

槐花中黄酮类物质提取工艺的研究

丁利君, 吴振辉, 蔡创海

(韩山师范学院)

摘要: 该文研究了水提法、传统的醇回流法提取槐花总黄酮类化合物的提取工艺, 并使用正交试验确定了最佳提取条件。试验结果表明: 槐花中总黄酮类物质的最佳提取条件为 B2C3A2, 即用 2 倍量 40% 的乙醇连续回流提取 3 次(每次 2 h)。

关键词: 槐花; 最佳提取条件; 回流; 正交试验法; 黄酮类化合物

中图分类号: S377

文献标识码: A

文章编号: 100226819(2002)0120142203

槐花 (*Pogonatum chinense* flowers) 为豆科植物槐 (*Sophora japonica* L.) 的干燥花及花蕾, 我国大部分地区都有生长。槐花中含有丰富的芸香甙(芦丁, rutin), 三萜皂甙, 槐花米甲素、乙素、丙素等多种物质。槐花米甲素是和芸香苷不同的黄酮类物质, 它在花蕾中的含量为 14% 左右。据医书记载, 槐花有多种医疗保健功能, 如《日华子本草》中说槐花“治五痔, 心疼, 眼疾”; 《本草纲目》中说“炒香频嚼, 治失音及喉痹”; 《医林纂要》中记载其“泻肺逆, 泻心火, 清肝火, 坚肾水”^[1]。

现代医学研究表明, 黄酮类物质具有降低心肌耗氧量, 使冠脉、脑血管流量增加, 抗心律、软化血管, 降血糖、血脂, 抗氧化, 消除抗体内自由基、抗衰老作用, 增加机体免疫力^[1,2]。有关文献报道了从葛根、水芹、苦荞麦、银杏叶等中提取黄酮类物质^[2~7], 而对于槐花中黄酮类物质的提取尚未见有文献报道。本文对槐花中黄酮类物质的提取方法(主要研究了水提法和醇提法)进行了研究。槐花中黄酮类物质的提取及含量的测定, 有利于更深入地研究槐花的药理与保健作用, 为槐花的开发评价槐花的质量提供了理论依据和检测手段。

1 材料与方法

1.1 材料和仪器

材料: 槐花, 购于潮州市中药店; NaNO₂, AL(NO₃)₃, NaOH, 95% 乙醇(均为 A. R 级)。

仪器: 724 分光光度计(上海光学仪器厂); 光电分析天平 TG328B 型(上海第二天平仪器厂); 干燥箱 101A 1 型(上海市实验仪器总厂); 电热套; 离心机等。

1.2 黄酮物质提取方法及含量测定

1.2.1 槐花中黄酮类物质的提取方法

1) 醇提法: 材料 干燥 粉碎 醇回流 抽滤 离心 离心液合并 浓缩 定容 测定吸光度, 计算总黄酮类物质的含量。

2) 温浸法: 分别在 50、60、70、80 和 90 等不同温度下, 各取 5 g 槐花干料进行水浴恒温, 以蒸馏水作溶媒, 第一次用 10 倍溶媒温浸 1 h, 第二次 8 倍溶媒温浸 0.5 h, 合并二次滤液并浓缩, 然后于 50 mL 容量瓶定容。

3) 煎煮法: 取 5 g 槐花干料, 以蒸馏水作溶媒, 分别用 10 倍溶媒和 8 倍溶媒于电热器直接加热, 保持微沸, 第一次加热 1 h, 第二次 0.5 h。合并二次滤液并浓缩, 然后于 50 mL 容量瓶定容。

4) 加碱浸提法: 各取 5 g 槐花干料, 分别以 pH 值为 8、9、10 和 11 的 NaOH 溶液作溶媒, 在 80℃ 水浴恒温温浸。第一次用 10 倍溶媒温浸 1 h, 第二次用 8 倍溶媒温浸 0.5 h, 合并二次滤液并浓缩, 然后于 50 mL 容量瓶定容。

1.2.2 工作曲线回归方程的确定^[3,4]

配制一系列不同浓度的芦丁标准溶液于波长 $K = 500 \text{ nm}$ 处测定吸光度, 依芦丁浓度——吸光度标准工作曲线, 得出回归方程:

$$Y = 0.1026A - 0.00195 \quad \text{相关系数 } r = 0.9999$$

Y —芦丁溶液的浓度, g/L; A —溶液的吸光度。

1.2.3 待测液的配制及黄酮类物质的测定

将提取液转移至 100 mL 容量瓶中, 用与提取时间同浓度的乙醇定容, 摇匀。取 0.4 mL 定容后的提取液置于 10 mL 容量瓶中, 用 30% 乙醇补充至 5 mL, 加入 0.28 mL NaNO₂ (1:20), 摇匀, 放置 5 min 后加入 0.28 mL AL(NO₃)₃ (1:10), 6 min 后再加入 2 mL 1 mol/L 的 NaOH 溶液, 混匀, 用 30% 乙醇稀释至刻度, 10 min 后于 500 nm 处的波

长测定吸光度, 试剂为空白。

$$\text{总黄酮类物质含量}(\%) = [(Y \times 10 \times 100) \div (0.4 \times 5 \times 1000)] \times 100 = Y \times 50$$

Y: 根据所测定溶液的吸光度值, 用回归方程计算出的相对应的总黄酮类物质含量。

1.3 回流中主要影响因素的确定

在材料粉碎度以及所用溶剂一定条件下, 溶剂的浓度、回流的时间、提取温度以及溶媒量等因素都会对提取率产生影响, 试验选用乙醇为提取剂, 由于在回流时的温度难以控制, 因此本试验只讨论了乙醇浓度、乙醇用量和回流时间对提取率的影响。

2 结果与分析

2.1 乙醇浓度对醇提法提取效果的影响

分别用 20%、40%、60%、80% 的乙醇, 以 60 mL 为 1 倍量对槐花 10 g 进行回流提取 1 h, 提取液按照 1.2.3 试验步骤操作。结果如表 1。

表 1 乙醇浓度对槐花中黄酮类物质提取效果的影响

浓度/%	20	40	60	80
吸光度	1.769	2.116	1.897	1.886

试验表明: 槐花在浓度为 40% 的乙醇回流时所测得的吸光度最高。

2.2 不同溶媒量对醇提法提取效果的影响

分别用 1、2、3 倍量的乙醇对槐花中黄酮类物质进行提取 1 h, 试验结果如表 2。试验结果表明: 槐花在 2 倍量溶媒时吸光度较高。

表 2 不同溶媒量对槐花中黄酮类物质的提取效果

溶媒量	1 倍	2 倍	3 倍
吸光度	2.116	2.520	2.350

2.3 不同时间对醇提法提取效果的影响

槐花以溶媒量为 2 倍量 40% 的乙醇从中提取总黄酮类物质。试验结果如表 3。试验结果表明: 物料回流 2 h 的提取效果最好, 往后随着时间增加, 吸光度反而降低, 这可能是回流时间太长, 有部分乙醇被挥发, 而导致沸点逐渐增大, 从而破坏某些黄酮类化合物。

2.4 水提法的提取效果

2.4.1 不同水浴温度对温浸法提取效果的影响

由图 1 可知, 各取 5 g 干槐花 5 份, 各加蒸馏水 50 mL 和 30 mL 于 50、60、70、80 和 90

表 3 不同回流提取时间的提取情况

Table 3 Effect of backwatering time on extracting result of Flavonoid

时间/h	1	2	3	4
吸光度	2.177	2.420	1.943	1.527

进行温浸提取两次时, 随着温度的升高, 提取液中的总黄酮类物质也增加, 即温度提高温浸效果较好, 主要是由于黄酮类物质易溶于热水。

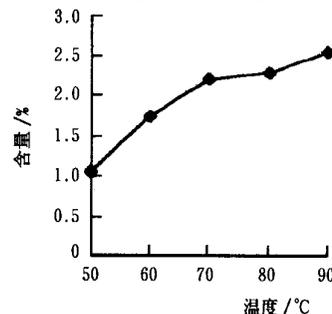


图 1 温度对温浸法的影响

Fig. 1 Effect of temperature on water extracting method

2.4.2 不同 pH 值对温浸法提取效果的影响

由图 2 可知, 各取 5 g 干槐花 4 份, 在 80 °C 恒温水浴分别以 pH 值为 8、9、10、11 的 NaOH 溶液分两次温浸 1 h 和 0.5 h。pH 值降低时, 由于提取不完全, 提取液中的总黄酮类物质含量较低; pH 值为 11 时虽然黄酮含量较高, 但含有的 NaOH 溶质较多而造成分析结果出现干扰, 因此 pH 值为 10 时的加碱温浸法效果最好。

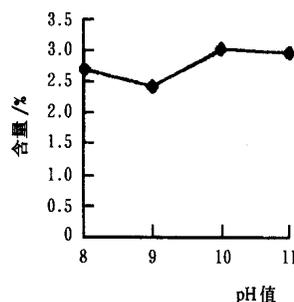


图 2 80 °C 水温条件下 pH 值对温浸法的影响

Fig. 2 Effect of pH value on water extracting method

2.4.3 不同水提法对提取效果的影响

由图 3 可知, 取干槐花 5 g 3 份, 分别以蒸馏水作溶媒于 90 °C 恒温水浴、以蒸馏水作溶媒于电热器上直接加热、以 pH 值为 10 的 NaOH 溶液于 80 °C 恒温水浴各提取 1 h 和 0.5 h。以 80 °C 恒温水浴, pH 值为 10 的加碱温浸法提取效果最好; 90 °C 水浴温浸法的效果次之, 煎煮法最差。这主要是煎煮法的过程中, 溶剂比较容易蒸发, 损失较多, 造成用煎煮法提取液中总黄酮类物质含量最低。

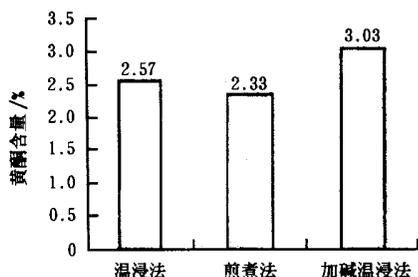


图 3 不同水提法效果比较

Fig 3 Comparison of different water extracting method

2 5 正交试验确定提取的最佳条件

比较了水提方法与醇提法,醇提法效果较好。为全面考查醇提法影响因素,设计了溶媒量、溶媒浓度和提取次数的 3 因素 3 水平正交试验,结果见表 4。

试验结果及方差分析表明:乙醇回流法从槐花中提取总黄酮类化合物的最佳条件为 B2C3A 2,即用 2 倍量 40% 乙醇回流 3 次(每次 2 h),影响因素的大小依次为:乙醇浓度、提取次数、溶媒量;此法提取的槐花中总黄酮类物质含量为 13.04%。

表 4 L₉3³ 的醇提法正交试验结果及分析

Table 4 The result and analysis of L₉3³ orthogonal experiment of alcohol extracting method

因素	溶媒量(倍)	溶媒浓度ö%	提取次数	吸光度	测定液中总黄酮含量 Y _{ögl} L ⁻¹	总黄酮类物质提取率(相对被提取槐花质量)ö%
1	1(1)	20(1)	2(2)	2.507	0.2553	12.77
2	1(1)	40(2)	1(1)	2.301	0.2341	11.71
3	1(1)	60(3)	3(3)	2.456	0.2500	12.50
4	2(2)	20(1)	1(1)	1.485	0.1504	7.52
5	2(2)	40(2)	3(3)	2.560	0.2607	13.04
6	2(2)	60(3)	2(2)	2.320	0.2360	11.80
7	3(3)	20(1)	3(3)	1.875	0.1924	9.62
8	3(3)	40(2)	2(2)	2.260	0.2300	11.50
9	3(3)	60(3)	1(1)	2.248	0.2287	11.44

因素	溶媒量(A)	溶媒浓度(B)	提取次数(C)
E	18.47	14.91	15.32
°	16.16	18.08	18.01
,	16.21	17.85	17.51
Q	288.35	289.27	288.55
S	1.16	2.08	1.36
P	287.19		

方差来源	平方和(S)	自由度(f)	均方 s öf	F	临界值
溶媒量	1.16	2	0.58	1	F _{0.05} (2,2) = 19
乙醇浓度	2.08	2	1.04	1.79	F _{0.01} (2,2) = 99
提取次数	1.36	2	0.68	1.17	F _{0.1} (2,2) = 9
总和	4.60	6			F _{0.2} (2,2) = 4

3 结 论

对槐花中黄酮类物质的提取,水提法以 80 的恒温水浴, pH 值为 10 的氢氧化钠液提取效果最好。

醇提法的正交试验的结果表明:槐花中黄酮类物质的最佳提取条件是: B2C3A 2,即用 2 倍量 40% 乙醇回流 3 次(每次 2 h)。醇提法提取总黄酮类物质的含量是 13.04%,而碱提法只有 6.06%。所以,槐花中黄酮类物质的最佳提取方法是醇提法。此法的提取工艺简单,条件容易控制,提取率高,提取剂乙醇易除净,所以可将此工艺应用于工业化生产,所生产的提取液可浓缩成膏状物,也可直接将其添加到保健饮料、糖果、绿豆羹、饼干或口服液中,作为保健食品、营养食品和功能食品的原料,应用前景良好。

[参 考 文 献]

- [1] 江苏新医学院编 中药大辞典[M] 上海:上海科技出版社,1997. 2433~ 2434
- [2] 郭建平等 葛根药理作用研究进展[J] 中草药,1995 (3): 163~ 165
- [3] 曾祥群 葛根总黄酮提取工艺[J] 食品工业科技,2000 (3): 33~ 34
- [4] 陈运中 苦荞麦黄酮含量的测定[J] 食品科学,1998 (3): 54~ 56
- [5] 郑玉之等 银杏叶提取物及其在保健食品中的应用[J] 食品工业科技,1996(2): 47~ 48
- [6] 毕丽君等 水芹中总黄酮类化合物最佳提取工艺的研究[J] 食品科学,1999(12): 35~ 37
- [7] 韩鲁佳等 黄芩甙提取分离方法及工艺研究[J] 农业工程学报,2000,16(6): 118~ 122

Key words: maltogenase; Enzyme; extremely high maltose syrup

Starch Paste Clarity and Its Influence Factors (129)

Du Xianfeng¹, Xu Shiyong², Wang Zhang² (1. *Department of Food Engineering, Anhui Agricultural University, Hefei 230036, China;* 2. *School of Food Science, Wuxi University of Light Industry, Wuxi 214036, China*)

Abstract: Starch paste clarity and the effects of molecular structure, retrogradation and some food ingredients such as NaCl, sucrose and citric acid were investigated. The experimental results indicate that the starch molecular structure is the principal factor to influence the starch paste clarity. NaCl was proved to be able to reduce starch paste clarity, and sucrose, citric acid are proved to be able to increase the clarity. During the initial storage period (4, 0~4 d), the clarity decreased abruptly, then it gradually decreased to its minimum as the leached but amylose completely formed a three-dimensional network with amylopectin embedded in and reinforced to the interpenetrating amylose gel matrix.

Key words: starch paste; clarity; molecular structure; food ingredient; retrogradation

Experimental Study on Extruded Rice Used as Beer Adjunct (132)

Shen Dechao, Meng Yang (*Engineering Technology College, Northeast Agricultural University, Harbin 150030, China*)

Abstract: In this paper the influence of the parameters of the extrusion system of rice used as beer adjunct on indexes observed was studied by experiments in the laboratory. These parameters are the diameter of nozzle, barrel temperature, moisture content of rice and screw speed. The indexes include total reduced sugar concentration and filtration rate. The research results indicate that the extruded rice can be used as beer adjunct.

Key words: rice; extrusion; beer adjunct

Rheological Law of the Crop Stem Fibrous Material During Compression Process ... (135)

Yang Mingshao, Zhang Yong, Li Xuying (*Inner Mongolia Agricultural and Husbandry University, Hohhot 010018, China*)

Abstract: The exploitation of the loose crop material needs compression process. To determine the structural parameters and optimize the design of compression equipment, the law of compression process, the relationship among main parameters in the whole compression process and the characteristics of the material to be compressed are investigated. Based on the achievement acquired by our research group, a basic law of the stress and strain variation pattern was obtained by use of advanced measuring means, which provided a theoretical basis for the further study and optimum design of compression equipment.

Key words: crop material; rheology; compression

Extraction Technology of Soluble Polysaccharides From Wild Patrinia Villosa (138)

Zhu Jiajin (*College of Agricultural Engineering and Food Science, Zhejiang University, Hangzhou 310029, China*)

Abstract: Patrinia villosa (P. V.) can cure diarrhea, but it can also do good to constipation. Not only the leaves and the stems can relax the constipation, but also the water-solution extracted from P. V. has the similar function. In order to know the effective ingredients to prevent and cure constipation extracted in the water-solution, the extraction technology of soluble polysaccharides from wild P. V. was studied, and single factor test and orthogonal experiment design methods ($L_9(3^3)$) were applied to analyze the influence of each factor in solid-liquid ratio, temperature and time on the extraction percent of the polysaccharides from the water solution of P. V.. Experimental results indicated that temperature and solid-liquid ratio significantly affect the extraction percent of the soluble polysaccharides. The optimum solid-liquid ratio is 1:20, temperature is 100 and the extraction time is 6 hours. In this condition, the extraction percent of soluble polysaccharides is 25.8%.

Key words: patrinia villosa; soluble polysaccharides; extraction technology; content measurement; phenol-sulfate method; constipation

Extraction of Flavonoid From Pagodatree Flower (142)

Ding Lijun, Wu Zhenhui, Cai Chuanghai (Department of Biology and Chemistry, H anshan N om al Institute, Chaozhou, Guangdong 521041, China)

Abstract: The extraction of the flavonoid from Pagodatree flower with water system and alcohol-water backwatering system was studied. With water extracting method, the effect of temperature, pH value, and different technology was studied. With alcohol extracting method, through the orthogonal experiment, the optimum technical condition is obtained. The results showed that the optimum technology of extracting flavonoid from the Pagodatree flowers is B2C3A2, namely, 2 times volume 40% alcohol-water, 3 extracting times (2 h each time), and the content of flavonoid is 13.04%.

Key words: pagodatree flowers; optimum technical conditions for extraction; backwatering; orthogonal experiment; flavonoid

· Agricultural Electronics and Information Technology ·

Information Processing System for Precision Agriculture Based on GPS and GIS ... (145)

He Yong, Fang Hui, Feng Lei (College of Agricultural Engineering and Food Science, Zhejiang University, Hangzhou 310029, China)

Abstract: To acquire spatial information quickly and analyze it properly, an information processing system which uses MapObjects to integrate GPS with GIS is researched. This information processing system has the function of visualizing and analyzing spatial data, showing and tracking moving point and calculating area of closed polygon. It can integrate GPS and GIS into Expert System and Decision Support System perfectly. A new way of information processing and a new model of information processing system for the research of precision agriculture are put forward.

Key words: precision agriculture; GIS; GPS; information processing system

Fuzzy Pattern Recognition Method Based on Image Contour Line (150)

Yu Qingcang¹, Yan Hongbin² (1. College of Agricultural Engineering and Food Science, Zhejiang University Hangzhou 310033, China; 2. Zhejiang Power System Training and Education Center, Hangzhou 310029, China)

Abstract: A simplified method was put forward on a binary image contour line extraction. Distance between centroid and dots on contour line was calculated, and based on the maximum centroid-dot distance, a feature vector including twelve direction segments and lengths of contour line was studied in this paper, and two distance algorithms based on euclid distance were promoted. The experimental results show that the correct recognition ratios on three kinds of paddy seed reach respectively 79.89%, 89.63% and 93.27%. This method can also be applied to fuzzy pattern recognition on fruits, machine parts etc. As random of object's lay direction and side were taken into account in this paper, and objects are recognized at the same direction and on the same side, so this method can also be promoted to the optic measurements on object rotation angle and any preindicated geometrical parameters.

Key words: paddy seed; image; contour line; fuzzy pattern recognition

Measuring Area of Leaves Based on Computer Vision Technology by Reference Object (154)

Xu Guili, Mao Hanping, Hu Yongguang (College of Machinery Engineering, Jiangsu University, Zhenjiang, Jiangsu 212013, China)

Abstract: A method of measuring area of leaves by reference object based on computer vision technology was studied. The sampling box of live leaves was developed, and its data were optimized. The measurement process was studied. The reference object was segmented from the original image by the method of threshold. The data of threshold was gained by means of derivation. The way of removing the noises in the image was studied. Generally, the results show that this method has better feasibility, higher precision and higher efficiency.

Key words: leaf area; computer vision; method of reference object; measurement

Reversible Airflow Drying System Based on Single-Chip Computer Control (158)