

# Studies on the preparing of KOMBUCHA using Blanching water of Fammulina velutipes

王子輯、李世傑、林麗雲

E-mail: 324825@mail.dyu.edu.tw

## ABSTRACT

This study was to research the preparing of Kombucha using blanching water of Fammulina velutipe (BWFV). The quality and polysaccharide content of the Kombucha were measured. The red tea was first prepared using tea: sugar: water at the ratio of 1: 5: 200. Then 900 mL of red tea and 100mL of commercial kombucha was added to made up to 1L. The BWFV was standardized by adjusting its soluble solid content to 2 ° Brix diluted with distilled water. This standardized BWFV was used for water substitution at 10%, 50%, 100% level during the traditional Kombucha batch production. Each experimental Kombucha batch was kept in room temperature for 12 day ' s fermentation. The change of pH, soluble solids, reducing sugar, total sugar, titratable acid and polysaccharide were monitored. Variation level of BWFV, including 5%, 10%, 20%, 30% of the total volume of water substitution, were used for preparing Kombucha for comparison based on chemical analysis and sensory evaluation. Based on our results, the more BWFV added, the more polysaccharide it will be produced in the Kombucha. Unfortunately, the consumer's acceptance and anti-oxidative activities both will be lower when more BWFV added. By using 10 days fermentation process, the 100% substitution BWFV treatment can both have maximum polysaccharide yield and acceptable sensory evaluation scores. The reducing power in Kombucha increased when more BWFV were added. The adding of BWFV was recommended to increase its functionality of Kombucha by increasing the polysaccharides content in the drink.

Keywords : Blanching water of Fammulina velutipe (BWFV)、Kombucha、Polysaccharide、Sensory evaluation

## Table of Contents

封面內頁	簽名頁	中文摘要	iv	英文摘要	v	致謝	vi	目錄	vii	圖目錄	x	表目錄	xii	1. 緒論	1	2. 文獻回顧	2	2.1 紅茶菇之簡介	2	2.1.1 紅茶菇之歷史背景	2	2.1.2 紅茶菇之相關成份及製備方法	2	2.1.3 紅茶菇之機能性	3	2.1.4 紅茶菇之毒性	4	2.2 多醣體之簡介	4	2.2.1 多醣體抗癌機制	5	2.2.2 金針菇多醣體之簡介	6	2.3 抗氧化之簡介	8	2.3.1 自由基之簡介	8	3. 材料方法	10	3.1 實驗材料	10	3.1.1 原料	10	3.2 實驗儀器及試藥	10	3.2.1 實驗儀器	10	3.2.2 實驗試藥	10	3.3 實驗架構	12	3.3.1 紅茶菇菌液之培養方法	12	3.3.2 金針菇殺菁液高濃度之製備	12	3.3.3 低濃度金針菇殺菁液之製備	13	3.4 成份分析	18	3.4.1 可溶性固形物	18	3.4.2 pH值	18	3.4.3 可滴定酸(Titratable acidity)	18	3.4.4 總糖之測定	18	3.4.5 還原糖之測定	19	3.4.5.1 標準曲線	19	3.4.5.2 紅茶菇發酵液之測定	19	3.4.6 多醣濃度之測定	20	3.5 抗氧化活性之分析	20	3.5.1 DPPH自由基清除能力測定	20	3.5.2 螯合亞鐵離子能力	20	3.5.3 還原力測試	21	3.6 嗜好性官能品評	22	4. 結果與討論	23	4.1 紅茶菇接種高濃度金針菇殺菁液發酵過程之探討	23	4.1.1 發酵期間成份分析探討	23	4.1.2 添加高濃度金針菇殺菁液官能品評之探討	31	4.2 紅茶菇接種低濃度金針菇殺菁液發酵過程之探討	34	4.2.1 發酵期間成份分析探討	34	4.2.2 添加低濃度金針菇殺菁液官能品評之探討	42	4.3 抗氧化分析結果	45	4.3.1 DPPH清除力結果之探討	45	4.3.2 亞鐵離子螯合分析結果之探討	50	4.3.3 還原力分析結果之探討	55	5. 結論	57	參考文獻	59	圖目錄		圖2.1 -D-glucan(葡聚糖)之結構	6	圖3.1 紅茶菇菌液擴大培養方式	14	圖3.2 紅茶菇菌液控制組培養方式	15	圖3.3 金針菇殺菁液以10%、50%、100%三種濃度取代水發酵12天	16	圖3.4 金針菇殺菁液以5%、10%、20%、30%四種濃度取代水發酵18天	17	圖4.1 以不同比例之金針菇殺菁液取代水進行紅茶菇發酵期間pH值之變化	24	圖4.2 以不同比例之金針菇殺菁液取代水進行紅茶菇發酵期間可溶性固形物之變化	25	圖4.3 以不同比例之金針菇殺菁液取代水進行紅茶菇發酵期間可滴定酸之變化	27	圖4.4 以不同比例之金針菇殺菁液取代水進行紅茶菇發酵期間總糖之變化	28	圖4.5 以不同比例之金針菇殺菁液取代水進行紅茶菇發酵期間還原糖之變化	29	圖4.6 以不同比例之金針菇殺菁液取代水進行紅茶菇發酵期間多醣含量之變化	30	圖4.7 以不同比例之金針菇殺菁液取代水進行紅茶菇發酵期間pH值之變化	35	圖4.8 以不同比例之金針菇殺菁液取代水進行紅茶菇發酵期間可溶性固形物之變化	36	圖4.9 以不同比例之金針菇殺菁液取代水進行紅茶菇發酵期間可滴定酸之變化	38	圖4.10 以不同比例之金針菇殺菁液取代水進行紅茶菇發酵期間總糖之變化	39	圖4.11 以不同比例之金針菇殺菁液取代水進行紅茶菇發酵期間還原糖之變化	40	圖4.12 以不同比例之金針菇殺菁液取代水進行紅茶菇發酵期間多醣含量之變化	41	圖4.13 紅茶菇發酵第0天對DPPH自由基清除活性之比較	46	圖4.14 紅茶菇發酵第2天對DPPH自由基清除活性之比較	47	圖4.15 紅茶菇發酵第4天對DPPH自由基清除活性之比較	48	圖4.16 紅茶菇發酵第6天對DPPH自由基清除活性之比較	49	圖4.17 紅茶菇發酵第0天對亞鐵離子螯合能力之比較	50	圖4.18 紅茶菇發酵第2天對亞鐵離子螯合能力之比較	52	圖4.19 紅茶菇發酵第4天對亞鐵離子螯合能力之比較	53	圖4.20 紅茶菇發酵第6天對亞鐵離子螯合能力之比較	54	圖4.21 紅茶菇於發酵期間還原力之比較	56	表目錄		表2.1 不同濃度金針菇殺菁水醣類含量之測定	8	表4.1 高濃度之金針菇殺菁液取代水於發酵第八天之嗜好性品評結果	32	表4.2 高濃度之金針菇殺菁液取代水於發酵第十天之嗜好性品評結果	33	表4.3 低濃度之金針菇殺菁液	
------	-----	------	----	------	---	----	----	----	-----	-----	---	-----	-----	-------	---	---------	---	------------	---	----------------	---	---------------------	---	---------------	---	--------------	---	------------	---	---------------	---	-----------------	---	------------	---	--------------	---	---------	----	----------	----	----------	----	-------------	----	------------	----	------------	----	----------	----	------------------	----	--------------------	----	--------------------	----	----------	----	--------------	----	-----------	----	--------------------------------	----	-------------	----	--------------	----	--------------	----	-------------------	----	---------------	----	--------------	----	---------------------	----	----------------	----	-------------	----	-------------	----	----------	----	---------------------------	----	------------------	----	--------------------------	----	---------------------------	----	------------------	----	--------------------------	----	-------------	----	--------------------	----	---------------------	----	------------------	----	-------	----	------	----	-----	--	------------------------	---	------------------	----	-------------------	----	--------------------------------------	----	--	----	-------------------------------------	----	--	----	--------------------------------------	----	------------------------------------	----	-------------------------------------	----	--------------------------------------	----	-------------------------------------	----	--	----	--------------------------------------	----	-------------------------------------	----	--------------------------------------	----	---------------------------------------	----	-------------------------------	----	-------------------------------	----	-------------------------------	----	-------------------------------	----	----------------------------	----	----------------------------	----	----------------------------	----	----------------------------	----	----------------------	----	-----	--	------------------------	---	----------------------------------	----	----------------------------------	----	-----------------	--

REFERENCES

- 1.水野卓、川合正允, 1997。菇類的化學、生化學。賴慶亮譯, 國立編譯館。台北, 台灣。
- 2.王伯徹、陳啟楨、華節, 1998。食藥用菇類的培養與應用。食品工業發展研究所。p56-57。
- 3.池川哲郎, 1989。抗腫瘍研究20年, 抗腫瘍研究20年編輯委員會編。長野縣農村工業研究所出版社。P9-22。
- 4.朱聖哲, 2006。培養模式與條件對紅茶菇的菌種活性、抗氧化力及茶多酚量之影響。國立中興大學研究所博士論文。台中。
- 5.李錦楓, 1976a。關於健康飲料海寶(紅茶菇)的真象。食品工業。8(3):35-37。
- 6.陳嘉雄, 2004。抗氧化物對神經細胞遭受神經毒害之保護作用研究。中山醫學大學生物化學研究所碩士論文, 台中。
- 7.彭秋妹、王家仁。1990。食品官能檢查手冊。食品工業發展研究所。新竹, 台灣。第23-45頁。
- 8.黃麗娜, 1996。食用菇菌絲體深層培養在食品工業上的應用。食品工業, 28(9):20-26。
- 9.賴敏男, 1976。紅茶菇的微生物及成份。食品工業。8(9):45-49。
- 10.Allen, C.M., 1998. Past research on kombucha tea. The kombucha FAQ part 6. Research and test results. [Http://persweb.direct.ca/chaugen/kombucha\\_faq\\_part06.html](http://persweb.direct.ca/chaugen/kombucha_faq_part06.html)
- 11.AOAC, 1984. Official methods of analysis. 14th ed. Association of Official Analytical Chemists. Washington D. C., USA, 28 : 200-2224.
- 12.Blank, P.J., 1996 Characterization of the tea fungus metabolites. *Biotech. Lett.* 18(2):139-142.
- 13.Center for Disease Control(USA),1995. Unexplained severe illness possibly associated with consumption of kombucha tea-Iowa, 1995. Morbidity and mortality weekly Report, 44, 48:892-900.
- 14.Decker, E. A., Welch, B., 1990. Role of ferritin as a lipid oxidation catalyst in muscle food. *J. Agric. Food Chem.* 38:674-677.
- 15.Dipti, P., Yogesh, B., Kain, A.K., Pauline, T., Anju, B., Sairam, M., Singh, B., Mongis, S.S., Devendra Kumar, G.I., & Selvamurthy, W., 2003. Lead induced oxidative stress: Beneficial effects of kombucha tea. *Biomedical and Environmental Sciences*, 16, 276-282.
- 16.Dubois, M., 1956. Colorimetric method for determination of sugar and related substances. *Anal. Chem.* 28:350-356.
- 17.Dufresne, C., & Carnworth, E., 2000. Tea, kombucha, and health: a review. *Food Research International*, 33, 409-421.
- 18.Frank, G. W., 1995. Personal experience. In “ Kombucha:Healthy beverage and natural remedy from the far East. ”, 9th ed., pp.128-142. Ennsthaler Verlag, Steyr.
- 19.Furukawa, M., Sugita, M., Kojima, T. and Hayashi, S., 1973. Antitumor effect of polysaccharides and its relationship to biogenetic amines in tumor tissue under vitamin B6 deficient feeding. *Chem. Pharm. Bull.*21(9): 2090-2093.
- 20.G.L. Miller, 1959. “ Use of dinitrosalicylic acid reagent for determination of reducing sugar, ” *Anal. Chem.*, 31(3):426~428.
- 21.Gordon, M. H., 1996. Dietary antioxidants in disease prevention. *Nat. Prod. Rep.* 13: 265-273.
- 22.Halliwell, B., Murcia, M. A., Chirico, S. and Aruoma, O. I. 1995. Free radicals and antioxidants in food and in vivo: What they do and how they work. *Crit. Rev. Food Sci. Nutr.*35:7-20.
- 23.Hartmann, A.M., Burleson, I. E., Holmes, A.K., & Geist, C.R.,2000. Effects of chronic kombucha ingestion on open field behaviors, appetitive behaviors, and organs in C57 mice: a pilot study. *nutrition*, 16, 755-761.
- 24.Hiroshi, H., Satoshi, M., Tomoaki, W., Senzo, S., Fusao, U., Masao, D. and Masao, B., 1978. Microbial flora and organic acid contents in tea fungus. *Food Microbiology* 19(3):273-281.
- 25.Ikekawa, T., Uehara, N., Maeda, Y., Nakanishi, M. and Fukuoka, F., 1969. Antitumor activity of aqueous extracts of edible mushrooms. *Cancer Res.* 29: 734-735.
- 26.Ikekawa T, Maruyama H, Miyano T, Okura A, Sawasaki Y, Naito K, Kawamura K, Shiratori K., 1985. Proflamin, a New Antitumor Agent-Preparation, Physicochemical Properties and Antitumor Activity. *Jap J Cancer Res.* 76(2):142-148.
- 27.Ikekawa T., 1995. Enokitake, *Flammulina Velutipes* - Host-Mediated Antitumor Polysaccharides. *Food Rev Int.* 11(1):203-206.
- 28.Jay, J.M., 1992. *Modern food microbiology*. 4th ed. New York: AVI.
- 29.Jayabalan R, Marimuthu S, Swaminathan K., 2007. Changes in content of organic acids and tea polyphenols during kombucha tea fermentation. *Food Chemistry* 102: 394 – 395.
- 30.Moncada, S. and Higgs, A. 1993. The L-arginine-nitric oxide pathway. *N. Engl. J. Med.* 329:2002-2011.
- 31.Oyaizu, M., 1986. Studies on products of browning reaction : Antioxidative activities of products of browning reaction prepared from glucosamine. *Jpn. J. Nutri.*44: 307-314.
- 32.Reiss, J.,1994. Influence of different sugars on the metabolism of the tea fungus. *Zeitschrift Lebensmitteluntersuchung und-Forschung A*, 198(3), 258-261.
- 33.Roussin, M.R., 1996. Analyses of kombucha ferments: report on growers. Salt lake city, Utah: Information resources, LC.
- 34.Shimada, K., Fujikawa, K., Yahara K. Nakamura, T., 1992. Antioxidative properties of xanthan on the autoxidation of soybean oil in cyclodextrin emulsion. *J. Agric. Food Chem.* 40(6): 945-948.
- 35.Sievers, M., Lanini, C., Weber, A., Schuler-Schmid, U., & Teuber, M., 1995. Microbiology and fermentation balance in a kombucha beverage obtained from a tea fungus fermentation. *Systematic and applied microbiology*, 18, 590-594.
- 36.Tsuru, R., Shinomiya, N., Katsura, Y., Gotoh, M., Noritake, M. and Nomoto, K., 1991. Effects of combined therapies with protein-bound polysaccharide (PSK, Krestin) and fluorinated pyrimidine derivatives on experimental liver metastases and on the immunologic capacities of the hosts. *Oncology.* 48:498-504.
- 37.Ueno, Y., Kohko, Y., Sajamaki, S., Itoh, Y., Tajahashi, M., Hirayama, Y. and Niitsu, Y., 1994. Immunochemotherapy in B-16-melanoma-cell transplanted mice with combinations of interleukin-2, cyclophosphamide, and PSK. *Oncology.* 51:296-302.
- 38.Wang, H.X., Liu, W.k., Ng, T.B., Ooi, V.E.C. and Chang, S.T., 1995. Immunomodulatory and antitumor activities of a polysaccharide-peptide complex from a mycelia culture of *Tricholoma* sp., a local edible mushroom. *Life Sci.* 57(3):269-281.
- 39.Yoshioka, Y., Sano, T. and Ikekawa, T., 1973. Studies on antitumor polysaccharides of *Flammulina velutipes*(Curt. Ex Fr.)Sing. *I. Chem. Pharm. Bull.* 21(8): 1772-1776.
- 40.Yoshioka, Y., Tabeta, R., Saito, H., Uehara, N. and Fukuoka, F., 1985. Antitumor polysaccharides from *P.ostreatus*(FR.)QUEL.: isolation and structure of  $\beta$ -glucan. *Carbohydr. Res.* 140:93-100.