A Study on the Antioxidant Activities in Different Parts of Gardenia Jasminoides

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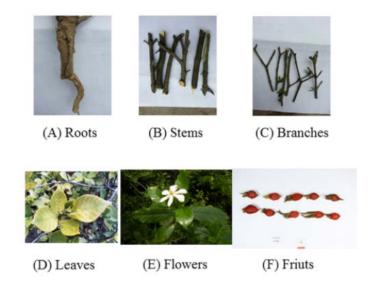
ABSTRACT.OBJECTIVE: The experiment attempts to study the antioxidant activity in different parts of Gardenia jasminoides including roots, stems, leaves, branches, flowers and fruits to explore the development and utilization of new medicinal parts of Gardenia jasminoides. METHODS: Apply microwave-assisted extraction(MAE) to extract total flavonoids from different parts of Gardenia jasminoides including roots, leaves, fruits, flowers and stems and register the ethanol concentration, and of temperature extraction time microwave-assisted extraction.RESULT: The total polyphenols content in different parts of Gardenia jasminoides was arranged from high to low as: leaves, branches, fruits, flowers, stems, roots; the total flavonoids content in different parts of Gardenia jasminoides was arranged from high to low as: leaves, Branches, fruits, stems, roots, flowers; the DPPH radical scavenging activity(IC50) in different parts of Gardenia jasminoides was arranged from high to low as: leaves, branches, fruits, flowers, stems, roots; the dry extract rate was arranged from high to low as: fruits, leaves, branches, flowers, stems, roots.

KEYWORDS: Gardenia jasminoides; antioxidant activity; component

1. Reagents and Materials

1.1 Experimental Raw Materials

The whole-plant Gardenia jasminoides Ellis was collected in Wanli, Nanchang City, Jiangxi Province in 2013 which was identified by Professor Fan Cuisheng at Jiangxi University of Traditional Chinese Medicine. The specimens are deposited in the Chinese Medicine Resources department of Jiangxi University of Traditional Chinese Medicine. The Gardenia jasminoides are divided into roots, stems, leaves, branches, flowers and fruits according to their parts, then keep them for future usage through the process of drying, flouring, and screening[1][2].



1.2 Experimental Reagent

Gallic acid (National Institute for the Control of Pharmaceutical and Biological Products, Lot No. 110813-201204), Rutin (National Institute for the Control of Pharmaceutical and Biological Products, Lot No. 110813-201204), Sodium carbonate anhydrous, Aluminium nitrate, Lithium sulfate, Bromine water (Tianjin Fengchuan Chemical Reagent Technologies Co., Ltd.), Tungstic acid, Sodium molybdate (Tianjin No.4 Chemical Reagent Factory), Concentrated phosphoric acid and Concentrated hydrochloric acid (Shantou Xinong Chemical Co., Ltd.), Sodium hydroxide, Aluminum Nitrate and Sodium Nitrite (Tianjin Hengxing Chemical Reagent Manufacturing Co., Ltd.).

1.3 Experimental Instruments

KQ-250 Ultrasound Cleaner (Kun Shan Ultrasonic Instruments Co.,Ltd.), HH-4 Digital Thermostat Water Bath Cauldron (Guohua Electrical Appliance Co., Ltd.), BS224S Electronic Balance (Beijing Sedori Instrument Co., Ltd.), 116 Swing Crusher (Ruian city, Yongli Pharmaceutical Machinery Co., Ltd.), UV-2100 Ultra Violet Spectrophotometer (Shanghai mapada Instruments Co., Ltd.), 0412-CENTRIFUGE Centrifuge (Shanghai Fangshu Instrument Factory), WKYIII-100 Adjustable Micropipette (Shanghai Jiaan Analyzer Factory), Bio-Tek Elx800 Microplate Reader(American Bio-Tek Microplate Reader).

2. Method of determination

2.1 Determination of total polyphenols in different parts of Gardenia jasminoides

Weigh roots, stems, leaves, branches, flowers and fruits of gardenia jasminoides Ellis exactly 0.5 g in 100 ml conical flask; add 20 ml of 80% ethanol; apply the approach of ultrasonic extraction for 20 minutes at room temperature; centrifuge; take a certain amount of supernatant in 10 ml Volumetric flask, add water to 4.0 ml; add 2 ml of Folin-Ciltocaen solution, shake well, keep for 5 minutes before using; add 2 ml of 10% sodium carbonate solution, shake well, store it in shade for 120min. Take 80% ethanol solution as blank group, measure the absorbance at 764 nm wavelength; calculate the concentration of the sample according to the standard curve of gallic acid and the content of total polyphenols in various parts of Gardenia jasminoides[3].

2.2 Determination of total flavonoids in different parts of Gardenia jasminoides

Weigh roots, stems, leaves, branches, flowers and fruits of gardenia jasminoides Ellis exactly 0.5 g in 100 ml conical flask; add 20 ml of 80% ethanol; apply the approach of ultrasonic extraction for 20 minutes at room temperature; centrifuge; take a certain amount of supernatant in 10 ml Volumetric flask, add water to 2.4 ml; add 0.4ml of 5% sodium nitrite solution, shake well, keep for 6 minutes before using; add 0.4ml of 0.4ml 10% aluminium nitrate solution, shake well, keep for 6 minutes before using; Add 4 ml 4.3% sodium hydroxide solution, dilute to required calibration, shake well, keep for 10-15 minutes. Take 80% ethanol solution as blank group, measure the absorbance at 508 nm wavelength; calculate the concentration of the sample according to the standard curve of fagopyrol and the content of total flavonoids in various parts of Gardenia jasminoides.

2.3 Determination of DPPH radical scavenging activity of Different Parts of Gardenia jasminoides

Take 0.02ml of certain concentration of the sample solution , add 0.18ml of 0.1 mmol/ml DPPH methanol solution, shake well; store in shade at room temperature for 30 minutes, measure the absorbance at 517 nm wavelength. Use the same concentration of methanol solution as positive control, then calculate as a percentage of DPPH scavenging free radical inhibition[4].

3 Data Processing

Each test was repeated 3 times. The DPPH radical scavenging activity in different parts of Gardenia jasminoides was evaluated by 50% inhibitory concentration (IC50) which was calculated by nonlinear regression from Windows Prism 4.0 software. The results are shown in Table 1 and Fig.1. The correlation between the contents of total polyphenols and flavonoids in different parts of Gardenia jasminoides and DPPH radical scavenging activity was analyzed. The

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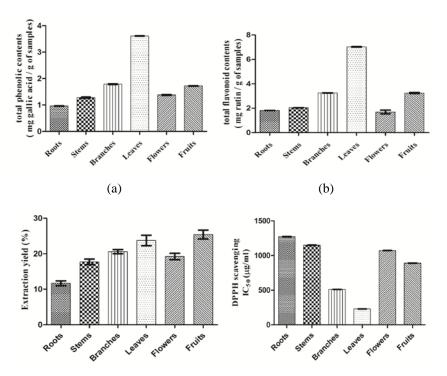
results were shown in Fig. 2.

Table 1 Total phenol and flavonoid contents, extraction yield and antioxidant activities of different parts of Gardenia jasminoides

Raw materials	TPC (mg/g)	TFC (mg/g)	DPPH radical	Yield(%)
			scavenging	
			activity	
			$IC_{50}(\mu g/mL)$	
Roots	$0.97 \pm 0.01^{**}$	$1.82 \pm 0.01^{**}$	1274.80±4.08 ^{**} ▲	11.68± 1.17**
Stems	$1.28 \pm 0.04^{**}$	2.05 ±	$1149.69 \pm 2.83^{**}$	17.72 ±
		0.01**		1.40**
leaves	$3.61 \pm 0.01^{**}$	$7.03 \pm 0.02^{**}$		23.74 ± 2.61
branches	$1.79 \pm 0.02^*$	3.26 ± 0.01	$511.75 \pm 1.32^{**}$	$20.61 \pm 1.03^*$
flowers	$1.38 \pm 0.03^{**}$	$1.69 \pm 0.15^{**}$	$1073.64 \pm 3.29^{**}$	$19.23 \pm 1.58^*$
fruits	1.72 ± 0.01	3.24 ± 0.04	890.87 ± 1.71 [▲]	25.42 ± 2.19
Trolox	-	-	10.75 ± 0.44	-

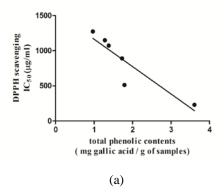
Trolox was used as a positive control, * P < 0.05, ** P < 0.001, indicating the comparison of extracts and fruit extracts.

 $\blacktriangle P < 0.05,$ indicating a comparison between the extract and water-soluble vitamin E.



(c) (d)

Fig. 1. The TPC, TFC, Yield and DPPH of different parts of Gardenia jasminoides



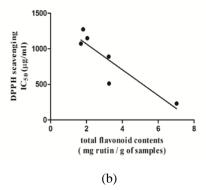


Fig.2 The relationship between DPPH radical scavenging activity and TPC,TFC of different parts extracts of Gardenia jasminoides

4. Results and Analysis

Folin-Ciocalteu and NaNO2-Al(NO3)3 were used to determine the contents of total polyphenols and flavonoids in different parts of Gardenia jasminoides (roots, stems, leaves, branches, flowers and fruits) and their antioxidant activities. According to Fig 1, the total polyphenol content in different parts of Gardenia jasminoides was arranged from high to low as(fig.1a): leaves > branches > fruits > flowers > stems > roots; the total flavone content in different parts of Gardenia jasminoides was arranged from high to low as (fig.1b): leaves > branches > fruits > stems > roots > flowers; the DPPH radical scavenging activity(IC50) in different parts of Gardenia jasminoides was arranged from high to low as (fig. 1d): leaves >

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branches > fruits > flowers > stems > roots; the extract yield was arranged from high to low as (fig.1c): fruits > leaves > Branches > flowers > stems > roots.

The contents of total polyphenols and flavonoids in different parts of Gardenia jasminoides and their antioxidant activities are greatly different. The content of total polyphenols and flavonoids in Gardenia leaves was the highest, which was 2-3 times as much as that in other aerial parts of Gardenia such as stems, branches, flowers and fruits, and 4 times as much as that in roots[5].

According to Fig 2, the antioxidant activity of extracts from different parts of Gardenia jasminoides was well correlated with the content of polyphenols and flavonoids, and the correlation coefficients were (R2) 0.8193 and (R2) 0.8096. The content of total polyphenols and total flavonoids in the extract of Gardenia jasminoides was the highest, and its DPPH radical scavenging activity (IC50) (230.68 (+1.33 ug/ml) was the strongest. The content of total polyphenols and total flavonoids in the extract of Gardenia jasminoides root was the lowest, and its DPPH free radical scavenging activity (IC50) (1274.80 (+4.08 ug/ml) was the weakest. The antioxidant activity of Gardenia leaves is the best, which provides a foundation for the development and utilization of new medicinal parts of Gardenia jasminoides.

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