



(RESEARCH ARTICLE)



## Determination of total phenolic and total flavonoid contents of *Jasminum grandiflorum* Lin

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### Abstract

The current study was focused on the standardization & validation of hydroalcoholic extract of *Jasminum grandiflorum* (JG). *Jasminum grandiflorum* Linn. Belonging to family Oleaceae is a well-known medicinal plant. It is an Ayurveda (herbal) medicinal plant. This review is to give comprehensive information on the chemical constituents and medicinal importance of *Jasminum grandiflorum*. It is commonly known as Chameli in Hindi. The process such as which involved the morphological, microscopical, and physical evaluation. Quantitative determination of phenols and flavonoid in *Jasminum grandiflorum* hydroalcoholic extract was carried out using chromatographic methods.

**Keyword:** *Jasminum grandiflorum*; Extraction; Phenols; Flavonoids

### 1. Introduction

Standardization of drugs means confirmation of its identity and determination of its quality and purity detection of nature of adulterant by various parameters like morphological, physical, chemical and biological observation. (Kumari, Rajesh, and Mita Kotecha *et al*, 2016).

Validation of an analytical method is the process by which it is established, by laboratory studies, that the performance characteristics of the method meet requirements for the intended analytical applications. The process confirm that the analytical procedure employed for a specific test is suitable for intended use and that they support the identity, quality, purity of the drug substances and drug products. (Gupta, 2015).

*Jasminum grandiflorum* commonly known as Royal Jasmine, is a highly fragrant flowering plant native to South Asia. The plant has been traditionally used for its therapeutic properties and its essential oil is extracted and used in perfumes, soaps, and other cosmetic products. In recent years, the photochemistry and therapeutic potential of *J. grandiflorum* has been extensively studied, revealing its wide range of applications in modern medicine.

In addition to its photochemistry properties, *J. grandiflorum* has also been found to have various therapeutic effects on the body. The plant has traditionally been used to treat a range of conditions, including anxiety, depression, and insomnia, and recent research has confirmed these traditional uses.

*Jasminum grandiflorum* Linn. Belonging to family Oleaceae is a well-known medicinal plant. It is an Ayurveda (herbal) medicinal plant. This review is to give comprehensive information on the chemical constituents and medicinal importance of *Jasminum grandiflorum*. It is commonly known as Chameli in Hindi. (Padmaa Paarakh, and P. M. Paarakh *et al*. 2009).

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### 1.1. Taxonomical classification

Kingdom	:	Plants
Subkingdom	:	Tracheobionts- Vascular plants
Division	:	Magnoliophyta- Flowering plants
Class	:	Magnoliopsida- Dicotyledons
Order	:	Scrophulariales
Family	:	Oleaceae- Olive family
Genus	:	Jasminum
Species	:	Grandiflorum

#### Vernacular names

- Hindi- Jati, Chameli
- Tamil- Jatimalli, Kotimalligai, Pitchi
- Sanskrit- Jati, Malati
- English- Spanish jasmine, common jasmine. (Bharathi, P. Rajasri, Shubashini K. Sripathi, and A. Naga Lakshmi *et al.*2020).

## 2. Materials and methods

The fresh and healthy leaves of *Jasminum grandiflorum* Linn were collected from various region of nagpur. The plant was identified and authenticated in Botany Department, RTMNU, Nagpur. The leaves were shade dried for 3-7 weeks until they were crisp, were size reduced and were used for further experimentation. The dried leaves were coarsely powdered and were defatted using Petroleum Ether (60-80 LR), with 50 cycle using Soxhlet apparatus 99% absolute ethanol and Distilled Water solvent used in selection of solvent system for maceration. The deciding ratio of solvent for maceration 1:1. Keep it for 7 days and Stir periodically 3-4 times / day. (Patel and Pawar *et al.*, 2012).

### 2.1. Standardization of total flavonoid content

#### 2.1.1. To study the effect of pH on $\lambda$ max

A few drops of NaOH were added to aliquots of the standard stock solution of rutin (2ml of 10 ug /ml) during the test. Two ml of rutin (10 u/ml) were placed in the second test tube, and a few drops of Hcl were also added. The absorbance of plain rutin, basic rutin, and acidic rutin was measured at various wavelengths. (Shraim and Amjad M. *et al.* 2021).

#### 2.1.2. To study the appropriate reagent in Aluminium chloride Method, for total flavonoid content determination

The first test tube added 4 ml Rutin (10 g/ml) ,1 ml of AlCl<sub>3</sub> (10%). Using a UV-VIS spectrophotometer with a liquid quartz cell as the blank methanol and AlCl<sub>3</sub> (10%) as the compensation. The second test tube obtained 4 ml of rutin (10 g/ml), added by 0.5 ml of NaNO<sub>2</sub> (5 M) was incubated for 2 minutes. A quartz cell NaNO<sub>2</sub> (5 M) and methanol used as the blank in a UV-vis spectrophotometer The third test tube added 4 ml of rutin (10 ug/ml), 0.1 ml of AlCl<sub>3</sub> (10%), and 0.1 ml of sodium acetate. After 15 minutes, the absorbance was measured at various wavelengths. Using a UV-VIS spectrophotometer with a liquid quartz cell as the blank methanol and sodium acetate as the compensation.

#### 2.1.3. To optimize reagent of time on the stability of rutin working standard solution

Test tubes were filled with aliquots of the standard stock solution of rutin (10ug/ml) taken 0.1 ml of, 0.1 ml of sodium acetate, and 0.1 ml of NaOH were added. A separate wavelength was used for recording absorbance immediately, after 15 minutes, and after 30 minutes. Calculate the percentage difference between the absorbance 0 min, after 15 minutes, and after 30 minutes.

### 2.2. Calibration curve of Rutin by Aluminium chloride Method

(Da Silva, Layzon Antonio Lemos, Bianca Ramos Pezzini, and Luciano Soares *et.al.*2015)

### 2.3. Preparation of Standard Stock Solutions (Rutin)

- **Stock Solution I** : 50 mg of rutin was dissolved in 50 ml of methanol, giving concentration of 1000 µg/ml.
- **Standard Solutions II** : 10 ml of stock solution I was dissolved in 100 ml of methanol, giving concentration of 100 µg/ml.
- **Working Standard Solutions** : 10 ml of rutin was dissolved in 50 ml of methanol, giving concentration of 20 µg/ml.
- From working standard solution 2, 4, 6, 8, 10 ml were diluted to 10 ml with methanol giving rutin concentrations of 4, 8, 12, 16, and 20 µg/ml respectively. 4 ml working standard Solution of rutin were separately taken in test tube with 0.5 ml of NaNO<sub>2</sub>, incubate for 2 minutes, and then add 0.1 ml of AlCl<sub>3</sub> after 15 minutes and four hours. The absorbance of the reaction mixtures was determined at 415 nm. Calculate the percentage difference between the four hours used for working standard solutions preparation.

#### 2.3.1. Calibration curve of quercetin by Aluminium chloride Method

- **Stock Solution I**: 10 mg of quercetin was dissolved in 100 ml of methanol, giving concentration of 100 µg/ml.
- **Stock solution II**: 12.5 ml of stock solution I was dissolved in 25 ml of methanol, giving concentration of 50 µg/ml.
- From working standard solution 1, 2, 3, 4, 5 ml were diluted to 10 ml with methanol giving quercetin concentrations of 5, 10, 15, 20 and 25 µg/ml respectively. 4 ml working standard Solution of quercetin were separately taken in test tube with 0.5 ml of NaNO<sub>2</sub>, incubate for 2 minutes, and then add 0.1 ml of AlCl<sub>3</sub> after 15 minutes and four hours. The absorbance of the reaction mixtures was determined at 415 nm. Calculate the percentage difference between the four hours used for working standard solutions preparation.

#### 2.3.2. Total Flavonoid content of hydroalcoholic extract of *Jasminum grandiflorum* equivalent to Rutin and Quercetin

10 mg of hydroalcoholic extract were accurately weighed and dissolved in 10 ml of methanol. The resulting solution was sonicated for 10 minutes and then filtered. From the filtrate, 1 ml of the solution was taken and diluted with methanol up to a total volume of 25 ml. To this diluted solution, 4 ml of the dilution was mixed with 0.5 ml of NaNO<sub>2</sub> and incubated for 2 minutes. After 15 minutes, 0.1 ml of AlCl<sub>3</sub> was added to the solution, and the mixture was left for four hours at a 415 nm wavelength. The results were expressed as mg of quercetin and rutin equivalent per gram of jasmine grandiflorum leaf.

### 2.4. Determination of total phenolic content

(Blainski, Andressa, Gisely Cristiny Lopes, and João Carlos Palazzo De Mello *et al.* 2013)

#### 2.4.1. Preparation of standard stock solutions (gallic acid)-

Gallic acid spectra - For the preparation of stock solution a 10mg gallic acid was dissolved in 10ml methanol.

Stock solution B was prepared by taking 1ml of stock solution A and diluting it to 10ml by methanol 1ml of the stock solution B was taken and diluted to 10ml with methanol to make standard solution C having the concentration 10µg/ml.

From standard solution C 2 ml solution was taken in which 1 ml of Folin-Ciocalteu reagent was added, and the volume was made up to 25ml by using 10.75% sodium carbonate. This solution was scanned in the range of 400 to 800 nm in UV spectrophotometer to obtain the spectra of the solution. Spectra was taken in the interval of 5, 15 and 30 minutes.

#### 2.4.2. To determine the total phenolic content (gallic acid Calibration curve)

For the preparation of stock solution a 10mg gallic acid was dissolved in 10ml methanol.

Stock solution B was prepared by taking 1ml of stock solution A and diluting it to 10ml by methanol 1ml of the stock solution B was taken and diluted to 10ml with methanol to make standard solution C having the concentration 10µg/ml.

From standard solution C 1,2,3,4,5 ml solution was pipette out and diluted to 10 ml with methanol respectively having the concentrations of 1,2,3,4,5 µg/ml. In each volumetric flask 1ml of Folin -Ciocalteu reagent was added, and the volume was made up to 25ml by using 10.75% sodium carbonate. The absorbance of each dilution was taken after 15 minutes and at 760 nm.

## 2.5. Total phenolic content of hydroalcoholic extract of *Jasminum grandiflorum* equivalent to gallic acid

For the preparation of stock solution a 10mg extract powder was dissolved in 10ml methanol.

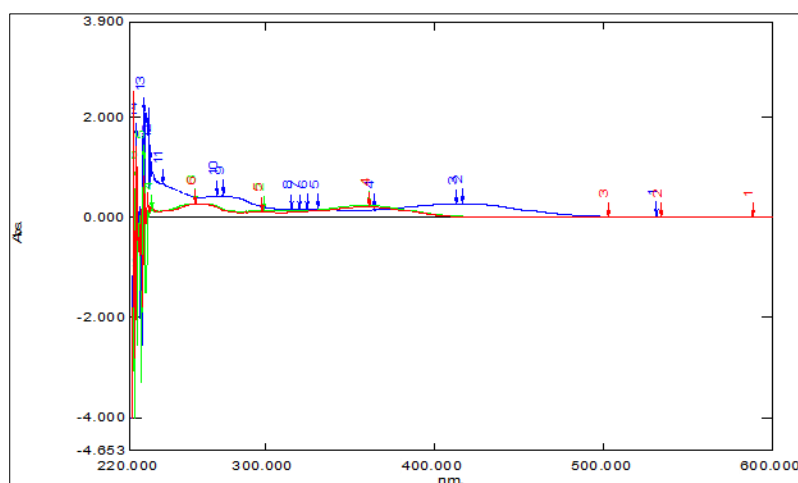
Stock solution B was prepared by taking 1ml of stock solution A and diluting it to 10ml by methanol 1ml of the stock solution B was taken and diluted to 10ml with methanol to make standard solution C having the concentration 10 $\mu$ g/ml.

From standard solution C 2 ml solution was taken in which 1 ml of Folin-Ciocalteu reagent was added, and the volume was made up to 25ml by using 10.75% sodium carbonate. The absorbance was measured at 760 nm.

## 3. Results and Discussion

### 3.1. Total flavonoid content

#### 3.1.1. To study the effect of pH on $\lambda$ max



**Figure 1** Spectra of pH on  $\lambda$  max

**Table 1** To determine the effect of pH on  $\lambda$  max

$\lambda$ max	Rutin	Rutin + NaOH	Rutin +HCl
259	0.262	0.436 (271)	0.286
361	0.210	0.160 (363)	0.237
415	-	0.268 (416)	-
510	-	0.016 (531)	-

$\lambda$  max 259nm of rutin it shift to 271nm at the basic pH and also intensity increase the basic pH from 0.262 for rutin in basic pH it is going to the 0.436.

It can observed that in basic pH have found additional maxima for the rutin. 415nm is acidic absorbance while it shows in neutral and acidic absorbance.

It was conclude that Rutin exhibits bathochromic shift for all of its maxima in its basic state. Furthermore, it was found that for each  $\lambda$  max, the intensity of absorption also increased. There was no change in  $\lambda$  max and their intensities while in an acidic environment.

## 3.1.2. To Optimize the stability of time on rutin working standard solution

**Table 2** The stability of time on rutin working standard solution

$\lambda$ max	0 min	15 min	30 min	$\lambda$ max	% difference between 0 min to 15 min	% difference between 15 to 30 min
268	1.536	1.306	0.0737	268	14.9%	85.06%
335	-0.322	-0.333	-0.269	335	-2.4%	16.4%
361	-1.1669	-1.135	-1.050	361	8.3%	10.0%
415	0.785	0.782	0.830	415	0.38%	-5.7%
510	0.465	0.479	0.522	510	-2.1%	-12.2%

It was conclude that absorbance was taken a 0 min, 15 min and 30min. It was found that, at 415 nm, the percentage difference between immediate to 15 minutes is less than immediate to 30 minutes. So we decide to take absorbance at 415 nm within 15 minutes time interval.

3.1.3. To study the appropriate reagent in  $AlCl_3$  method for Total flavonoid content determination**Table 3** To study the appropriate reagent in  $AlCl_3$  method

$\lambda$ max	Rutin + $NaNO_2$	Rutin + $AlCl_3$	Rutin + $AlCl_3$ + Sodium Acetate
259	1.536(268)	0.284(269)	0.135
361	-1.1669	0.131(363)	0.097
415	0.785	0.147	0.212
510	0.449	0.00	0.003

IT can observe that When Rutin and  $NaNO_2$  were combined and analyzed using spectrophotometry, the resulting spectra exhibited higher absorbance at a wavelength of 415 nm compared to the previously identified  $\lambda_{max}$  values of 259 nm, 361 nm, and 510 nm. However, when the spectra were obtained using a different solvent, the absorbance was reduced. Therefore, I chose to focus on the 415 nm wavelength since it displayed a higher level of absorbance in this particular condition. It was conclude that rutin absorbs greater at 415 wavelength when  $NaNO_2 + AlCl_3$  was present.

## 3.1.4. To determine the total flavanoid content (rutin Callibration curve)

**Table 4** Absorbance of different concentration of rutin

SN	Sample	Concentration( $\mu g$ /ml)	Absorbance (0 min)	Absorbance (After 4Hour)
1	Rutin	4	0.1619	0.1398
2	Rutin	8	0.223	0.2182
3	Rutin	12	0.3074	0.2659
4	Rutin	16	0.3561	0.3406
5	Rutin	20	0.425	0.4122

**Table 5** % Difference between immediate to 4 hour

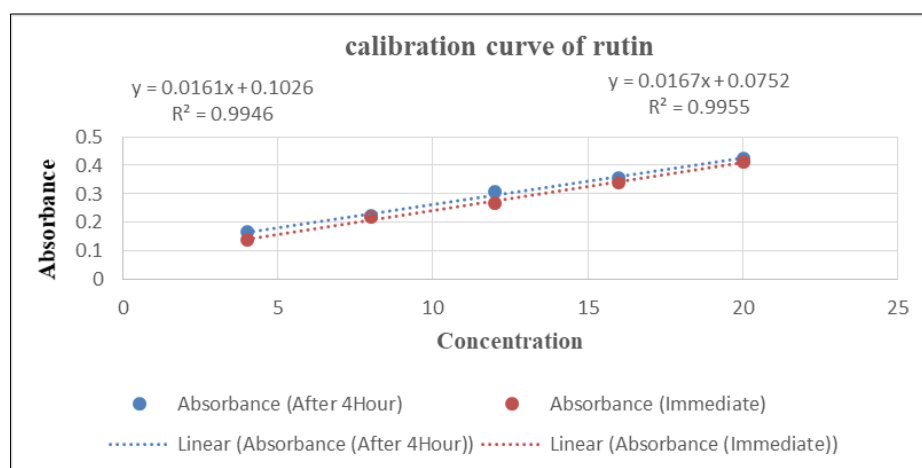
Concentration	%Difference between 0 min to 4 hours
4µg/ml	13.6 %
8µg/ml	5.13 %
12µg/ml	3.50 %
16µg/ml	4.35 %
20µg/ml	3.01%

$$R^2 \text{ value 0 min - } y = 0.0161x + 0.1026$$

$$R^2 = 0.9946$$

$$\text{After 4 hours- } y = 0.0167x + 0.0752$$

$$R^2 = 0.9955$$

**Figure 2** Calibration curve of rutin

The result of total flavonoid contents of the hydroalcoholic extract of *Jasminum grandiflorum* leaves. Equation of calibration curve of rutin standard immediate was  $y = 0.0161x + 0.1026$   $R^2 = 0.994$ . Equation of calibration curve of rutin standard after 4 hour was  $y = 0.0167x + 0.0752$   $R^2 = 0.9955$ . The total flavonoid contents crude extract determined by Aluminium chloride method was reported as rutin equivalents.

**Observation-** The stability of the rutin solution is not established. Because after preparing the solution, it should be seen as soon as possible because after four hours it won't be stable. It needs to be used immediately.

It was observed that due to a decrease in standard stock solution absorbance after 4 hours, we cannot use the same solution.

### 3.1.5. To determine the total flavonoid content (quercetin Calibration curve)

The result of total flavonoid contents of the hydroalcoholic extract of *Jasminum grandiflorum* leaves is given in table no 11. Equation of calibration curve of quercetin standard immediate was  $y = 0.299x + 0.1135$ ,  $R^2 = 0.992$ . Equation of calibration curve of quercetin standard after 4 hour was  $y = 0.295x + 0.1205$ ,  $R^2 = 0.991$ . The total flavonoid contents crude extract determined by Aluminium chloride method was reported as quercetin equivalents.

**Table 6** Absorbance of different concentration of rutin

SN	Sample	Concentration( $\mu\text{g/ml}$ )	Absorbance (0)	Absorbance (After 4Hour)
1	Quercetin	5	0.2743	0.279
2	Quercetin	10	0.3803	0.3847
3	Quercetin	15	0.5817	0.5791
4	Quercetin	20	0.7206	0.7319
5	Quercetin	25	0.851	0.8441

**Table 7** % Difference between 0 min to 4 hour

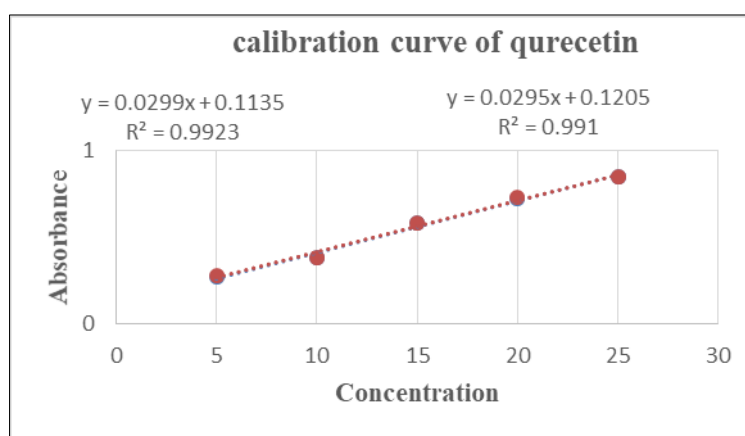
Concentration	%Difference between 0 min to 4 hours
5 $\mu\text{g/ml}$	-1.71 %
10 $\mu\text{g/ml}$	-1.15 %
15 $\mu\text{g/ml}$	0.44 %
20 $\mu\text{g/ml}$	-1.56 %
25 $\mu\text{g/ml}$	0.81 %

$$R^2 \text{ value-0 min} - y = 0.0299x + 0.1135$$

$$R^2 = 0.9923$$

$$\text{After 4 hours} - y = 0.0295x + 0.1205$$

$$R^2 = 0.991$$

**Figure 3** Calibration curve of quercetin

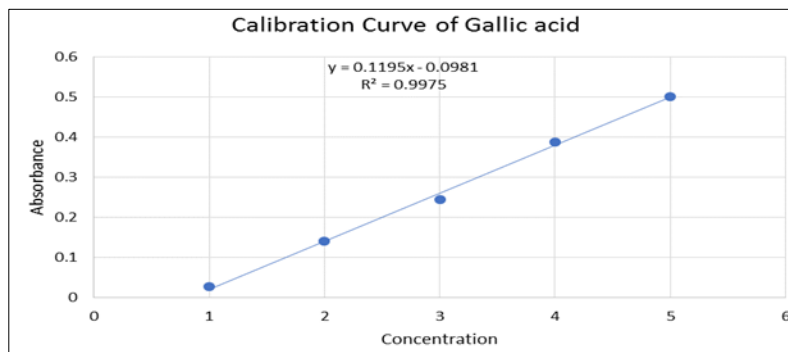
It can observe that it has been determined that the quercetin solution is stable. Because the solution should be stable after preparation. It must be used immediately and four hours later. It was observed that After 4 hours, there are very tiny differences in absorbance, and we can continue using the same standard stock solution.

### 3.1.6. Total flavonoid content of hydroalcoholic extract equivalent to Rutin and Quercetin

Total flavonoid content equivalent to Rutin of *Jasminum grandiflorum* was found to be 16.22 %.

Total flavonoid content equivalent to quercetin of *Jasminum grandiflorum* was found to be 8.075%.

### 3.2. Total phenolic content



**Figure 4** Calibration curve of gallic acid

#### 3.2.1. Total phenolic content of extract equivalent to gallic acid

Total phenolic content equivalent to gallic acid of *Jasminum grandiflorum* was found to be 18.8%.

## 4. Conclusion

The present investigation revealed that the leaves of *Jasminum grandiflorum* contain significant amount of phenols and flavonoids. The objective of this study was to get information of the amount of phenolics and flavonoids in different parts of *Jasminum grandiflorum*. Further intention of this study is to correlate relationship of these secondary metabolites to possible biological activities and evaluate *Jasminum grandiflorum* as a potential source of natural bioactive chemicals.

## Compliance with ethical standards

### Disclosure of conflict of interest

No conflict of interest to be disclosed.

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