International Food Research Journal 20(1): 285-289 (2013)
Journal homepage: *http://www.ifrj.upm.edu.my*

Microencapsulation of natural antioxidant powder from *Aloe vera* (L.) skin using foam mat drying method

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Article history

Received: 27 March 2012
Received in revised form: 14 May 2012
Accepted:15 May 2012

Keywords

Antioxidant
Aloe vera (L.) skin powder
maltodextrin
tween 80
temperature
microencapsulation

Abstract

In this study, the microencapsulation of natural antioxidant powder from *Aloe vera* (L.) skin was carried out using 10% maltodextrin, 0.3% tween 80 and drying at 60^{0}C; and the product was found to have free radical scavenging activity using DPPH (88.31%), total phenol (34.921%), and α tocopherol by HPLC (87.789mg/g). The GC-MS analysis for the phyto-component shows that the major compounds identified were squalene (16.84%), limonene (14.17%), 7- tetradeecane (13.13%) n-Hexadecanoic acid (11.91%) and α-Tocopherol (4.18%). The FTIR analysis shows that the functional components in the *Aloe vera* skin powder were phenol, aromatic, substituted alkenes, aromatic acid halide, aliphatic acid halide, eter R-O-R, Nitro NO_2, Keton R-CO-R,vinilidene, carboxylic acid, metilene and OH. SEM analysis showed that drying process by foam mat method resulted in a structure that has the capacity to absorb water easily and can dissolved in cold water. The present findings, especially on the microencapsulation of *Aloe vera* skin powder will provide a new opportunity to improved the bioactive properties of the phyto-component as antioxidant agent.

Introduction

Aloe vera (L.) is a tropical or subtropical plant widely use in folk medicine, cosmetic, supplement and food material (Eshun and He, 2004). *Aloe vera* (L.) skin is seldom used in food processing, however, Miladi and Damak (2008) noted that *Aloe vera* (L.) skin contained pharmaceutical compound such as antioxidant, making it a potential source of raw materials to be processed into natural antioxidant powder.

The processing of *Aloe vera* (L.) skin into natural antioxidant powder requires encapsulation using maltodextrin as protective compound, Tween 80 as foaming agent and low drying temperature (60^{0}C). Finotelli and Rocha-Leão (2010) suggested that the addition of maltodextrin as encapsulation can protect the release of nutrient components as well as protecting important compound such as antioxidant from extreme temperature, whilst Iswari (2007) noted that tween 80 as foaming agent help figuration of a good suspension. Thaisong and Rojanakorn (2011) suggested that drying temperature between 60-75^{0}C was the optimum best temperature in maintaining the quality of powder.

Rajkumar *et al*. (2007) noted that the process of drying with the addition of foaming agent will produce a good quality product. This is in agreement with the findings by Kudra and Ratti (2008) who reported that the drying process by using foaming material will decrease the drying time. However, the objectives of this study were to find out the effect of microencapsulation on natural antioxidant compounds in the powder of *Aloe vera* (L.) skin using maltodextrin, tween 80 and drying at 60^{0}C.

Materials and Methods

Sample preparation

Aloe vera (L.) skin of 10 months old were obtained from Kalimantan Barat, Indonesia. After harvesting, the *Aloe vera* (L.) skin were separated from the gel, washed by aquades and was extracted at 80^{0}C for 60 min. The filtrates obtained were then mixed with 10% maltodextrin, 0.3% tween 80 and homogenized using a mixer at a speed of 1800 rpm for 10 min. The rough obtained were scattered on the baking pan with alumunium foil and dried at 60^{0}C for 6 h. The dried rough was blended and sifted using sieve 100 mesh.

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Determination of antioxidant activity

The *Aloe vera* (L.) skin powder were analyzed for its antioxidant activity using the DPPH (2,2-diphenyl-2-picrylhydrazyl) radical scavenging assay. Sample (200 g) was dissolved in 100 mM Tris–HCl buffer (800 µl, pH 7.4) followed by the addition of 1 ml 500 µM DPPH. The solution was homogenized using a shaker and storage in dark room for 20 min. Spectrophotometry was used to determine the absorbance at 517 nm.

Determination of α-tocopherol

The *Aloe vera* (L.) skin powder were analyzed for α-tocopherol content using HPLC (High Performance Liquid Cromatography) under the following conditions: column used were inertsil NH_2 µm 250 x 4.6 mm, and the flow rate was 1 ml/ minute. The column temperature was 30^0C, detector UV 290nm and using ethyl acetate/n hexane 30/70 as gas carrier.

Determination of phytocomponent

The *Aloe vera* (L.) skin powder were analyzed for phytocomponent using GC-MS QP2010S-Shimadzu under the following conditions: column used were Rtx-5MS, 30 m length and inner diameter of 0.25 mm and the initial column temperature was 40°C and final temperature was 260°C (5°C/min), while the injector temperature was 250°C with split mode injector and split ratio of 68 and pressure of 14.0 kPa. The flow rate was 1.3 ml/min and the flow within the column was 0.50 ml/min. The detector temperature was 300^0C and using Helium as the gas carrier with EI (Electron Impact); and the samples volume injected was 1µl. Compounds were identified by comparing retention indices/comparing mass spectra of each compound with those of authentic samples and library.

Determination of functional compounds

The *Aloe vera* (L.) skin powder was analyzed for its functional compounds using FTIR (Fourier Transform Infra Red). The IR spectra were recorded on FTIR-8400S (Shimadzu Deutchland GmbH) spectrophotometer in KBr and polyethylene pellets. Samples were weigh-in at 0.01 g and homogenized with 0.01 g KBr anhydrous by mortar agate. The sample and KBr mixture were pressed by vacuum hydrolic (Graseby Specac) at 1.2 psi to obtained transparency pellet. Scanned sample passed through infra red, where its continuing wave by detector connected to computer with set values of tested sample spectrum. Samples were usually scanned in the absorption area of 500-4000 cm^{-1}. The results of analysis consisted of chemical structure, molecular binding form and certain functional group of tested sample as basic of spectrum type.

SEM studies

The microstructure of *Aloe vera* (L.) skin powder were analyzed using SEM (Scanning Electron Microscopy) JSM T-100, JEOL, Japan. Samples were dehydrated by putting them into critical point drying equipment, then fastened with a special glue to stub (samples holder). Samples were left to dry for ±1 day. Samples were coated with pure gold or carbon for 1 h at a coating evaporator machine prior to be observed and their microscopic photos taken by scanning electron microscope (SEM) machine.

Results and Discussion

Free radical scavenging activity

The antioxidant activity of *Aloe vera* (L.) skin powder determined using DPPH assay (%) was 88.31%, which is higher than synthetic antioxidant such as BHT (butylated hydroxytoluene) at 70.5% and α-tocopherol at 65.65 as reported by Anilakumar *et al*. (2010). The higher level of antioxidant activity observed in the encapsulated *Aloe vera* (L.) skin powder are probably due to its relative resistant to the effect of the drying temperature and the effect of encapsulation using maltodextrin and tween 80. Patras *et al*. (2009) suggested that the increase in free radical antioxidant could be due to better extractability of antioxidant component and higher level of phenolic content. Elsewhere Pengseng *et al*. (2010) reported that phytochemicals such as phenolic content, ascorbic acid, tocopherol and pigment also contribute to total antioxidant activity and has a good correlation between the antioxidant activity and its total phenolic compound content.

Total phenol content

Total phenol content of *Aloe vera* (L.) skin powder was 34.921%. Saénz *et al*. (2009) and Desai and Park (2005) noted that maltodextrin can improve the stability of phenol compound as maltodextrin can protect phenol compound from oxidation effect, oxygen, water and extreme temperature. Whereas Pengseng *et al*. (2010) noted that the use temperature between 25-90^0C does not caused damaged to the antioxidant components in material, thus will probably explained why the total phenol in the *Aloe vera* (L.) skin powder examined in this study is within the limits and are safe within the protected bioactive compound.

α-tocopherol content

α-tocopherol content obtained from *Aloe vera* (L.) skin powder was 87.789 mg/ 100g and its typical

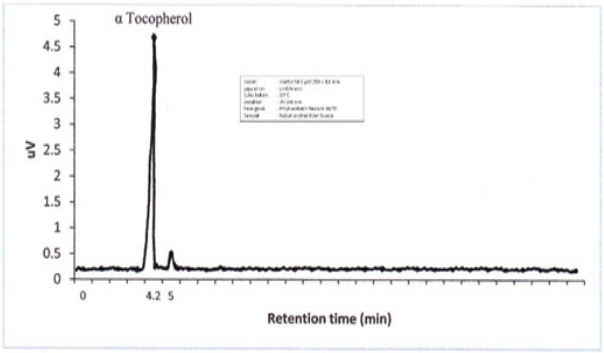

Figure 1. HPLC chromatogram of α-tocopherol in *Aloe vera* (L.) skin powder

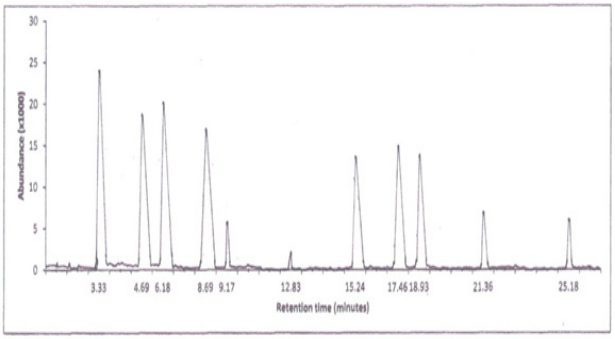

Figure 2. GC-MS chromatogram of *Aloe vera* (L.) skin powder

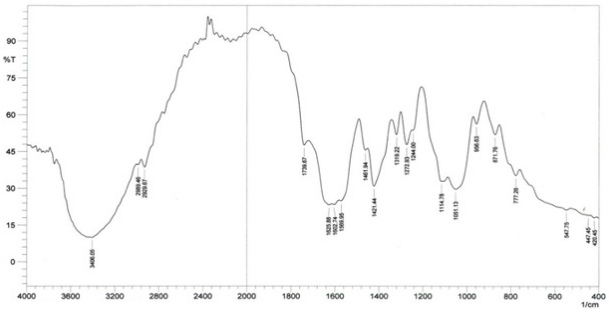

Figure 3. Infra red spectrum of *Aloe vera* (L.) skin powder

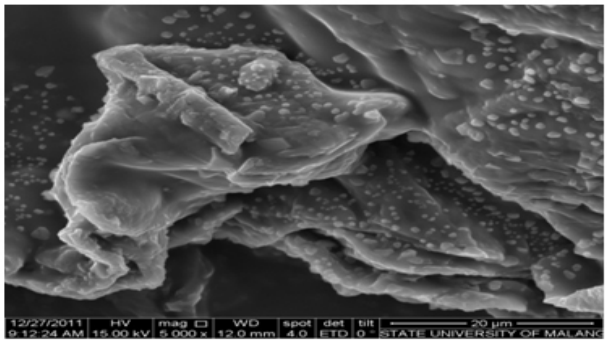

Figure 4. Microstructure of *Aloe vera* (L.) skin powder

chromatogram is shown on Figure 1. α-tocopherol content on *Aloe vera* (L.) skin powder obtained were influenced by drying methods, in addition to the effect of encapsulation using maltodextrin and tween 80. Estiasih and Sofia (2009) suggested that drying using foaming method increased the surface area, therefore increasing the contact of product which facilitate the oxidation process to occurred and affects the tocopherol. This is due to the known facts that tocopherol stability is very much affected by various conditions in the environment. Saguy *et al.* (2003) noted that the effects of heating can cause α-tocopherol to become unstable due to reactions such as hydrolysis, oxidation and polymerization during direct contact with heat. Therefore, Lešková *et al*. (2006) suggested that its negative alteration will lead decrease the effect of α-tocopherol as antioxidant agent.

Identification antioxidant compounds of Aloe vera (L.) skin

A typical gas chromatogram of *Aloe vera* (L.) skin powder is shown in Figure 2 and a list of the compounds identified appears in Table 1. Eleven compounds of *Aloe vera* (L.) skin powder were identified using GC-MS and the major compound identified was squalene (16.84%), limonene (14.17%), 7- tetradeecane (13.13%) and n-Hexadecanoic acid (11.91%). Lakshmi *et al*. (2011) reported that the major phyto-components possessing activity as antimicrobial and antioxidant were tetradecanoic acid, methyl ester, hexadecanoic acid, and squalene. Whilst Botes *et al*. (2008) reported that campesterol and coumaric were identified as phytosterols in *Aloe greatheadii* var. *davyana* extracts and were associated with antioxidant properties. Coopoosamy (2010) also reported limonene and carvone as volatile compounds on leaf exudates of *Aloe excelsa* (Berger).

Identification antioxidant compounds by FTIR

The infra red spectrum of *Aloe vera* (L.) skin powder as shown in Figure 3 was in the wave length range of 420.45 to 3406.05 cm^{-1}, with 20 functional compounds detected (Table 5). According to Table 2 and Figure 3, the presence of broad bands at 3406.05 cm^{-1} can be attributed to (OH) stretching vibrations. The wave length at 2989.46 to 2929.67 cm^{-1}; 1319.22 to 1569.95 cm^{-1}; and 1051.13 to 1272.93 cm^{-1} can be attributed to metilene group, aromatic group and eter (R-O-R), respectively. Rajendran *et al*. (2007) reported that the functional compounds of *Aloe vera* (L.) when present are at the wave length at 611.4 cm^{-1}; 717.5 cm^{-1}; 1051.1 cm^{-1}; 1398.3 cm^{-1}; 1623.9 cm^{-1}; 1730.0 cm^{-1}; 2912.3 cm^{-1}; 3155.3 cm^{-1} and 3398.3 cm^{-1}.

SEM study

A representative scanning electron micoroscopy (SEM) conducted at magnification 5000x in Figure 4 shows the microstructure of *Aloe vera* (L.) skin powder containing bubbles or spotted on the surface due to the use of Tween 80 as foaming agent.

Table 1. Identified compounds of *Aloe vera* (L.) skin powder

Peak	Compounds	% RA
1	Squalene	16.84
2	7-Tetradecane	13.13
3	Limonene	14.17
4	n-Hexadecanoic acid	11.91
5	Campesterol	3.96
6	B-Sitosterol	1.43
7	9-octadecanoic methyl ester	9.53
8	Carvone	10.43
9	Comaric	9.58
10	Lupeol	4.80
11	α-Tocopherol	4.18

Table 2. Functional compounds of *Aloe vera* (L.) skin powder analyzed by using FTIR

No	Wave length (cm^{-1})	Vibration type	Functional compound
1	420.45	C-OH bend	Phenol AR-H
2	447.45	OH bend	Phenol AR-H
3	547.75	Def cincin 2p	Aromatic
4	777.26	CH_2 kel.ben.wag	Alkena substituted
5	871.76	C-C or C-Cl stretch	Aromatic acid halide
6	956.63	C-Cl stretch	Aliphatic acid halida
7	1051.13	C-O-C stretch eter siklis	Eter ROR
8	1114.78	C-O-C stretch eter siklis	Eter ROR
9	1244	C-O-C stretch vinil eter	Eter ROR
10	1272.93	C-O-C stretch alkil aril eter	Eter ROR
11	1319.22	NO_2 stretch aromatic	Nitro NO_2
12	1421.44	Ring aromatic stretch (4p)	Aromatic
13	1461.94	NO_2 stretch aromatic	Nitro NO_2
14	1569.95	NO_2 stretch aromatic	Nitro NO_2
15	1602.74	C=C stretch konj	Keton RCOR
16	1625.74	C=C stretch	Vinilidena
17	1739.67	C=O stretch monomer	Carboxylic acid
18	2929.67	CH stretch into alkena	Metilena CH_2
19	2989.46	CH stretch into alkena	Metilena CH_2
20	3406.05	OH stretch bonded	OH

Kudra and Ratti (2006) noted that the drying process combined with foaming agent will lead to the formation of cavity on material surfaces, therefore water will drained out faster when the drying process was done without foaming agent. This observation may suggest that the density of powder produced is lower compared to the non-foamed ones as the dry product contains a lot of bubbles. This is in agreement with the findings by Iswari (2007) who reported that drying product by foam mat method produced a structure that can absorb water easily and thus food can dissolved in cold water.

Conclusion

Microencapsulation of *Aloe vera* skin powder will provide a new opportunity to improved the bioactive properties of the phyto-component as antioxidant agent. The higher level of antioxidant activity observed in the encapsulated *Aloe vera* (L.) skin powder are probably due to its relative resistant to the effect of the drying temperature and the effect of encapsulation using 10% maltodextrin and 0.3% tween 80

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