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Optimisation of functional cabbage drinks with lime juice using Response Surface Methodology (RSM)

(Pengoptimuman formulasi minuman kubis fungsian dengan jus limau menggunakan kaedah gerak balas permukaan)

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Abstract

Functional beverages have become popular due to its appeal to consumers who are seeking specific health benefits in their foods and beverages. Thus, a new functional beverage from cabbage extract with rich alkaloid content was developed. The acceptability of the cabbage drink was with respect to the initial cabbage extract concentrations (5.0%, 12.5% and 20.0%) and lime juice as natural flavour (0%, 2.0% and 4.0%). A central composite design (CCD) was applied to study the response pattern for monitoring sensory quality and acceptability as well as its physico-chemical properties of the formulated cabbage drink. In Response Surface Methodology (RSM), both sensory evaluation with hedonic rating scale and Quantitative Descriptive Analysis (QDA) were used as the response factors to identify the best ratio of cabbage extract to lime juice. Quantitative Descriptive Analysis indicated that 13.93% cabbage extract incorporated with 3.36% of lime juice resulted in a sensory score of 5.37, 5.04 and 5.36 for taste, aroma and overall acceptability respectively. The addition of juice also successfully improved the taste of the cabbage drink. The pH and total soluble solid for the optimum formulation was 2.67 and 9.62 °Brix respectively. The formulated cabbage drink has further raised the functional value of the product through alkaloid content, particularly amounting to 518.4 ppm with 81% antioxidant activity measured via DPPH assay.

Keywords: functional beverage, cabbage juice, lime juice

Introduction

Cabbage, scientifically known as *Brassica oleracea*, represents one of the major vegetable crops grown worldwide constituting an important part of a well-balanced diet. It is a native of the Mediterranean region and southwestern Europe, extending northward to southern England. It can be grown easily in cold weather, requires moist soil and can tolerate maritime exposure. Horticultural selection of the species has led to the development of a

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number of cultivars and although essentially temperate, *B. oleracea* cultivars are currently grown for food everywhere (Ferreres et al. 2007).

Several studies have found that consumers need a constant supply of phytochemicals containing plants to achieve optimal health benefits. Different plants have distinct compounds with several chemical structures, thus offering different protective mechanisms at different levels (Chu et al. 2002; Ninfali and Bacchioca 2003; Ferreres et al. 2007). *Brassica oleracea* is known to contain some potential health promoting compounds that may reduce the risk of diseases. Examples of these are anticarcinogenic compounds such as vitamin C and E and phenolic compounds that includes glucosinolates, phenolic acids, flavonols and anthocyanidins (Mattila and Helistrom 2007; Park et al. 2014; Park et al. 2014a and b). Among all the phytochemicals, only the glucosinolates group exists dominantly in the Brassicaceae species. The glucosinolates group consists of nitrogen and sulfur molecules attached together with glucopyranosyl moiety. Upon cellular disruption, glucosinolates are hydrolysed to various bioactive breakdown products by the endogenous enzyme myrosinase. Isothiocynates and indole glucosinolate metabolites are two major groups of autolytic breakdown products of glucosinolates (Francisco et al. 2009). Sinigrin is one of the major glucosinolate constituents that is widely used as a standard marker for Brassicaceae species verification as reported by Mazumder et al. (2016a) and Ishida et al. (2014). Sinigrin has also been reported to possess a lot of biological activities (Mazumder et al. 2016b), especially antioxidants as reported by Oh et al. (2016) and Fernandez-Leon et al. (2014).

The preliminary sensory study found that lime extract was significantly accepted by the panelist as a natural flavour to be used in the cabbage drink compared to melon juice and mixed fruit juice. Lime is a good source of vitamin C and often used to accent the flavour of foods and beverages. Lime contains unique flavonoid compounds called flavonol glycosides that have antioxidant activity and anticancer properties (Ojinnaka and Ojimelukwe 2012). Previously, the finding showed that it also has higher Vitamin C, A, calcium, iron and phosphorus compared to lemon.

Response Surface Methodology (RSM) was developed by Box and collaborators in the 1950's (Burns et al. 2006; Bezerra et al. 2008). It is a powerful mathematical model with a collection of statistical techniques wherein interactions between multiple process variables can be identified with fewer experimental trials. It has been widely used since the early 1980's to examine and optimise the operational variables for experiment designing, model developing and factors and conditions optimisation (Cheynier et al. 1983). In food processing, especially in the field of beverage, optimum conditions is one of the most critical stages. Statistical methodologies involved mathematical models for designing and analysing the process results (Bas et al. 2007; Tan et al. 2012).

The main purpose of this study was to optimise certain parameters using RSM application in order to produce an optimal formulation for the cabbage drinks based on percentage of cabbage extract and lime juice with alkaloid content (sinigrin) in order to obtain antioxidant rich drinks.

Materials and methods

Sample preparation

Cabbage extract was prepared according to Daud et al. (2011). The cabbage (1.0 kg) was cut into small particle size (2.0 mm) in order to increase surface contact. The cabbage was boiled with four litres of water for 5 h in a distillation system in order to separate the odour from the main extract. After that, the main cabbage extract was allowed to cool at room temperature for 2 h.

Determination of sinigrin

The cabbage drinks were processed according to the flowchart as outlined in *Figure 1*. The sinigrin standard was purchased from Sigma (85440) in the form of its potassium salt. The sinigrin content in the extract was analysed using a spectrophotometer (UV-visat wavelength 227 nm) that is normally used for analysis of sinigrin as reported in many previous studies (Gallaher et al. 2012; Malabed et al. 2014; Vastenhout et al. 2015). Different concentrations of sinigrin standard solutions were prepared starting from 1,000 ppm and further diluted by half in series to form six serial dilutions of standards. A calibration curve of concentration vs. absorbance was constructed in order to determine the sinigrin content in the cabbage drinks (*Figure 7b*).

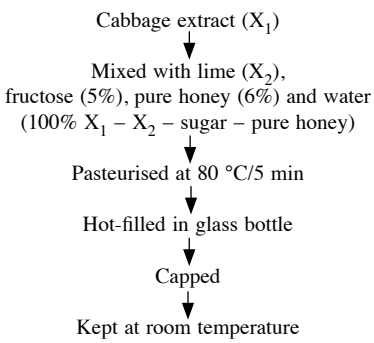

Figure 1. Preparation of cabbage drinks

Experimental design

Response Surface Methodology (RSM) involving the sensory evaluation with hedonic rating scale QDA was used to identify the best ratio of cabbage extract to lime juice. The two variable factors and their levels were studied as follows; a) percentage of cabbage extract (5 – 20%) and b) percentage of lime juice (0 – 4%) (*Table 1a*). The factor was coded as –1 and +1 respectively, to represent the minimum and maximum levels, whereas the 0 (zero) represents the central point of the factorial design as shown in *Table 1b*.

The complete design consisted of 13 experimental runs (samples) which included 5 replications of the centre point (*Table 2*). The samples were subjected to sensory tests, determination of sinigrin content (ppm) and physico-chemical analysis. Models with greater value for coefficient of

Table 1a. Factors and levels for response surface study

Factor	Low level (–1)	High level (–1)
X_1 – Cabbage extract (%)	5	20
X_2 – Lime juice (%)	0	4

Table 1b. Experimental design for the formulation of cabbage drinks

Std. run no.	Coded independent variables		Decoded independent variables	
	X_1	X_2	Cabbage extract (%)	Lime juice (%)
1	–1	–1	5.0	0.0
2	1	–1	20.0	0.0
3	–1	1	5.0	4.0
4	1	1	20.0	4.0
5	–1	0	5.0	2.0
6	1	0	20.0	2.0
7	0	–1	12.5	0.0
8	0	1	12.5	4.0
9	0	0	12.5	2.0
10	0	0	12.5	2.0
11	0	0	12.5	2.0
12	0	0	12.5	2.0
13	0	0	12.5	2.0

Table 2. The predicted model for sensory data at formulation phase

Final equation	R^2	R^{2adj}	F value	Lack of fit
Taste = 3.620 + 0.107 X_1 + 0.668 X_2 – 4.766E – 003X_1^2 – 0.136 X_2^2 + 0.010 X_1X_2 **	0.8071	0.6694	5.86	Not significant
Aroma = 3.287 + 0.038X_1 + 0.838X_2 – 6. 896E – 004 X_1^2 – 0.116 X_2^2 – 3.333E – 003 X_1X_2 **	0.9567	0.9258	30.93	Not significant
Overall acceptability = 3.036 + 0.183 X_1 + 0.589 X_2 – 8.398E – 003 X_1^2 – 0.093 X_2^2 + 1.0E – 002 X_1X_2 **	0.8084	0.6715	5.91	Not significant
Sinigrin = 560.472 – 4.571 X_1 – 17.113 X_2 + 0.168 X_1^2 + 3.176 X_2^2 + 0.227 X_1X_2 *	0.8893	0.8102	11.24	Not significant

*Significant at p <0.01
**Significant at p <0.05

determination (R^2) and adjusted coefficient of determination (R^{2adj}) are significant in the model design but not significant (p <0.05) in lack of fit. These are some of the important points to indicate a good predictive model for the responses.

Statistical analysis

The Design-Expert 6.0.8 (DX6) Statistical Programme (Stat-Ease, Inc., 2000) was used to design the experimental work for RSM. This software was also used to conduct the regression and ANOVA analysis of the data obtained.

Proximate analysis

Dietary fibre, carbohydrate, calories, calcium, sodium and iron contents of the sample were determined according to AOAC method (2000). The methods of determining protein, fat and sugar content were based on Kirk et al. (1991). Vitamin A and vitamin C was determined using the method of analysis for Sullivan and Carpenter (1993).

DPPH assay

The assay was based on the method described by Daud et al. (2017). The antioxidant potential of the formulated cabbage drink was assessed based on their scavenging activity against the stable 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical. A solution of DPPH was prepared by dissolving 5.0 mg DPPH in 2 ml of methanol and the solution was kept in the dark at room temperature. A hundred and ninety five µl of formulated cabbage drink was added to a 96-well microtitre plate. After that, 5 µl of methanolic DPPH solution was added. The plate was shaken to ensure thorough mixing before being placed in the dark and wrapped with aluminum foil. After 30 min, the optical density of the solution was analysed using an ELISA reader (Spectramax Plus, Molecular Devices) at a wavelength of 517 nm. Percentage inhibition by sample treatment was determined by comparison with methanol treated control group. Quercetin (Sigma, USA) was used as a positive control. In the DPPH method, the antioxidant activity is described by percent inhibition:

$$\text{Percent inhibition} = [(\text{Abs control} - \text{Abs sample})/\text{Abs control}] \times 100$$

Sensory evaluation

Preference test

The sensory tests were carried out on a laboratory scale according to Stone and Sidel (2004) at MARDI Johor Bahru with 10 panelists and the order of presentation was randomised as described by Saniah and Rahani (2011). The samples of cabbage drinks were presented in glasses with three digits random numbers. Sensory attributes of the preference test were based on degree of liking (DOL), taste, aroma of cabbage and

overall acceptability using a 7-point hedonic scale (1 = Dislike extremely; 4 = Neither like nor dislike; 7 = Like extremely) as described by Meilgaard et al. (1999).

The QDA, using 5-inch line scales (Saniah and Rahani 2011) with descriptors anchored at both ends, was applied to describe the intensity of sweetness, sourness and cabbage aroma (undesirable aroma). The scales were anchored from left with 'not sweet' to 'very sweet', 'not sour' to 'very sour' and 'no cabbage aroma' to 'very hard cabbage aroma' for the sensory attributes of sweetness, sourness and cabbage aroma respectively. The panelists were pre-screened and trained using the descriptive testing method (Meilgaard et al. 1999). The trained panelists evaluated the factors by marking across the scales for intensity and the markings were converted to numerical values by measuring from left to right of the line (Stone et al. 1974) in the sensory room.

Results and discussion

Model fitting from RSM

Using RSM, the relationship among the variables, i.e. cabbage extract and lime juice were expressed mathematically in the form of polynomial model, which gave response as a function of relevant variables. The independent and dependent variables were fitted to the second order model equation and were examined for the goodness of fit. The analyses of variance (ANOVA) were performed to test the adequacy of the model. The parameters found to be significant at the probability level (P) equal to or less than 5% were considered in order to obtain the fitted models to predict the responses. In this case, the responses for the DOL for taste, aroma and overall acceptability were significant at the 0.05% level. The coefficient of determination (R^2) was suggested to determine a good fit model. The R^2 should be achieved at the minimum value of at least 80% as reported by Joglekar and May (1991). Other researchers like Henika (1982) considered a minimum value of 85% for R^2 as a very good model to predict the sensory score. The fitted model developed from this study was used to predict sensory score for cabbage drinks produced at the different cabbage extract-lime juice levels and the experimental sensory score obtained were found to be close with the predicted sensory score with a value of R^2 >80% (*Table 2*). The sensory evaluation results showed that the models for DOL for taste (p <0.05), aroma (p <0.01), sinigrin (p <0.01) and overall acceptability (p <0.05) had achieved the coefficients of determination (R^2) of 0.8071, 0.9567, 0.8893 and 0.8084 respectively (*Table 2*). This indicated that nearly 80 – 90% of the total variation can be predicted by the models. All models also presented insignificance in the lack of fit (p >0.05), which is desirable for the model.

Sensory evaluation

Examination of the Fit Summary output also revealed that the linear model was statistically significant for DOL for taste, aroma and overall acceptability. The cabbage drink formulation without the addition of lime juice received a low mean score of below 4.0 which was closer to 'slightly dislike' for taste, aroma as well as overall acceptability (*Table 3*). However, the score for aroma was high when the percentage of lime juice increased in the cabbage drink formulations (*Figure 2*) where the DOL score for aroma was >5.0.

Figure 3 illustrates that the predictive model generated by the mean score indicated the optimised region for taste, whereby a sample with medium level of cabbage extract (12.5 – 14.0%) and lime juice (3.0 – 3.5%) was predicted to be the best liked.

The overall acceptability was positively associated with the amount of cabbage extract and lime juice (*Figure 4*) with quadratic effects at the coefficients of determination (R^2) of 0.8084. It can be observed from QDA test for sweetness that indicated the panelists preferred the less sugar or 'not very sweet' drink. *Figure 5* illustrates the possibility of optimised

Table 3. The mean sensory scores, pH, TSS and sinigrin content for the response variables of cabbage drinks

Run	DOL of taste	DOLof aroma	Overall taste acceptability	pH	TSS (°Brix)	Sinigrin (ppm)	Intensity of		
							Sweetness	Sourness	Cabbage aroma
1	3.8	3.4	3.7	3.7	9.2	544.8	2.5	0.7	1.5
2	3.9	3.8	3.6	2.31	9.2	538.6	2.0	0.8	1.6
3	4.7	4.9	4.8	2.5	9.4	528.9	1.8	1.9	1.0
4	5.4	5.1	5.3	2.63	9.8	536.3	1.9	3.3	1.0
5	5.2	4.7	4.7	2.68	9.2	519.4	1.6	1.3	1.0
6	4.8	4.75	4.0	2.87	9.6	522	1.9	1.8	2.1
7	4.4	3.7	3.8	2.68	9.0	524.3	2.9	0.5	2.0
8	5.0	4.9	5.1	2.58	9.8	523.6	1.8	2.8	1.8
9	5.2	4.9	4.9	2.8	9.4	520.7	1.5	1.5	1.5
10	5.0	4.5	4.9	2.8	9.4	516.3	1.9	2.0	1.9
11	5.8	4.9	5.7	2.79	9.4	511.6	2.0	2.3	0.7
12	5.1	4.9	5.25	2.79	9.4	516.6	1.5	1.8	1.4
13	5.2	4.8	5.1	2.81	9.6	508.5	1.5	1.6	1.0

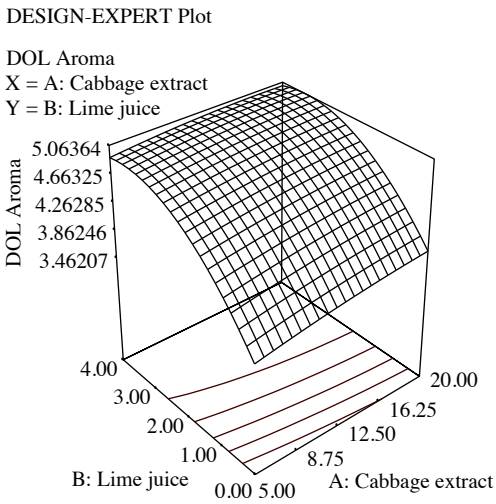

Figure 2. Response surface plot for aroma

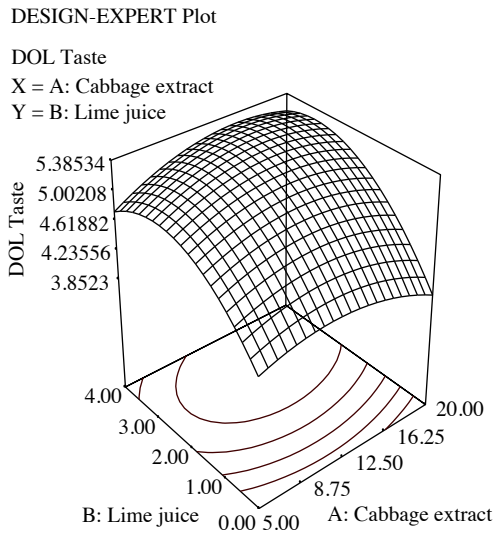

Figure 3. Response surface plot for taste

region for sinigrin content within a range of 513.5 – 520.6 ppm obtained from cabbage extract (12.5 – 14.0%) and lime juice (3.0 – 3.4%). Only a slight difference in pH was observed while the TSS value remained without any changes.

Optimisation of basic formulation and verification of the model

Graphical optimisation function was used to superimpose the contour plots generated from three response surface variables. Superimposing of sensory contour (*Figure 6*) resulted in an acceptable cabbage drink formulation based on the optimised point of three sensory contour plots whereby

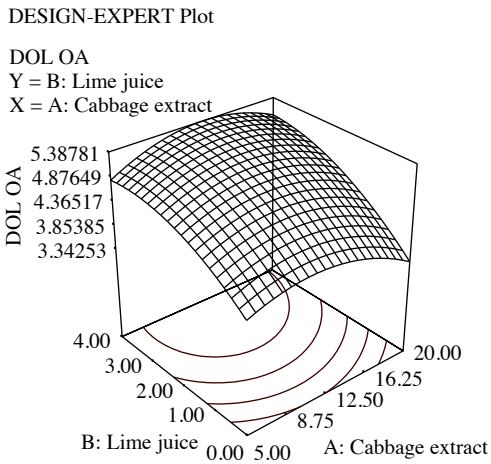

Figure 4. Response surface plot for overall acceptability

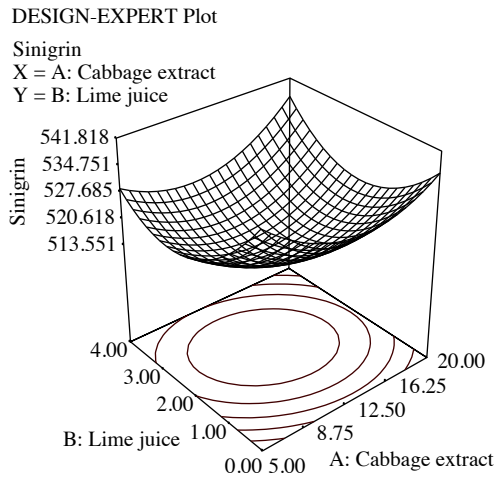

Figure 5. Response surface plot for sinigrin content

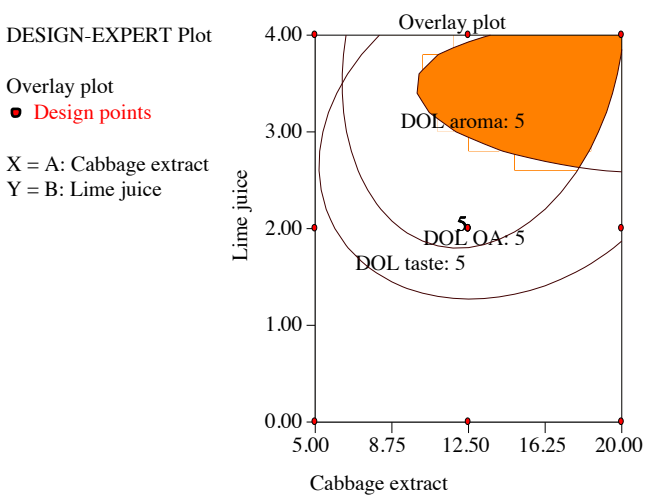

Figure 6. The overlay plot for the three response surface variables of cabbage drinks

the suitable range of cabbage extract and lime juice was shown as feasible region.

The multiple response optimisations were also performed using numerical optimisation functions. In this optimisation study, the percentage of cabbage extract and lime juice were set as 'in range'. It is desirable to have a maximum sensory score for all responses, particularly the degree of liking (DOL) for taste, aroma and overall acceptability. Therefore, the goals for this group of responses were set at 'maximise'. The amount of sinigrin, pH, total soluble solids (TSS) and the intensity of sweetness, sourness and cabbage aroma were set as 'in range'. The possible optimal solution for the percentage and combination of cabbage extract and lime juice in the cabbage drink is based on aroma, taste and overall acceptability as shown in *Table 4*.

From the optimisation model, it was shown that the best DOL for the sensory

test was obtained when 13.93% of cabbage extract was used in combination with 3.36% lime juice. The incorporation of 13.93% of cabbage extract and 3.36% lime juice suggested obtaining the optimum score of sensory attributes evaluated with a desirability of 0.858. The optimum taste, aroma and overall acceptability of this formulated cabbage drink were 5.37, 5.04 and 5.36 or close to 'like slightly' respectively (*Table 4*), as obtained from the optimised point of three sensory contour plots (*Figure 6*).

Analysis of sinigrin

Analysis of sinigrin was carried-out using the UV-vis spectrophotometer at a wavelength of 227 nm. *Figure 7a* shows optimal detection of sinigrin at 227 nm which was in agreement with previous reports by Vastenhout et al. (2015), Malabed et al. (2014) and Gallaher et al. (2012). The calibration curve (*Figure 7b*) was successfully constructed based on concentration of sinigrin and absorbance in order to determine the concentration. The coefficient of correlation (R^2) for sinigrin

Table 4. Possible optimal solutions of cabbage extract drinks

Cabbage extract	Lime juice	Taste	Aroma	Overall acceptability	pH	TSS (°Brix)	Sinigrin (ppm)	Sweetness	Sourness	Cabbage aroma	Desirability
13.93	3.36	5.37	5.04	5.36	2.67	9.62	518.37	1.76	2.50	1.31	0.858 Selected

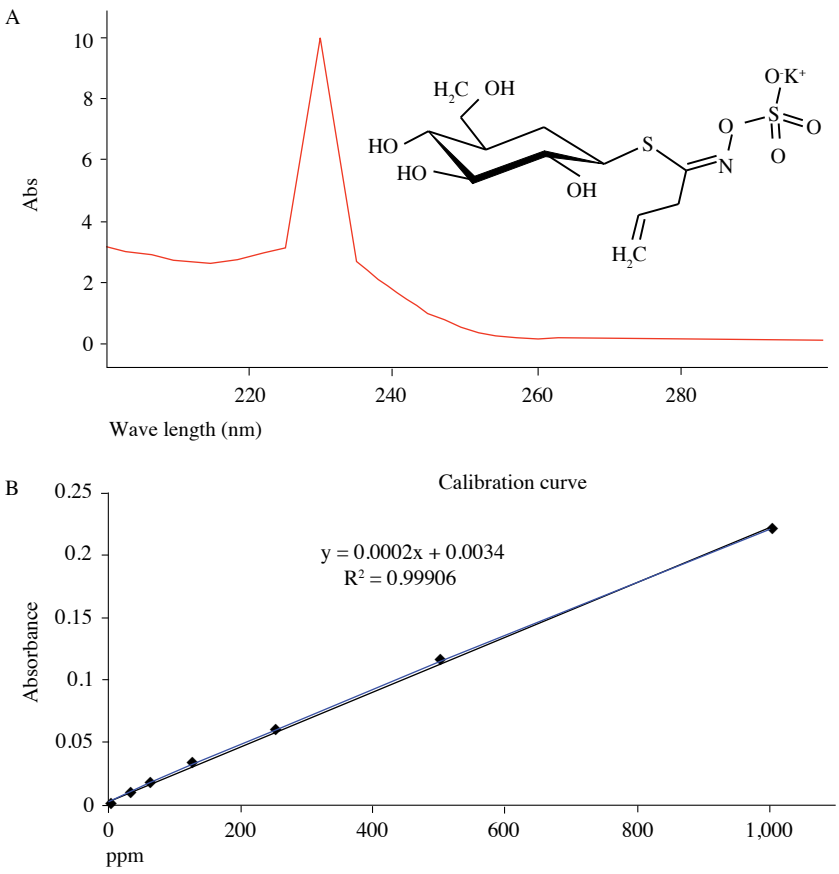

Figure 7. (A) Optimal wavelength for detection of sinigrin and (B) calibration curve for determination of sinigrin using spectrophotometer

at wavelength 227 nm was 0.9991, with an equation of y = 0.0002x + 0.0034.

Nutritional value, antioxidant activity and sinigrin content of cabbage drink

The nutritional value, antioxidant (AOX) activity of formulated cabbage drink and control sample (without cabbage extract) were evaluated and the results are shown in *Table 5*. The antioxidant activity of the formulated cabbage drink was higher than the control with values of 81% and 61% respectively, with sinigrin content of 518.4 ppm in the formulated cabbage drink. Other nutritional values remained similar except for sodium content which increased about 3 folds.

Table 5. Nutritional value and antioxidant activity of cabbage drink

Sample	Cabbage drink	Control sample (without cabbage extract)
Protein (% w/v)	0.1	0.4
Fat content (%w/v)	0.1	0.4
Total carbohydrate (%w/v)	9.1	8.5
Calories (kcal/100 ml)	38	39
Sugar (% w/v)	8.7	8.3
Dietary fibre (% w/v)	ND	ND
Vitamin A (IU/100 ml)	3	24
Vitamin C (mg/100 ml)	ND	ND
Sodium (mg/100 ml)	4.9	1.7
Calcium (mg/100 ml)	2.2	1.8
Iron (mg/100 ml)	0.3	0.2
Antioxidant activity (% DPPH)	81%	61%

Conclusion

From this study, it was found that by increasing the percentage of lime juice in the formulation, we could also increase the aroma, taste and acceptability. The optimum level for combination of cabbage extract and lime juice was found to be at 13.93% and 3.36% respectively, with a pH of 2.67 and TSS of 9.62 °Brix. The overall acceptability score for the formulation based on sensory is 5.36, with 5.37 and 5.04 for taste and aroma respectively. The antioxidant activity for the formulated cabbage drink was found to be higher compared to the control and this could be due to the presence of alkaloid content, especially sinigrin.

The RSM had significantly reduced the time to formulate an optimum cabbage drink formulation. Therefore, based on this study, it is suggested that the models obtained could be used to optimise the formulation process of functional cabbage drinks. Generally, the production of functional cabbage drink could further vitalise the beverage industry in the country and directly benefit the consumers through their consumption of a healthy product.

References

AOAC (2000). Association of official analytical chemists. Official methods of analysis of AOAC international, 17th Edition, Washington DC, USA

Bas, D., Ismail, H. and Boyaci, J. (2007). Modelling and optimization: Usability of response surface methodology. *Journal of Food Enginering* 78: 836 – 845

Bezerra, M.A., Santelli, R.E., Oliveira, E.P., Villar, L.S. and Escaleira, L.A. (2008). Response surface methodology as a tool for optimization in analytical chemistry. *Talanta* 76: 965 – 977

Burns, R.E., Scarminio, I.S. and Neto, B.B. (2006). Statistical Design-Chemometrics, Elsevier, Amsterdam

Cheynier, V., Feinberg, M., Chararas, C. and Ducauze, C. (1983). Application of response surface methodology to evaluate bioconversion experimental conditions. *Journal of Applied and Environmental Microbiology* 45: 634 – 639

Chu, Y.F., Sun, J., Wu, X. and Liu, R.H. (2002). Antioxidant and antiproliferative activities of common vegetables. *Journal of Agricultural and Food Chemistry* 50: 6910 – 6916

Daud, M.N.H., Fatanah, D.N., Abdullah, N. and Ahmad, R. (2017). Evaluation of antioxidant potential of *Artocarpus heterophyllus* L. J33 variety fruit waste from different extraction methods and identification of phenolic constituents by LCMS. *Journal of Food Chemistry* 232: 621 – 632

Daud, M.N.H., Jabit, M.L., Noh, J.M., Normah, A. and Nabilah, M.F. (2011). Identification and isolation of methyl gallate as a polar chemical

marker for *Labisia pumila* Benth. *Journal of Tropical Agricultural and Food Science* 39(2): 279 – 284

Fernandez-Leon, A.M., Lozano, M., Gonzalez, D., Ayuso, M.C. and Fernandez-Leon, M.F. (2014). Bioactive compounds content and total antioxidant activity of two savoy cabbage**s.***Czech Journal of Food Sciences* 32(6): 549 – 554

Ferreres, F., Sousa, C., Valentao, P., Seabra, R.M., Pereira, J.A. and Andrade, P.B. (2007). Tronchuba cabbage (*Brassica oleracea* L. var. costata DC) seeds: Phytochemical characterization and antioxidant potential. *Journal of Food Chemistry* 101: 549 – 558

Francisco, M., Moreno, D.A., Cartea, M.E., Ferreres, F., Viguera, C.G. and Velasco, P. (2009). Simultaneous identification of glucosinolates and phenolic compounds in *Brassica rapa*. *Journal of Chromatography A* 1216: 6611 – 6619

Gallaher, C.M., Gallaher, D.D. and Peterson, S. (2012). Development and validation of a spectrophotometric method for quantification of total glucosinolates in cruciferous vegetables. *Journal of Agricultural and Food Chemistry* 60: 1358 – 1362

Henika, R.G. (1982). Use of response surface methodology in sensory evaluation. *Food Technology* 36: 96 – 101

Ishida, M., Hara, M., Fukino, N., Kakizaki, T. and Morimitsu, Y. (2014). Glucosinolate metabolism, functionality and breeding for the improvement of Brassicaceae vegetables. *Breeding Science* 64: 48 – 59

Joglekar, A.M. and May, A.T. (1991). Product excellence through experimental design. In: *Food Product Development*. New York: Van Nostrand Reinhold

Kirk, R.S., Sawyer, R. and Egan, H. (1991). Pearson's chemical analysis of foods. 9th Edition, Longman Scientific and Technical, Harlow, Essex, UK

Malabed, R.S., Noel, M.G., Aton, B.C. and Toribio, E.A.F. (2014). Characterization of the glucosinolates and isothiocyanates in mustard (*Brassica juncea* l.) extracts and determination of its myrosinase activity and antioxidant capacity. DLSU Research Congress, March 6 – 8. p. 1 – 7 De La Salle University, Manila, Philippines

Mattila, P. and Helistrom, J. (2007). Phenolic acids in potatoes, vegetables and some of their products. *Journal of Food Composition and Analysis* 20: 152 – 160

Mazumder, A., Dwivedi, A. and Du Plessis, J. (2016a). Sinigrin and its therapeutic benefits. *Molecules* 21(4): 1 – 11

Mazumder, A., Dwivedi, A., Du Preez, J.L. and Du Plessis, J. (2016b). *In vitro* wound healing and cytotoxic effects of sinigrin-phytosome complex. *International Journal of Pharmaceutics* 498: 283 – 293

Meilgaard, M., Civille, G.V. and Carr, B.T. (1999). Sensory evaluation technique 3rd Ed. USA. CRC press LLC

Ninfali, P. and Bacchioca, M. (2003). Polyphenols and antioxidant capacity of vegetables under fresh and frozen conditions. *Journal of Agricultural and Food Chemistry* 51: 2222 – 2226

Oh, S., Kim, K. and Choi, M. (2016). Antioxidant activity of different parts of Dolsan Leaf Mustard. *Food Science and Biotechnology* 25(5): 1463 – 1467

Ojinnaka, M.C. and Ojimelukwe, P.C. (2012). Effect of salt and lime on *Bacillus* fermented castor oil bean condiment. *Global Research Journal of Agricultural and Biological Science* 3(5): 381 – 389

Park, S., Arasu, M.V., Jiang, N., Choi, S.H., Lim, Y.P., Park, J.T., Al-Dhabi, N.A. and Kim, S.J. (2014). Metabolite profiling of phenolics, anthocyanins and flavonol in cabbage (*Brassica oleracea* var capitata). *Journal of Industrial Crops and Product* 60: 8 – 14

Park, S., Arasu, M.V., Lee, M.K., Chun, J.H., Seo, J.M., Al-Dhabi, N.A. and Kim, S.J. (2014b). Analysis and metabolite profiling of glucosinolates, anthocyanins and free amino acids in inbred lines of green and red cabbage (*Brassica oleracea L.*). *Journal of Food Science and Technology* 58(1): 203 – 213

Park, S., Arasu, M.V., Lee, M.K., Chun, J.H., Seo, J.M., Lee, S.W., Al-Dhabi, N.A. and Kim,S.J. (2014a). Quantification of glucosinolates, anthocyanins, free amino acids and vitamin C in inbred lines of cabbage (*Brassica oleracea L.*). *Journal of Food Chemistry* 145: 77 – 85

Saniah, K. and Rahani Z.C. (2011). Acceptability and textural quality of Sri Pontian cassava nuggets using Response Surface Methodology quantitative descriptive analysis (QDA). *Journal of Tropical Agricultural and Food Science* 39(2): 131 – 139

Sullivan, D.M. and Carpenter, D.E. (1993). Methods of analysis for nutrition labeling. p. 33 – 83, AOAC International, Arlington, VA

Stone, H. and Sidel, J. (2004). *Sensory evaluation practices*. Orlando: Academic Press

Stone, P.H., Quirk, W.J. and Somerville, R.C.J. (1974). The effect of small-scale vertical mixing of horizontal momentum in a general circulation model. *Monthly Weather Review* 102: 765 – 771

Tan, Q.L.P., Kieu, X.N.T., Kim, N.H.T. and Hong, X.N.T. (2012). Application of response surface methodology (RSM) in condition optimization for essential oil production from *Citrus latifolia*. *Journal of Food Agricultural* 24(1): 25 – 30

Vastenhout, K.J., Tornberg, R.H., Johnson, A.L., Amolins, M.W. and Mays, J.R. (2015). HPLC-based method to evaluate kinetics of glucosinolate hydrolysis by sinapis alba myrosinase. *Analytical Biochemistry* 465: 105 – 113

Abstrak

Minuman fungsian telah menjadi popular berikutan pemintaan pengguna yang mencari khasiat kesihatan yang spesifik dalam makanan dan minuman mereka. Sehubungan dengan itu, minuman fungsian baru daripada ekstrak kubis yang kaya dengan kandungan alkaloid telah dihasilkan. Penerimaan minuman berfungsi kubis terhadap kandungan sebenar ekstrak kubis (5%, 12.5% dan 20.0%) dan jus limau sebagai perisa semulajadi (0%, 2.0% dan 4.0%) telah dikaji. *Centre Composite Design* (CCD) telah diaplikasikan bagi mengkaji corak reaksi pemantauan kualiti sensori dan penerimaan begitu juga dengan fiziko-kimia minuman kubis berformulasi. *Response Surface Methodology* (RSM) kombinasi kedua-dua penilaian sensori dengan skala penentuan hedonik dan *Quantitative Descriptive Analysis* (QDA) telah digunakan sebagai faktor respon bagi menentukan nisbah terbaik ekstrak kubis kepada jus limau. Analysis QDA menunjukkan penggunaan 13.93% ektrak kubis ditambah dengan 3.36% jus limau menghasilkan rasa, aroma dan penerimaan keseluruhan pada tahap optimum dengan skor sensori masing-masing 5.37, 5.04 dan 5.36. Penambahan jus juga telah berjaya memperbaiki rasa minuman kubis yang terhasil. Nilai pH dan jumlah pepejal larut bagi formulasi optimum masing-masing 2.67 dan 9.62 °Brix. Formulasi minuman kubis telah berjaya meningkatkan nilai fungsian produk melalui kandungan alkaloid pada kadar 518.4 ppm dengan aktiviti antioksida 81% yang dinilai melalui teknik DPPH.