of substitution are significantly elastic, improvement of production cost management and transportation should be done in order to increase the pricing competitiveness. The quality improvement consistent with the regulations should be encouraged as several importing countries in EU also trends to set the complicated regulations and trade barriers which govern Thai frozen shrimp to their market. Fourthly, development of traceability system together with creating a good image on dealing with IUU fishing particularly in the issue involved in human and workers' rights[18] should be seriously incessantly enforced, because they are much serious and attributed to EU's trade bans. Finally, government should expedite negotiation of FTA

[18] Thai frozen shrimp exported to EU have ever almost been banned as Thai shrimp was raised by fish meal associated with illegal labors.

agreement between Thailand and EU which has not been signed yet in order to grab their tariff advantages of Thai frozen shrimp. Future studies are recommended to study the impact of EU's GSP withdrawal in long run as well as other negative or positive effects derived from EU's GSP suspension to be additional and useful guideline for exporters and government's policy implementation.

Table 2. Result of CMs analysis of Thai frozen shrimp export to EU15 during 2014 -2015

[Unit : metric tons]

Effect components	Change in Volume of exports	Percent %
Structural Effect	**-243**	**-3.89**
Growth Effect	-647	-10.38
Market Distribution Effect	404	6.48
Residue Effect	**-6,157**	**-98.73**
Pure Residue Effect	-373	-5.98
Static Structural Residue Effect	-5,784	-92.75
Second-Order Effect	**164**	**2.63**
Pure Second-Order Effect	184	2.95
Dynamic Structural Residue Effect	-19	-0.30
Change in actual volume of exports	**-6,236**	**100.00**

ACKNOWLEDGEMENT

I would like to thank the department of agricultural & resource economics and graduate school at Kasetsart University for kindly support. Moreover, I am also especially thankful to Dr. Nuttapon Photchanaprasert, my advisor, for very helpful recommendations and valuable advices.

REFERENCES

[1] Department of International Trade Promotion of Thailand. Shrimp Industry Report. 2015. Accessed Octorber 20, 2016. www.ditp.go.th/contents_attach/93306/93306.pdf

[2] Office of agricultural economics. The value of export of 10 important agricultural commodities, 2016. Accessed Octorber 20, 2016. http://www.oae.go.th/oae/report/export_import//exp_topten.php?imex=2

[3] Trade Map. Trade statistics for international business development. 2016. Accessed November 20, 2016. http://www.trademap.org/

[4] UNCTAD. Trade statistics for international business development. 2015. Accessed November 27, 2016. http://unctad.org/en/PublicationsLibrary/itcdts bmisc25rev4_en.pdf

[5] The European Commission. "REGULATION (EU) No 978/2012 (2012)." Official Journal of the European Union, (2012).

[6] The European Commission. “COMMISSION DELEGATED REGULATION (EU) No 1421/2013.” Official Journal of the European Union, (2013).

[7] Viner, Jacob. "The Customs Union Issue. Carnegie Endowment for International Peace." New York (1950).

[8] Baldwin, Robert E., and Tracy Murray. "MFN tariff reductions and developing country trade benefits under the GSP." The Economic Journal 87, no. 345 (1977): 30-46.

[9] Sapir, Andre. "Trade benefits under the EEC generalized system of preferences." European Economic Review 15, no. 3 (1981): 339-355.

[10] Choudhry, Sonam, Murali Kallummal, and Poornima Varma. "Trade Creation and Trade Diversion in the India-Sri Lanka Free Trade Agreement: A Sector Specific Analysis." Journal of Economic Policy and Research 8, no. 1 (2012): 36.

[11] Mondal, Bitan, and Smita Sirohi. "Impact of ASEAN-India free trade agreement on Indian dairy trade: A simulation analysis." Economic Affairs 61, no. 3 (2016): 461.

[12] Clague, Christopher K. "The trade effects of tariff preferences." Southern Economic Journal (1972): 379-389.

[13] Jammes, Olivier, and Marcelo Olarreaga. "Explaining Smart and GSIM." The World Bank (2005).

[14] Richardson, J. David. "Some sensitivity tests for a" constant-market-shares" analysis of export growth." The Review of Economics and Statistics (1971): 300-304.

[15] Jepma, Catrinus. "Extensions and application possibilities of the constant market shares analysis. The case of the developing countries' export." (1986).

[16] Ahmadi-Esfahani, Fredoun Z., and Paul H. Jensen. "Impact of the US-EC price war on major wheat exporters' shares of the Chinese market." Agricultural Economics 10, no. 1 (1994): 61-70.

[17] Ahmadi-Esfahani, Fredoun Z. "Wheat market shares in the presence of Japanese import quotas." Journal of Policy Modeling 17, no. 3 (1995): 315-323.

[18] Duan, Yufeng. "Ex-post competitiveness assessment of Alberta agri-food exports in target products and markets: 1988-96." (1999).

[19] Sustainable Fisheries Partnership (SFP). Report on the Shrimp Sector: Asian Shrimp Trade and Sustainability. 2016. Accessed, 15 April, 2017. https://www.sustainablefish.org/

[20] Nabin, Munirul H., Xuan Nguyen, and Pasquale M. Sgro. "Technology transfer, quality standards, and North–South trade." Review of International Economics 21, no. 4 (2013): 783-796.

[21] Wati, Lina Asmara, Wen-I. Chang, and Mochammad Muslich Mustadjab. "Competitiveness of Indonesian shrimp compare with Thailand shrimp in export market." WACANA, Jurnal Sosial dan Humaniora 16, no. 1 (2013): 24-31.

AUTHOR INDEX

iconsci.rmutp.ac.th

Co-organizer

Mahidol University
Royal Thai Army
IBM Thailand Co., Ltd.
Deakin University

Rajamangala University of Technology Phra Nakhon : RMUTP
399 Samsen Rd. Vachira Phayaban Dusit Bangkok 10300 Thailand

02 665 3888
rmutp.ac.th