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Optimisation of Spray Drying Conditions of Boiling Extract Liquid of *P. Amboinicus* Lour for Maximum Antioxidant Activity

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ABSTRACT

Background: Optimisation of spray drying of boiling extract liquid of P. amboinicus Lour. encapsulated in maltodextrin for maximum antioxidant activity was conducted using response surface methodology comprising of 22 factorial experiments followed by the steepest ascent method. Fresh samples of P. amboinicus Lour. were randomly collected from Kuantan, Malaysia. The antioxidant activities of spray dried boiling extract encapsulated in maltodextrin were assessed using the 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging assay. **Objective:** The objective was to optimize the spray drying of the boiling extract liquid of P. amboinicus Lour. plant, for maximum antioxidant activity. **Results:** The results showed that the highest yield of antioxidant activity of spray dried boiling extract was obtained when the spray dryer was operated at the combination of levels of operational variables X1 (temperature) = 177.65oC and X2 (concentration of maltodextrin) = 48.65%. **Conclusion:** These findings may have a bearing on the application of spray drying technology in preparing herbal extracts, and may prove useful to the herbs industries.

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INTRODUCTION

Cell damage caused by free radicals appears to be a major contributor to degenerative diseases of aging (Percival, 1998). Antioxidants are nutrients that can lower the activity of free radicals and prevent body cells from being damaged due to the reaction involving free radicals. Antioxidants include vitamin E, vitamin C, zinc, selenium and carotenoids (Packer and Colman, 1999), and can be found in many natural substances–especially vegetables and fruits. Fruits and vegetables have protective effects against cellular damage caused by exposure to high levels of free radicals (Ames *et al*., 1993). Micronutrients in fruits and vegetables provide health benefits to the human diet and are inversely related to cancer risk (Ziegler, 1991). Commonly used synthetic antioxidants such as butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT) are suspected of being responsible for liver damage and carcinogenesis in laboratory animals (Williams *et al*., 1999). Thus there is a great interest in obtaining antioxidants from natural sources (Oktay *et al*, 2003), and utilising them as dietary supplements, food additives, functional food ingredients, pharmaceuticals and cosmetic products (Shahidi and Wanasundara, 1992).

P. amboinicus Lour. from Indonesia has been shown to be a rich source of bioactive compounds (Ong, 2008). In Malay and Chinese herbal medicine, it is used to treat coughs, nausea, headaches and indigestion (Singh and Panda, 2005; Ong, 2008). Consumption of young leaves as salad can increase blood circulation and repair skin texture thus resulting in younger looking skin (Noraida, 2007). The crushed leaves are also used as a local application for headache, insect bite and burns while its fresh juice is used as remedies for asthma, rheumatism and bronchitis (Ong, 2008; Che Aniha, 2008; Singh and Panda, 2005). In Sumatra Island, Indonesia, Bataknese lactating women have consumed the leaves of *P. amboinicus* Lour. in the form of soup during the first month to stimulate breast milk production (Damanik *et al*. 2001; Damanik *et al*., 2004). This soup is also used to control postpartum bleeding and to provide energy (Ong, 2008).

To achieve lower storage costs, higher concentration, and stability of active substances, standardized dried plant extracts need to be produced through new technologies (Oliveira *et al*., 2006). In powders manufacturing,

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spray drying has been adopted due to its ability to create a product with precise quality specifications in continuous operation (Souza and Oliveira, 2006). It turns a solution or slurry into a dried powder in a single step (Chegini and Ghobadian, 2007; Patel *et al*, 2009). A spray drying aid is usually used in the food and other industries to preserve the properties of the products during spray drying (Kha *et al*., 2010). The product to be dried (the load) and the drying aid (usually some sort of modified starch, e.g. maltodextrin) is homogenized as a suspension in water (the slurry). The slurry is then fed into the spray dryer.

In the present work, spray drying has been adopted to produce the dried extract from the boiling extract liquid of the leaves of *P. amboinicus* Lour., and maltodextrin has been chosen as the drying aid. The objective was to optimize the spray drying of the boiling extract liquid of *P. amboinicus* Lour. plant, for maximum antioxidant activity. The Response Surface Methodology (Box and Wilson, 1951) was used, comprising of 22 Factorial Experiments followed by the Steepest Ascent Method. Since temperature affects the activity and stability of compounds due to chemical and enzymatic degradation, and losses by volatility or thermal decomposition (Mouere *et al*., 2001), the combination of levels of independent operational variables X1 (temperature) and X2 (concentration of maltodextrin) are optimised to maximise the antioxidant activity yield of the dried extract as the dependent variable.

MATERIALS AND METHODS

Chemicals:

Maltodextrin was obtained from San Soon Seng Food Industries Sdn. Bhd. (Selangor, Malaysia). Methanol (MeOH) was obtained from Merck (Darmstadt, Germany). All chemicals used were of analytical grade.

Equipments and Instrumentation:

The spray dryer Model SD 06 was supplied by Lab Quip (Cumbria, England). The magnetic stirrer Model EMS-HP 7000 was supplied by Erla (Selangor, Malaysia). The UV-VIS spectrophotometer Model U-1800 was supplied by Hitachi (Tokyo, Japan).

Preparation of P. amboinicus Lour. Plant Boiling Extract Liquid:

Fresh *P. amboinicus* Lour. were randomly collected from Kuantan, Malaysia. Extracts were prepared by the technique of decoction (Ong, 2008). About 150g plant was weighed and washed. Then it was boiled in 1.50L distilled water based on ratio 1:100 (150 g in 1.50 L; concentration of 100 mg/mL). The sample for spray drying was a mixture of individual samples each boiled at a different extraction temperature and for a different extraction time. The antioxidant activity of the mixture was determined prior to feeding it into the spray dryer.

Spray Drying of the P. amboinicus Lour. Plant Boiling Extract Liquid:

The levels of experimental variables for spray drying of the *P. amboinicus* Lour. plant boiling extract liquid in the first experiment were obtained based on initial trials which indicate roughly the ranges of spray drying temperature and % concentration of maltodextrin which would be suitable. These levels were then fitted into a 2^2 factorial design. The levels of experimental variables in the first 2^2 factorial experiments are shown in Table 1.

Table 1: Level of Experimental Variables in the 2^2 factorial experiments.

	Levels of Experimental Variable			
Experimental Variables	α = -1	α = 0	α = +1	Units
Temperature	150	165	180	^{o}C
% concentration of Maltodextrin	10	20	30	%

Before spray drying, the weight of maltodextrin to be added into 200 ml of boiling extract liquid was calculated and then the samples of *P. amboinicus* Lour. plant boiling extract liquid were added with the maltodextrin in the % concentrations as in Table 1. The levels of experimental variables in terms of α are shown in Table 2.

Table 2: Actual values of experimental variables at specified levels of α.

No	Temperature (^{o}C), X_1	Maltodextrin (%), X_2	Volume of Liquid Extract (ml)	Weight of Maltodextrin (g)
1	150 (-1)	10 (-1)	200	20
2	180 (+1)	10 (-1)	200	20
3	150 (-1)	30 (+1)	200	60
4	180 (+1)	30 (+1)	200	60

Each mixture of boiling extract liquid and maltodextrin was stirred thoroughly by using a magnetic stirrer. The mixture was then fed into the spray-dryer. The yield of dried extract powder was then weighed. The dried extract was then diluted to obtain the same concentration of the original boiling extract liquid on which its initial DPPH scavenging activity was measured. The DPPH scavenging activity of dried extract (final antioxidant activity) was measured and compared to initial DPPH scavenging activity of the boiling extract liquid (56.33%). The physical appearance of the dried extract is also reported.

Measurement of DPPH Scavenging Activity:

The antioxidant assay is based on measurement of the loss of DPPH colour at 517 nm after reaction with the test compound and the reaction is monitored by a UV-VIS spectrophotometer. The % of the DPPH remaining is calculated as % scavenging activity:

$$[A_A - A_B) / A_A] \times 100\%. \tag{1}$$

where A_B is absorbance of DPPH* solution with methanol, A_A is absorbance of a DPPH solution with a tested fraction solution.

In this assay, 0.4 ml of extract was diluted with 3.6 ml of distilled water. Then, from the dilution, 0.3 ml was placed in a test tube. 0.7 ml of methanol and then 2 mL of fresh methanolic solution of DPPH (0.004%) were added. These solution mixtures were mixed thoroughly before they were kept in the dark for 30 min. The absorbance was measured at 517 nm using a UV spectrophotometer.

The Method of Factorial Experiments:

There are many types of factorial experiments, one of which is that involving 2 levels, ie the 2^n factorial experiments (Montgomery, 2001). The method of factorial experiments has been designed to allow the effects of a number of experimental variables on the yield to be investigated simultaneously. It gives the "main effects" and the "interactive effects" of changing the experimental variables from a lower level to higher level. The main effect of an experimental variable is defined as the average of the effects of changing its value from the lower level to upper level among all the experiments. It is derived by assuming that the experimental variable is an independent variable and all the variations in its effect are due to experimental errors only. The interactive effects between two or more experimental variables are calculated on the assumption that the experimental variables are not independent but are in fact interacting between them.

The factorial experiments make use of a mathematical method known as the Yates' Method (Yates, 1937) to analyse the main effects and the interactive effects. In giving the main effects and the interactive effects the result of the analysis by Yates' Method (Yates, 1937) also indicate whether the "yield response surface" in the area examined is curved or uncurved, and if it is uncurved, whether it is flat with the respect to the experimental variables or increasing or decreasing with respect to one or more experimental variables and if so, in which direction. In this method, each of the experimental variables that is relevant to the yield is given 2 levels equidistant from the centre point, far enough from each other so that the effect of the difference in levels can be detected in the change in the yields. The levels of the experimental variables used in the 2^2 factorial experiments, at α = (-1) and α= (+1), can be found in Table 1.

The Method of the Path of Steepest Ascent:

The method of the Path of Steepest Ascent involves moving through the experimental region along a path that yields the maximum increases in the response. Taking the centre-point of the first 2^2 Factorial Experiments as the starting point of the ascent, we need to move towards a position P with coordinates ($\bar{x}_{1,}\ \bar{x}_2$) which maximizes the response $y(\bar{x}_{1,}\ \bar{x}_2)$. The change in the value of y depends on the distance of the jump J from the starting point to the position P, given by:

$$J = \sqrt{\ }\ (\bar{x}_{1,}\ \bar{x}_2). \tag{2}$$

These new experimental points can be tested, and the point where the yield differs significantly from that predicted by the linear regression equation describing the area of response surface previously investigated in the 2^2 Factorial Experiments is made the new centre-point around which a new factorial experiment can be constructed.

RESULTS AND DISCUSSION

A 2^2 Factorial Experiment comprise of 4 experiments. The yield expression to be optimized was the antioxidant activity (%) in the spray dried extract. The plan of the 2^2 Factorial Experiments and the untreated results are shown in Table 3.

Table 3: The plan of the 2^2 Factorial Experiments and the Untreated Results.

No	Temperature X_1	Maltodextrin X_2	Spray dried Extract (g)	Antioxidant Activity (%)
1	-1	-1	0.13	-36.20
2	+1	-1	3.5	-30.80
3	-1	+1	4.7	13.75
4	+1	+1	6.0	25.23

From the table above, it shows that the experiment no. 4 (X_1 = +1 and X_2 = +1)

From the Table 3, experiment no. 4 (X_1 = +1 and X_2 = +1) gave the highest antioxidant activity of the spray dried extract while the experiment no. 1 (X_1 = -1 and X_2 = -1) gave the lowest antioxidant activity. The initial antioxidant activity of liquid extract was 56.33%. The experiment no.1, no. 2, no. 3 and no. 4 recorded a decrease of 164.26%, 153.86%, 75.59% and 55.21% respectively.

The physical appearance of the dried extract was also compared among them (Figure 1 to Figure 4). The results showed that the physical appearance of the dried extract produced with the spray dryer operated at X_1 = 180^{o}C and X_2 = 30% was the most powdery among them (Figure 4). The amount of dried extract with the spray dryer operated at X_1 = 150^{o}C and X_2 = 10% was smallest compared to all others. The result also showed that an increase of maltodextrin percentage in the liquid extract caused an improvement of physical appearance (more powdery) and an increase in the amount of dried extract as seen in Figure 3 and Figure 4.

Fig 1: Physical appearance of the dried extract with spray dryer operated at X_1 = 150^{o}C and X_2 = 10%.

Fig. 2: Physical appearance of the dried extract with spray dryer operated at X_1 = 180^{o}C and X_2 = 10%.

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Fig. 3: Physical appearance of the dried extract with spray dryer operated at X_1 = 150^{o}C and X_2 = 30%.

Fig. 4: Physical appearance of the dried extract with spray dryer operated at X_1 = 180^{o}C and X_2 = 30%.

A linear regression between the dependent variable *y* and the independent variables X_1 and X_2 was performed, giving the linear regression coefficient as in Table 4. The coefficient a_2 of X_2 is of the same magnitude as the intercept a_0, indicating that the surface is steep.

Table 4: Regression Coefficients of the Linear Equation for the Response Surface.

Coefficients	Values
b_0	9.745
b_1	2.870
b_2	9.745

Based on these results, a steepest ascent approach was followed, involving the two independent operational variables ie spray drying temperature (X_1) and maltodextrin concentration (X_2).

For the Method of the Path of Steepest Ascent, the calculation is as below:

Calculating $\frac{\partial y}{\partial x_i}$ by differentiating,

$$y_N = a_0 + a_1X_1 + a_2X_2$$

$\frac{\partial y}{\partial x_1}$ = 2.870, $\frac{\partial y}{\partial x_2}$ = 9.745

Calculating ℓ ,

ℓ is given by ℓ = $\sqrt[J]{\left[\sum(\frac{\partial y}{\partial x_i})^2\right]}$

$$= \frac{J}{\sqrt{[2.870^2 + 9.7452^2]}}$$

$$= \frac{J}{\sqrt{[8.2369 + 94.9650]}}$$

$$= \frac{J}{\sqrt{[103.2019]}}$$

$$= \frac{J}{10.159}$$

$$\ell = \frac{J}{10.159}$$

The chosen range of the value of *J* from 1 to 5 and the corresponding ℓ values and the value of each variable X_i was calculated at each different value of *J* by using the equation $x_i = \ell \frac{\partial y}{\partial x_i}$ and shown in the Table 5. By reversing from the coded level in terms of α to the real level of the operational variables, the real values of operational variables are as presented in brackets in Table 5. These new operational points were tested and the results of the new experiments are shown in Table 5. The point of *J* = 3 gave the highest yield of antioxidant activity of spray dried extract compared to the other points tested. Hence the combination of levels of operational variables at *J* = 3 was selected as the best combination.

Table 5: The real values of operational variables and the results of Method of the Path of Steepest Ascent.

J	ℓ	X_1, Temperature (°C)	X_2, Maltodextrin (%)	Yield of Spray-dried Extract (g)	Antioxidant Activity (%)
1	0.098	0.281 (169.20)	0.955 (29.55)	8.4	17.52
2	0.1968	0.565 (173.43)	1.918 (39.10)	5.2	7.59
3	0.2953	0.843 (177.65)	2.8779 (48.65)	10.6	27.8
4	0.3937	1.130 (181.86)	3.837 (58.20)	9.9	21.09
5	0.4922	0.4942 (186.08)	4.796 (67.75)	7.6	24.42

On physical appearance, the dried extracts for all combinations of levels of operational variables at points *J* = 1 to 5 (Figures 5 to 9) look similarly powdery, but the amounts vary considerably with the amount at *J* = 3 (being highest in antioxidant activity) being highest.

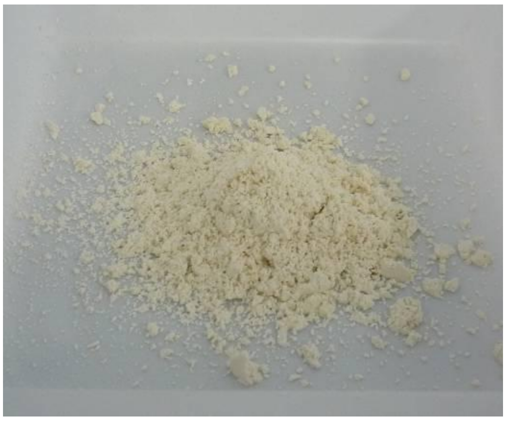

Fig. 5: Physical appearance of the dried extract with spray dryer operated at X_1 = 169.2°C and X_2 = 29.55%.

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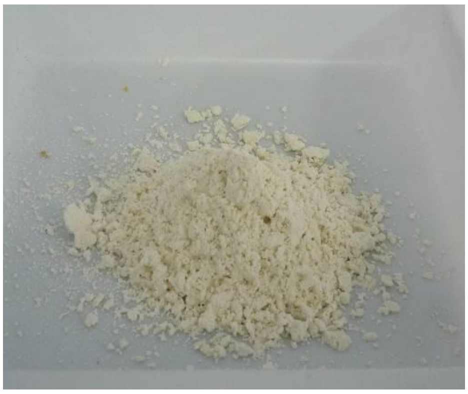

Fig. 6: Physical appearance of the dried extract with spray dryer operated at X_1 = 173.4^{o}C and X_2 = 39.1%..

Fig. 7: Physical appearance of the dried extract with spray dryer operated at X_1 = 177.65^{o}C and X_2 = 48.65%.

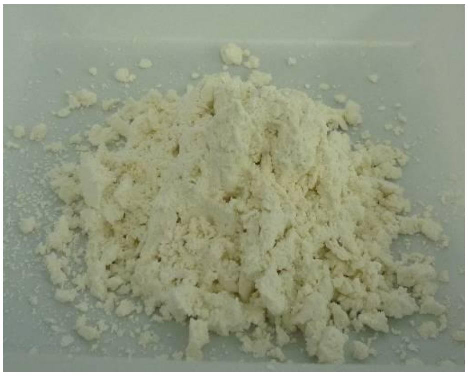

Fig 8: Physical appearance of the dried extract with spray dryer operated at X_1 = 181.86^{o}C and X_2 = 58.20%.

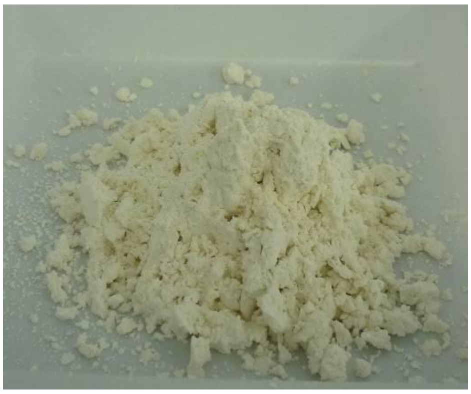

Fig. 9: Physical appearance of the dried extract with spray dryer operated at X_1 = 186.08^{o}C and X_2 = 67.75.

Conclusions:

The combination of levels of operational variables where the spray dryer is operated at temperature of 177.65^{o}C and concentration of maltodextrin of 48.65% produced the highest yield of antioxidant activity of spray dried extract. This concludes that these operational variables was the best combination in yielding the best results of antioxidant activity in spray drying yield of boiling extract liquid of *P. Amboinicus* Lour. These findings may have a bearing on the application of spray drying technology in preparing herbal extracts, and may prove useful to the herbs industries.

Glossary of Terms

No	Symbols	Meaning	Units
1	X_1	The experimental variable temperature	ºC
2	x_1	A value of X_1	ºC
3	$\bar{x}_1$	The specific value of X_1 at point P	ºC
4	X_2	The experimental variable % concentration of maltodextrin	%
5	x_2	A value of X_2	%
6	$\bar{x}_2$	The specific value of X_2 at point P	%
7	y	Yield of antioxidant activity	%
8	P	A point in the yield response surface	%
9	J	Jump distance	-
10	a_i	Coefficient of the variables in the linear regression equation	-
11	a_0	Intercept of the linear regression equation	%

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REFERENCES

Ames, S.N., M.K. Shigrenaga and T.M. Hagen, 1993. Oxidants, antioxidants and degenerative disease of aging. *Proceedings of the National Academy of Sciences of the United States of America*. 90: 7915-7922.

Box, G.E.P. and K.B. Wilson, 1951. On the experimental attainment of optimum condition. *Journal of the Royal Statistical Society*, 13: 1-45.

Che Aniha, C.I., 2008. *99 Herba; Kekal Sihat dengan Petua Nenek Moyang*, 2nd ed. Shah Alam: Alaf 21 Sendirian Berhad.

Chegini, G.R. and B. Ghobadian, 2007. Spray Dryer Parameters for Fruit Juice Drying. *World Journal of Agricultural Sciences*, 3: 230-236.

Damanik, R., N. Damanik, Z. Daulay, S. Saragih, R. Premier, N. Wattanapenpaiboon and M.L. Wahlqvist, 2001. Consumption of bangun-bangun leaves (*Coleus Amboinicus* Lour) to increase breast milk production among Bataknese women in North Sumatera Island, Indonesia. *Asia Pacific Journal of Clinical Nutrition.* 10: 67.

6. Damanik, R., N. Watanapenpaiboon and M.L. Wahlqvist, 2004. The use of a putative lactagogue plant on breast milk production in Simalungun, North Sumatra, Indonesia. *Asia Pacific Journal of Clinical Nutrition*, 16: 87

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

Moure, A., J.M. Cruz, D. Franco, J.M. Dominguez, J. Sineiro, H. Dominguez, M.J. Núňez and J.C. Parajō, 2001. Natural antioxidants from residual sources. *Food Chemistry*, 72: 145-171.

Noraida, A., 2007. *Penyembuhan Semula Jadi dengan Herba*. Kuala Lumpur: PTS Millennia Sdn. Bhd.

Oktay, M., I. Gulcin and O.I. Kulrevioglu, 2003. Determination of in vitro antioxidant activity of Fennel (*Foeniculum vulgarae*) seed extract. *Lebensmittel-W. Scenchaft und Technologie*, 36: 263-271.

Oliveira, W.P., R.B. Bott and C.R.F. Souza, 2006. Manufacture of standardised dried extracts from medicinal Brazilian plants. *Drying Technology*, 24(4): 523-533.

Ong, T., 2008. *Rahsia Herba*. Shah Alam: Alaf 21 Sendirian Berhad.

Packer, L. and C. Colman, 1999. *The Antioxidant Miracle*. Canada: John Wiley and Sons, Inc.

Patel, R.P., M.P. Patel and A.M. Suthar, 2009. Spray drying technology: an overview. *Indian Journal of Science and Technology,* 2(10): 44-47.

Percival, M., 1998. Antioxidants. Revised ed. *Clinical Insights.*10: 1-4.

Scott, M.K., M.B. Connie and D.C. Montgomery, 2005. A modified path of steepest ascent for split-plot experiments. *Journal of Quality Technology*, 37: 75-83.

Shahidi, F. and P.K.J.P.D. Wanasundara, 1992. Phenolic antioxidants. *Critical Reviews in Food science and Nutrition,* 32: 67-103.

Singh, M.P. and H. Panda, 2005. *Medicinal Herbs with Their Formulations*. Delhi: Daya Publishing House.

Souza, C.R.F. and W.P. Oliveira, 2006. Powder properties and system behaviour during drying of *Bauhinia fornicata* link extract. *Drying Technology*, 24(6): 735-749.

Williams, G.M., M.J. Iatropoulus and J. Whysner, 1999. Safety assessment of butylated hydroxyanisole and butylated hydroxytoluene as antioxidant food additives, *Food and Chemical Toxicology,* 37: 1027-1038.

Ziegler, R.G., 1991. Vegetables, fruit and carotenoids and the risk of cancer, *The American Journal of Clinical Nutrition,* 53: 251-259.