International Food Research Journal 18: 583-587 (2011)

Evaluation of the antioxidant potential of some Malaysian herbal aqueous extracts as compared with synthetic antioxidants and ascorbic acid in cakes

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Abstract: Efficacy of some Malaysian herbal aqueous extracts, BHA/BHT (synthetic antioxidants) and ascorbic acid in retarding oxidative rancidity was tested with cakes. The development of lipid oxidation products during 15 days at room temperature was evaluated by means of Peroxide Value (PV) and Thiobarbituric acid-reactive substances (TBARS) value. The six formulations consists of control sample (cake without addition of antioxidant) (F1), cake added with curry leaves extract (F2), cake incorporated with *kesum* leaves extract (F3), cake added with *tenggek burung* leaves extract (F4), cake incorporated with ascorbic acid (F5) and cake added with BHA/BHT (F6). Formulation with the incorporation of *tenggek burung* leaves extract showed powerful oxidative stability effect compared to the formulations with othe plant extracts and control sample. However, cakes with BHA/BHT showed the strongest oxidative stability throughout the storage period. Therefore, it is suggested that *tenggek burung* leaves extract can be added into the food system for effectiveness as antioxidant to prolong the shelf life of the product.

Keywords: Antioxidant, extracts, BHA/BHT, ascorbic acid, cakes

Introduction

Bakery products are widely consumed and are becoming a major component of the international food market (Kotsianis *et al*., 2002). Cake is one of the most common bakery products consumed by people in the world. Nowadays, cake manufacturers face a major problem of lipid oxidation which limits the shelf life of their products (Lean and Mohamed, 1999). Bakery products such as cakes particularly those with high lipid content tend to become rancid after prolonged storage owing to the oxidation of polyunsaturated fatty acids (Smith *et al*., 2004; Ray and Husain, 2002). Foods containing higher content of polyunsaturated fatty acids are more prone to oxidation (Aardt *et al*., 2004). One of the most important changes that occur to food is lipid oxidation. Lipid oxidation lowers the quality and nutritional value of food (Suja *et al*., 2004). The susceptibility of lipid to oxidation is one of the major cause oxidative stress, resulting in the development of rancidity, unpleasant tastes and odours as well as changes in colour (Pezzuto and Park, 2002).

Addition of antioxidant is effective in delaying the oxidation and extending the shelf life of food (Jadhav *et al*., 1996; Decker, 1998). In the food industry, the oxidative deterioration of fats and oils is prevented by synthetic antioxidants such as butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT). Although efficient in preventing oxidation, only a few synthetic compounds are currently approved for use in the food industry because of their potential toxicity and carcinogenic (Botterweck *et a*l., 2000). To satisfy consumers' preference for natural food additives over synthetic ones, there is increasing importance searching for natural antioxidants from herbs, fruits, vegetables and spices as a less harmful alternatives to synthetic antioxidants (Daker *et al*., 2008). Recently, special attention has given to the use of natural antioxidant because of the worldwide tend to avoid or minimise synthetic food additives (Krings and Berger, 2001). Plant extracts obtained from some fruits and vegetables have been reported to be effective antioxidants (Cao *et al*., 1996; Vinson *et al*., 1998). In this investigation water was used as the extraction solvent to extract the hydrophilic antioxidants present in the plants. For use in foods, plant extracts made with water are nutritionally more relevant and would have obvious advantages in relation to certification and safety (Moller *et al*., 1999).

This study was carried out to investigate the effectiveness of three types of local plants water extracts *kesum* (*Polygonum minus*), curry leaves (*Murraya koenigii*) and *tenggek burung* (*Melicope lunu-ankenda*) incorporated in the cakes stored for

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15 days in comparison with ascorbic acid, BHA/BHT and a control treatment without plant extracts.

Materials and Methods

Production of cake

The cakes were formulated into six formulations i.e. without plant extract, with 200 ppm curry leaves extract, 200 ppm *kesum* extract, 200 ppm *tenggek burung* extract, 200 ppm ascorbic acid and 200 ppm BHA/BHT. The margarine (without antioxidant) supplied from Fryola Bakery Fat and castor sugar were beaten in a mixing bowl on medium speed until light and fluffy (creamy) for 12 mins. The whole eggs (white and yolk) were added one by one. Addition of eggs had to be careful ensuring that each addition is completely incorporated. The batter was beaten until it is smooth for 5 mins. After that, the antioxidant (natural or synthetic) was added into the batter for 1 min at low speed. With the mixer at low speed, sifted cake flour were fold in carefully without over mixing into the batter for 2 mins. All of these mixtures were put in the pan and baked in an oven at an approximately temperature of 160°C for 60 mins. Once baked, the cakes were allowed to cool for 30 mins and then stored in plastic containers at ambient temperature about 15 days for further analysis.

Plant material and extraction

Extraction using water was determined according to the method of Duh and Yen (1997). 20 g of the fresh sample (*Murraya koenigii*, *Polygonum minus* and *Melicope lunu-ankenda*) were extracted with boiling water (600 ml) for 10 minutes. After boiling, sample was blended and filtrated with Whatman No. 4. Water was removed by evaporating using Rotary Evaporator (Model: BüCHI Rotavapor R-200) at the temperature of 70°C. After water been removed, extracts were frozen in blast freezer (SANYO VIP Series -86°C) at -70°C for 2 days and freeze dryer (CHRIST Alpha 2-4) at -16°C for 3 days. Then, the extracts were stored in the amber bottles at 0°C for analysis.

Peroxide value (PV)

Peroxide value was determined using AOAC methods (1984). Fat extraction was carried out using Kinsella *et al*. (1977) extraction method with slight modification.

Thiobarbituric acid (TBA) value

The 2-thiobarbituric acid (TBA) distillation method of Tarladgis *et al*. (1960) was used to determine lipid oxidation.

Results and Discussion

Peroxide value

The effect of natural and synthetic antioxidants on Peroxide Value (PV) of cake samples over 15 days of storage period under room temperature was illustrated in Figure 1. PV range of 10-20 mEq/kg, food product is considered rancid but still acceptable. But if more than 20 mEq/kg, it considered food product already rancid and unacceptable to consume (Pearson, 1970). All samples were able to maintain PV less than 10 mEq/kg until the fifteen day of storage.

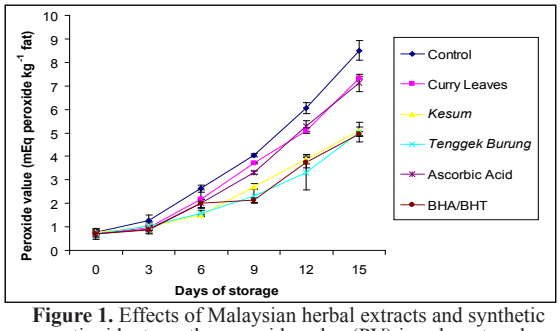

Figure 1. Effects of Malaysian herbal extracts and synthetic antioxidants on the peroxide value (PV) in cakes stored at room temperatures for 15 days

In present study, all samples were considered not rancid and still acceptable. Control exhibited the highest PV throughout the storage period, showing a high oxidation process. A slow rise in PV was observed for all the treated samples, revealing the effectiveness of natural plant extracts and standards in the stabilisation of cakes during storage under room temperatures. Among all samples, the cakes treated with *kesum*, *tenggek burung* and BHA/BHT showed the lowest PV throughout storage period followed by ascorbic acid, curry leaves and control sample. Cakes added with BHA/BHT showed the lowest PV at the end of storage with no significant different compared to cakes incorporated with *kesum* and *tenggek burung* extract. Among all plant extracts added in cakes, *tenggek burung* extract had the lowest PV during storage. However, the PV cakes incorporation with *tenggek burung* and *kesum* extracts were not significantly different. This indicated that *kesum* and *tenggek burung* extract possess a good antioxidant power which are comparable to BHA/BHT after 9 days of storage. These results suggested that *tenggek burung* and *kesum* were effective in suppressing the oxidation of cakes comparable to that of BHA/BHT.

The antioxidative effect may have contributed to the oxidative stability of cakes with addition of natural and synthetic antioxidants. When added into the cakes, antioxidants prevent the lipid peroxides formed during storage and delayed oxidation. This could be due to the slow permeation rate of antioxidant components into lipid bilayer of the cakes.

The results is in agreement with those reported by Noriham *et al*. (2004), that *kesum* and *tenggek burung* showed antioxidant activity against lipid oxidation in mechanically deboned chicken meat sausages. Plant extracts are well known for their phenolic compounds and the antioxidant activity of many natural extracts is due to phenolic extracts (St. Angelo *et al*., 1990; Wong *et al*., 1995). BHA/BHT combinations were generally known as fat soluble antioxidant and had very good carry-through in fats (Budavari, 1989). On the other hand, ascorbic acid possessed low antioxidant activity in supressing oxidation in cakes. After 6 days of storage, the effectiveness of ascorbic acid against lipid hydroperoxides gradually decreased until at the end of storage. Ueda (1986) reported that addition of ascorbic acid at the intial stage of autoxidation produced ascorbic acid-derived free radicals which supressed the oxidation, but had no effect at a later stage when maximum PV had been reached. This suggests that the supression might be caused by the reaction of ascorbic acid with some intermediate product from the intial stage of autoxidation. Sardakei and Howell (2008) proposed ascorbic acid had to combine with other standard antioxidants such as Vitamin E or citric acid to slow down oxidation processes effectively.

The incorporation of natural and synthetic antioxidants was not affected by temperature (baked at 160-180°C) as indicated by the PV and TBA results. In addition, the antioxidants were not affected by storage over a period of 15 days, as *kesum*, *tenggek burung* and BHA/BHT exhibited higher antioxidant activity in cakes than other treated samples during storage period. The phenolic contents in plant extracts were not destroyed during baking might be explained by the increase of extractable (or bioavailable) phenolic content due to the influence of high temperature. High temperature during baking probably destroys some of the complexes of hydrolysable phenols with other food components resulting in increase of extractable phenolic content in plant extracts (Vitali *et al*., 2009). These statement also were in agreement with Gelinas and McKinnon (2006) and Holtekjolen *et al*. (2008) reported that baking increase the total phenolic content of the samples while others have claimed that phenolic compounds are destroyed during baking (Leenhardt *et al*., 2006). Additionally, literature data indicate that increased antioxidant potential might be attributed to the production of certain Maillard reaction products that results from a condensation reaction between amino acids (or proteins) and reducing sugars or lipid oxidation products under the influence of high temperature and might occur during baking (Jing and Kitts, 2004; Sun *et al*., 2006; Wagner *et al*., 2007; Yilmaz and Toledo, 2005).

The cakes prepared by the incorporation of antioxidants from *kesum* and *tenggek burung* extract were accepted well during the storage study which indicates the efficiency of antioxidants in preventing the onset of rancidity comparable with BHA/BHT.

Thiobarbituric acid (TBA) value

TBA values for six types of cakes which include control sample were observed until day 15 at 532 nm absorbance. The results showed that TBA value and red colour intensity for control sample is higher than other sample that has been tested. Incorporation of Malaysian herbal extracts and synthetic antioxidants significantly ($p<0.05$) reduced TBA values compared to the control sample throughout the storage. Figure 2 showed that there were an increased in TBA values for all samples throughout the storage at room temperature for 15 days. TBA value less than 0.576 mg / kg^{-1} sample are considered not rancid, whereas values of 0.65 – 1.44 mg / kg^{-1} sample are regarded as rancid but still acceptable and values greater than 1.5 mg / kg^{-1} sample are said to be rancid and unacceptable (Ke *et al*., 1984).

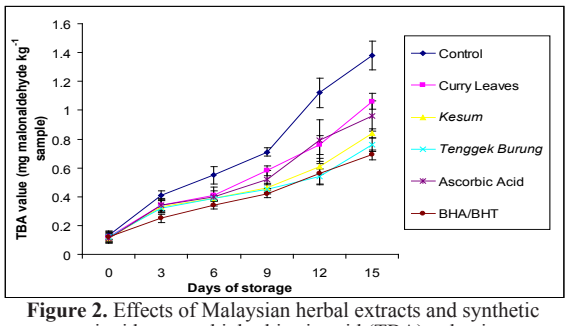

Figure 2. Effects of Malaysian herbal extracts and synthetic antioxidants on thiobarbituric acid (TBA) value in cakes stored at room temperatures for 15 days

At the end of storage (day 15) all treatments includes control sample resulted as rancid but still acceptable. The TBA value for control cake and cake added with curry leaves extract exhibited more than 1.0 mg / kg^{-1} sample at the end of storage period. The TBA value for control sample showed the highest value compared to other sample studied. All treated samples resulted in significantly lower ($p< 0.05$) TBA values when compared to the control, which indicates that the natural extracts and synthetic antioxidants incorporated into cakes exhibited antioxidant properties. Among all plant extracts tested, cake added with *tenggek burung* extract had the lowest TBA values throughout the storage period compared to samples with other plant extracts and ascorbic acid. However, the most effective antioxidant in preventing lipid oxidation in cakes throughout storage, as measured by TBA values was

BHA/BHT sample followed by sample added with *tenggek burung*. Sample with *kesum* extract was more effective in preventing lipid oxidation than control cake and cakes incorporated with curry leaves extract and ascorbic acid. The increased in TBA values in cakes incorporated with *tenggek burung* and *kesum* was very slow and remained below 1.0 mg / kg^{-1} sample up to 15 days.

Bakery products manufacturer uses a wide range of ingredients of which the fats and oils offer the highest potential risk of rancidity in the autoxidation of fats, unsaturated fatty acids are oxidised to hydroperoxides which on subsequent decomposition yield a number of saturated and unsaturated aldehyde and ketones (WHO, 1972). One of the most important changes occuring during food production and storage is lipid oxidation (St. Angelo, 1996). This generally involves the degradation of polyunsaturated fatty acids and the production of secondary decomposition products including carbonyls and hydrocarbon compounds (Claxson *et al*., 1994) and leading to the development of off-flavours or off-odours (Ladikos and Lougovoist, 1990). Results in this study confirm that *tenggek burung* in cakes can delay lipid oxidation and reduce the potential risk induced by lipid oxidation products better than other plant extracts tested. TBA values of cakes that had been treated with *kesum*, *tenggek burung* and curry leaves extracts were much lower than the control thereby indicating protection to the cakes against autoxidation. Several studies have reported on the relationship between phenolic content and antioxidant activity in certain plant products (Velioglu *et al*., 1998). Previous results showed that large amount of phenolics in *tenggek burung* may cause its strong antioxidant activity in cakes. *Tenggek burung* phenolics may act in a similar fashion as reductones by donating the electrons and reacting with free radicals to convert them to more stable product and terminate free radical chain reaction. This can be indicated that marked antioxidant activity of *tenggek burung* extract in cakes seem to be the result of their radical scavenging activity and reducing power. This study shows that polyphenolic antioxidant compounds in Malaysian herbal extracts are distributed and remianed functional as antioxidant in cakes system.

Conclusion

This study concluded that all Malaysian herbs water extracts provide antioxidant benefits to cakes during room temperature storage. Between these three extracts, *tenggek burung* demonstrated the most potent effect in terms of antioxidative activity which comparable to that of BHA/BHT. However, cakes with BHA/BHT showed the strongest oxidative stability throughout the storage period. Therefore, it is suggested that *tenggek burung* extract could be used to extend the shelf life of cakes for effectiveness as natural antioxidant agent.

Acknowledgements

We would like to thank to the E-SCIENCE (PROJECT CODE: 03-01-01-SF0032) under Ministry of Science, Technology and Innovation (MOSTI) and Universiti Teknologi MARA for financial assistance to conduct this research.

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