Effects Of Medium Composition And Fed-batch Cultures Control On Polysaccharide Production By Phellinus igniarius Ferment

唐溱筑、柳源德、林芳儀

E-mail: 364877@mail.dyu.edu.tw

ABSTRACT

Phellinus igniarius is a yellowish-orange parasitic mushroom commonly found on the mulberry tree. P. igniarius are rich in polysaccharides. Many studies have pointed out that P. igniarius ’ s polysaccharides demonstrate antitumor activity, and have other medicinal properties such as: anti-cancer, hypertension reduction, down- regulation of cholesterol and stimulation of immune efficacy. In this study we investigate the effects of different carbon and nitrogen sources on the mycelium biomass and exopolysaccharide (EPS) production in cultures of P. igniarius in a shake flask. Then, we used 5L fed-batch fermenters to investigate the effects of the culture conditions on the mycelium biomass and exopolysaccharide production by the fermentation of P. igniarius for further assessment of the antibacterial and anti-oxidation activity. The results show that P. igniarius ’ s optimal carbon and nitrogen sources were glucose and yeast extract, respectively. Employing glucose as a carbon source, we obtained an additional mycelium biomass of 11.29 mg/mL and exopolysaccharide production was increased by 6.38 mg/mL. Using yeast extract as a nitrogen source, we obtained an additional mycelium biomass of 8.5 mg/mL and exopolysaccharide production was increased by 0.81 mg/mL. We then investigated the effects on mycelium biomass and exopolysaccharide production by fermentation of P. igniarius at different temperatures and different initial pH in a 5L Mechanical Agitating Fermentor. The results showed that when the culture temperature was at 25 ℃ we obtained the greatest boost in mycelium biomass and exopolysaccharide production. It was also found that the optimal pH levels for the culture were pH 6 and pH 5.5; products of each condition were 11.00 mg/mL and 7.21 mg/mL, respectively. To measure in vitro antioxidant activity, the DPPH radical scavenging assay was employed. When the intracellular polysaccharide (IPS) concentration was 10mg/mL, the highest scavenging ratio was up to 86.9%. In the reducing power test, absorbance values were up to 1.57 when the exopolysaccharide concentration was at 10mg/mL. And finally, in the ferrous ion chelating ability test, when the exopolysaccharide concentration was 10mg/mL, the ferrous ion chelating ability reached to 93.5%.

Keywords : Phellinus igniarius、Fed-batch fermentations、antioxidant activity

Table of Contents

REFERENCES

參考文獻 1.丁偉。2007。桑黃菌生物學特性研究。青島農業大學。碩士論文。 2.王欽博。2011。桑黃抗氧化活性成分的篩選及其分離純化。上海師範大學生命與環境科學學院。碩士論文。 3.回晶、李輝、朱春玉、李其久、胡鳳慶。2009。桑黃子實體與菌絲體營養成分的比較分析。Special Wild Economic Animal and Plant Research。2:59-61。 4.何丹、範雪嬌、楊鳴鳴、所起鳳、杜文婷、劉戟。2011。洋蔥總黃酮透過血腦屏障抑制腦膠質瘤的作用研究。中藥藥理與臨床 24(3)84-87。 5.吳子瑋。2009。不同型式發酵槽深層培養桑黃菌 Phellinus linteus 生產胞外多醣之研究。東海大學食品科學研究所。碩士論文。 6.宋柳徵、張佩。2010。桑黃多糖對免疫細胞調節作用研究進展。中國現代醫生。48(12):23-24。 7.李宜明、沈業壽、季俊虬。2006。桑黃菌質多糖體外抑瘤及抗環磷醯胺致突變的作用。中國科學技術大學學。36(7): 700-703。 8.周長文、王芳、陶淑玲。2010。藥用植物桑黃抑制高血糖症的研究。荷澤醫學專科學校學報。22(1):1-2。 9.孟慶龍、潘景芝、陳麗、王琦。2011。桑黃胞內及胞外多醣抗癌作用的研究。時珍國醫國藥。22(5):1130-1132。 10.孟慶龍、潘景芝、陳麗、王琦。2011。桑黃發酵產物的抑菌作用。食品科學 32(3):56-59。 11.姜明、劉岩、王偉功、趙桂雲。2010。不同pH值和培養基對桑黃菌絲生長的影響。食用菌學報。8:192-194。 12.胡金霞、楊焱、張勁松、唐慶九、劉豔芳、都瑞霞、馮娜。2009。桑黃醇提物抗氧化和保護神經細胞損傷的研究。上海農業學報25(2):58-61。 13.孫錦秀、戈延茹、夏國華、王曉芳。2011。不同來源桑黃子實體總黃酮含量的比較。中外健康文摘。8(15):144。 14.張益鳴。2008。培養pH對液態醱酵生產桑黃多醣體之影響及其抗氧化特性之研究。明新科技大學。碩士論文。 15.張萬國、胡晉紅、蔡溱。2002。 桑黃增強人外周血單個核細胞產生干擾素的研究。基層中藥雜誌。16(3):5-6。 16.梁志弘。2009。桑黃之液態培養及其生理活性。國立中興大學食品暨應用生物科技學系研究所。博士論文。 17.梁佳、孫夢伊、張騰、裴浩宏、張萌萌、范桂枝。2011。響應曲面法優化桑黃菌絲體中三?蔽熒L波提取工藝。中國農學通報。27(10):235-238。 18.莫順燕、楊永春、石建功。2003a。桑黃化學成分研究。中國中藥雜誌。28:339-341。 19.莫順燕、楊永春、石建功。2003b。桑黃黃酮A和B的分離與合成。化學學報。61:1161。 20.陳欣、龔蘭、劉冠卉。2010。食用真菌多糖提取條件的優化及其還原力的比較。食品科學。31(14):140-144。 21.陳奕廷。2005。 探討 pH 值和通氣量對 Penicillium brevicompactum 生產Mycophenolic acid 之影響。國立中央大學化學工程與材料工程研究所。碩士論文。 22.陳建男。2007。啤酒花抗氧化成分之研究。私立大同大學生物工程研究所。碩士論文。 23.傅海慶、陳紹軍、駱文燦、陳漢清、林河通。2007。桑黃菌液體發酵培養研究。中國食品學報。3(7):58-63。 24.曾念開、王秋穎、蘇明聲、王懷凱。2007。營養及環境因素對鮑氏針層孔菌菌絲生長的影響。食用菌學報。 4:6~8。 25.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。 26.翟瓊、馮月飛。2011。真菌桑黃的研究進展。華章。(6):280。 27.趙瀾、張紅鋒。桑黃粗多醣對腫瘤細胞增殖及轉移相關能力的抑制作用。2008。東華師範大學學報。2:78-84。 28.劉金榮、江發壽、李艷、洪成林、曹永翔、趙文斌。1998。藥用真菌桑黃甾類成分的提取和鑑定。農墾醫學。20:141。 29.劉艷芳、楊焱、賈葳、張勁松、唐九慶、唐傳紅。2006。藥用真菌桑黃總黃酮測定方法研究。食用菌學報。13(2):45-48。 30.龍明有。2006。桑黃醱酵生產多醣體及醱酵產物抗氧化特性研究。明新科技大學化學工程系。碩士論文。 31.謝江甯、宋素芬、李香、徐秀泉。2012。桑黃總三?蔽煽ㄗ?峔靼擱~抗腦膠質瘤U251活性。中國實驗方劑學雜誌18 (5): 24~26。 32.羅國晏。2008。深層培養條件及發酵槽種類對桑黃菌Phellinus igniarius 胞外多醣體產量及生物活性之影響。私立東海大學食品科學系研究所。碩士論文。 33.Brown, G. D., and Gordon, S. 2003. Fungal β-glucans and mammalian immunity. Immunity. 19(3):311 – 315. 34.Chi, J. H., T. M. Ha., Y. H. Kim., Y. D. Rho., 1996. Studies on the mainfactors affecting the mycelial growth of Phellinus linteus. Korean Journal of Medical Mycology. 24:214-222. 35.Guo. X., Zou. X., Sun. M., 2010, Optimization of extraction process by response surface methodologyand preliminary characterization of polysaccharides from Phellinus igniarius. Carbohydrate Polymers. 80:344-349. 36.Hwang. H. J., Kim. S. W., Lim. J. M., Joo. J. H., Kim. H. O., Kim. H. M., Yun. J. W., 2005. Hypoglycemic effect of crude exopolysaccharides produced by a medicinal mushroom Phellinus baumii in streptozotocin-induced diabetic rats. Life Sciences. 76:3069-3080. 37.Kim. B. C., Choi. J. W., Hong. H. Y., Lee. S. A., Hong. S., Park. E. H., Kim. S. J., Lim. C. J., 2006. Heme oxygenase-1 mediates the anti-inflammatory effect of mushroom Phellinus linteus in LPS-stimulated RAW 264.7 macrophages. Journal of Ethnopharmacology. 106:364 – 371. 38.Kim. G. Y., Roh. S. I., Park S. K., Ahn S. C., Oh. Y. H., Lee. J. D., Park. Y. M., 2003. Alleviation of Experimental Septic Shock in Mice by Acidic Polysaccharide Isolated from the Medicinal Mushroom Phellinus linteus. Biol Pharm Bull. 26(10):1418—1423. 39.Ko, Y. T. and Y. L. Lin. 2004. 1, 3-β-glucan quantification by a fluorescence microassay and analysis of its distrivution in foods. Journal of Agricultural and Food Chemistry. 52:3313-3318. 40.Liu. Q. N., Liu. R. S., Wang. Y. H., Mi. Z. Y., Li. D. S., Zhong. J. J., Tang. Y. J., 2009. Fed-batch fermentation of Tuber melanosporum for the hyperproduction of mycelia and bioactive Tuber polysaccharides. Bioresource Technology. 100:3644-3649. 41.Luo. J., Liu. J., Ke. C., Qiao. D., Ye. H., Sun. Y., Zeng. X., 2009. Optimization of medium composition for the production of exopolysaccharides from Phellinus baumii Pilat in submerged culture and the immuno-stimulating activity of exopolysaccharides. Carbohydrate Polymers.78:409 – 415. 42.Mantovani. M. S., Bellini. M. F., Angeli. J. P. F., Oliveira R. J., Silva. A. F., Ribeiro. L. R., 2008. β-Glucans in promoting health: Prevention against mutation and cancer. Mutation Research. 658:154 – 161. 43.Novak. M., Vetvicka. V., 2009. Glucans as Biological Response Modifiers. Endocrine, Metabolic & Immune Disorders—Drug Targets. 9:67-75. 44.Papinutti. L., 2010. Effects of nutrients, pH and water potential on exopolysaccharides production by a fungal strain belonging to Ganoderma lucidum complex. Bioresource Technology. 101:1941 – 1946. 45.Sheng. J., Yu. F., Xin. Z., Zhao. L., Zhu. X., Hu. Q. 2007. Preparation, identification and their antitumor activities in vitro of polysaccharides from

Chlorella pyrenoidosa. Food Chemistry. 105:533 – 539. 46.Shih. I. L., Chou. B. W., Chen. C. C., Wu. J. Y., Hsieh. C., 2008. Study of mycelial growth and bioactive polysaccharide production in batch and fed-batch culture of Grifola frondosa. Bioresource Technology. 99:785-793. 47.Song. T. Y., Lin. H C., Yang. N. C., Hu. M. L., 2008. Antiproliferative and antimetastatic effects of the ethanolic extract of Phellinus igniarius (Linnearus: Fries) Quelet. Journal of Ethnopharmacology. 115:50 – 56. 48.Dinis. T. C. P., Madeira. V. M. C., Almeids. L. M., 1994. Action of phenolic derivatives (acetaminophen, salicylate, and 5-aminosalicylate) as inhibitors of membrane lipid peroxidation and as peroxyl radical scavengers. Arch Biochem Biophys. 315(1):161-9. 49.Wang, Y. J., Yao, S. J., Guan, Y. X., Wu, T. X., Kennedy. J. F., 2005. A novel process for preparation of (1→3)- β -D-glucan sulphate by a heterogeneous reaction and its structural elucidation. Carbohydrate Polymers. 59(1): 93-99. 50.Xu. W., Zhang. F., Luo. Y., Maa L., Kou. X., Huang. K., 2009. Antioxidant activity of a water-soluble polysaccharide purified from Pteridium aquilinum. Carbohydrate Research. 344:217 – 222. 51.Zou. X., Sun. M., Guo. X., 2006. Quantitative response of cell growth and polysaccharide biosynthesis by the medicinal mushroom Phellinus linteus to NaCl in the medium. Microbiology and Biotechnology. 22:1129 – 1133.