FORMULATION AND EVALUATION OF ORAL HERBAL GRANULES FOR ASTHMA

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Abstract

Purpose: Asthma is a chronic disease of the bronchiwhich usually requires continuous medical care. According to WHO, about 235 million people currently suffer from asthma. It is the most common chronic disease prevalent among children. Approaches in formulating novel herbal drug formulations that are easy to administer especially for chronic diseases can form a valuable therapy in addition to overcoming the side effects and disadvantages of synthetic drugs.

Method: Vasaka is easily available and largely used herb for asthma. Oral granules of dried aqueous extract of *Justicia adhatoda (*Vasaka)(125mg per dose i.e. per gram) were formulated especially for paediatric patients using wet granulation technique, as they show noncompliance towards solid preparations such as tablets and capsules due to fear of chocking and syrups on account of their bitter taste. The bitter taste of Vasaka extract was masked with sucralose, flavouring agents and citric acid.

Results and conclusions: The formulation was prepared with excipients and flavouring gents to improve patient compliance. Citric acid which also acts as sialagogue was used, thereby obviating the need of consuming water while administering the formulation.The granules apart from showing excellent flow propertiesdisintegrate within 20 seconds in oral cavity. As wet granulation technique is used, the process is cheaper and less time consuming as compared to production of tablets, capsules and syrups. Hence, this formulation can serve as an ideal candidate for commercialization on large scale and an inexpensive therapy as compared to currently available formulations in the market.

Keywords: *Justicia adhatoda*. granules, Vasaka granules, asthma.

1. Introduction

Asthma attacks all age groups but often starts in childhood. Asthma is a chronic disease characterized by recurrent attacks of breathlessness and wheezing, which vary in severity and frequency from person to person. During an asthma attack, the lining of the bronchial tubes swells, causing the airways to narrow and reducing flow of air into and out of the lungs [1].The causes of asthma are not completely understood. However, risk factors for developing asthma include inhaling asthma "triggers", such as allergens, tobacco smoke and chemical irritants. Asthma cannot be cured, but appropriate management can control the disorder. Drugs commonly used for asthma include Bronchodilators, Leukotriene antagonists, Mast cell stabilizers, Corticosteroids, Anti-IgEantibody [2]. Recently Dr Reddy's Laboratories announced that it has launched Montelukast Sodium Oral Granules, a bioequivalent generic version of Singulair (montelukast sodium) oral granules, in the US market [3]. Disadvantages of synthetic medicines include development of tolerance, expensive, high toxicity, greater incidences of side effects, lower patient compliance due to factors such as bad mouth feel, throat irritation etc.

According to Ayurveda, the general etiology of svasaroga is that all things, materials and conditions that could help increase vatadosa and kaphadosa are causally responsible for tamakasvasa. This develops from an increase in cough (kasa), undigested materials (ama), diarrhea, vomiting (vamathu), poison (visa), anemia (pandu), and fever (jvara); coming into contact with air containing dust, irritant gases, pollens, or smoke; injuring vital spots; using very cold water; and residing in cold and damp places. Excessive use of dry food and astringent food and irregular dietary habits may also trigger an attack. In addition, constipation, excessive fasting, excessive use of cold water, excessive sexual indulgence in adults, exposure to extremes of temperature, anxieties, grief, disturbance of peace of mind, and debility may all precipitate an attack.Management of asthma in Ayurveda judiciously encompasses use of herbal and herbomineral drugs in addition to advising a healthy lifestyle [4].

Various medications are described in Ayurveda which include Krsnadichurna, Bharangyadichurna, Srangydichurna,decoction of *Dolichosbiflorus* (kulattha), *Zingiberofficinalis* (sunthi), *Solanumxantho- carpum* (kantakari), and*Justicia adhatoda* (vasaka) [5].

Vasaka (*Justicia adhatoda*) has been used widely as a prophylactic treatment of asthma in the form of decoctions, syrups, lozenges and tablets.Vasaka leaves contain quinazoline alkaloids such as vasicinone, vasicine,vasicinol,vasicol, vasicinine, adhatodineand 6- hydroxyl vasicine. Vasicine is oxidized to its ketonic derivative Vasicinone and the latter exerts main activity as bronchodilator [6].

Granules can be used as a novel approach for using herbal drug Vasaka for treatment of asthma as they offer advantages such as faster disintegration and dissolution as compared to tablets and capsules, palatable as compared to syrups and decoctions, greater acceptability in pediatrics and geriatrics due to lesser risk of choking, time and cost required for their manufacture is lesser as compared to tablets.

2. Materials and methods

2.1 Preparation of Vasaka extract

Vasaka leaves were collected and authenticated from Botanical Survey of India (BSI). Leaves were chopped and subjected to extraction using water as solvent as 60°C. Decoction was prepared by evaporating the extract to one third of its volume. Decoction was poured onto a glass tray and dried at 100°C. Dried extract was pulverized and stored in a desiccator.

2.2 Selection of excipients

Starch was chosen as disintegrant, calcium phosphate dibasic and Pearlitol as bulking agent, magnesium stearate as antiadherant, coloring agent to impart colour and methyl and propyl parabens as preservatives. To mask the extreme bitter taste of Vasaka, sucralose was used as sweetener, citric acid as taste masker and sialagogue and for flavoring strawberry flavor was used.The coloring and flavoring agents used were of food grade quality [7, 8].

2.3 Preparation of Granules

Formulation 1(Table 1)

Granules were prepared by using wet granulation technique. Vasaka extract (powder) and citric acid were mixed in a mortar to which sucralose and strawberry flavor were added. This was followed by subsequent addition of starch, Pearlitol, calcium phosphate dibasic and the parabens. Sufficient quantity of distilled water was added to form a lumpy mass which was then passed through sieve no. 22 to form granules. Granules were dried in the oven. Magnesium stearate was added at the end [9].

Table 1: Formulation 1 (F1)

Sr.no	Ingredients	Quantity (mg)	Category
1	Vasaka	125	Antiasthamatic
2	Starch	150	Disintegrant
3	Magnesium stearate	2.5	Antiadherant
4	Pearlitol	312.5	Bulking agent
5	Calcium phosphate dibasic	250	Bulking agent
6	Citric acid	125	Taste masker, Sialagogue
7	Methyl paraben	2	Preservative
8	Propyl paraben	0.5	Preservative
9	Strawberry flavor	qs	Flavoring agent
10	Sucralose	qs	Sweetening agent
11	Color	qs	Coloring agent

qs: quantity sufficient

Formulation 2 (Table 2)

Vasaka extract (powder) was taste masked with sucralose and added to a beaker containing molten chocolate. The mixture was stirred and allowed to partly cool down so as to form a lumpy mass which was then passed through sieve no.22 to form granules. Granules were kept in refrigerator for 15 minutes.

Table 2: formulation 2 (F2)

Sr.no	Ingredient	Quantity (mg)	Category
1	Vasaka	125	Antiasthamatic
2	Sucralose	qs	Sweetening agent
3	Chocolate	875	Taste masker, flavoring agent

qs: quantity sufficient

2.4 Evaluation of granules

2.4.1. Angle of Repose

The angle of repose is the angle formed by the horizontal base of the bench surface and the edge of a cone-like pile of granules. After the cone from 5 g of sample was built, height of the granules forming the cone (h) and the radius (r) of the base were measured. The angle of repose (θ) was calculated as follows:

$$\theta = \tan^{-1}(h/r)$$

Results were only considered valid when a symmetrical cone of powder was formed. The funnel method was used to perform the test [10].**(Table 3)**

Table 3: Results for angle of repose, bulk and tapped density, % LOD and disintegration time.

Formula	Angle of repose	Bulk Density	Tapped density	% of LOD	Disintegration time
F1	27.38°	0.377	0.467	1.24	Within 20 sec
F2	29.88°	0.348	0.488	-	Within 20 sec

LOD: Loss on Drying

2.4.2. Bulk density

It is the ratio of total mass of powder to the bulk volume of powder.

Db = m / Vo

Where, m: Mass of the blend

Vo: Untapped Volume

A graduated glass cylinder was used to perform the test [11]. **(Table 3)**

2.4.3. Tapped Density

Tapped density is the ratio of mass of powder to the tapped volume. Tapped volume is the volume occupied by the same mass of the powder after a standard tapping of a measure.

Dt= m / Vi

Where, m: Mass of the blend.

Vi: Tapped Volume

Graduated glass cylinder was used for the test which was subjected to 50 tappings and the volume was noted [11]. **(Table 3)**

2.4.4.Loss on drying

This test was performed by drying a weighed quantity of the product in the oven at 105°C until constant weight was obtained [12]. **(Table 3)**

2.4.5. Disintegration time

The test was performed using a beaker containing simulated saliva fluid maintained at 37°C for evaluating fast disintegration. The formulation was added to it and the disintegration time was noted. **(Table 3)**

2.4.6. Microbial limit test:

Total viable count of bacteria and fungi was evaluated by pour plate method as per standard procedures [13].

2.4.7. Total alkaloidal content

One millilitre of the extract containing known amount of vasicine in saline solution was shaken well with 20 µl of ammonia solution and then gently heated on a water bath. After cooling, vasicine was extracted usingchloroform (3×2.5 ml). The mixture was shaken well, centrifuged and to the separated chloroform extract a little anhydrous sodium sulphate was added to remove the traces of moisture. The extract was evaporated to

dryness under vacuum. Vasicine was reconstituted with 1 ml of saline and the absorbance measured at $\lambda_{max}$of 281 nm using an UV spectrophotometer. A stock solution of vasicine 10 µg/ml in saline was prepared using an ultrasonic bath. From this stock solution, a series of dilutions (1, 2, 3, 4, 5, 6, 7, 8, 9 and 10 µg/ml) were made using saline. Absorbance was measured using UV spectrophotometer at λ_{max}281 nm and the standard graph plotted with concentration (µg/ml) on the abscissa and absorbance on the ordinate [14].

2.4.8. HPTLC

HPTLC precoated, silica gel G 60 F25 (Merck, Germany) plates were used for application of sample. A small quantity of extract was dissolved in mobile phase and sample was applied on precoated plate with the help of Linomat IV applicator. Solvent system optimized for TLC study was chosen for HPTLC study.

Chromatographic conditions: Following are the chromatographic conditions required to get an effective resolutions by selected mobile phase.

Stationary phase: HPTLC precoated, silica gel G 60 F254 (Merck, Germany)

Size: 10 x 10 cm

Developing chamber: Twin trough glass chamber

Mode of application: Band

Band size: 5 mm

Separation technique: Ascending

Temperature: 20 ± 5OC

Saturation time: 30 min

Scanning wavelength: 254 nm / 366 nm

Scanning mode: Absorbance/Reflectance [15].

2.4.9. Qualitative phytochemical screening of extract of *Justicia adhatoda*

The tests for saponins, alkaloids, tannins, flavonoids, steroids, carbohydrates and cardiac glycosides were done as per standard procedures [16].

3. Results and discussion

3.1 Drug profile

Vasaka was chosen as the drug as it is easily available, perennial plant and the collection is easier. It also has a well-established pharmacological profile proving its antiasthamatic activity. Vasicine (quinazoline alkaloid) is mainly responsible for the bronchodilatory and mucolytic effects of the drug [6]. Extraction was carried out using water as the alkaloid is readily soluble in water. This formulation is mainly intended for pediatric and geriatric use hence, the dose of the drug is taken to be 125mg. The dose has been fixed in reference to commercially available dosage forms of Vasaka which recommends number of tablets or teaspoons of syrup that correspond to 125mg of the drug for pediatric use [17, 18].

3.2 Excipient profile

Starch takes up water from the body fluids which cause it to swell and thereby leading to disintegration of the granules. Calcium phosphate dibasic and Pearlitol were used as bulking agents. Pearlitol also aids in faster disintegration and acts as a non-calorific sweetening agent. Magnesium stearate helps to prevent attrition between the granules and formation of fines. Methyl and propyl parabens are nontoxic, non-irritating and are used in combination to prevent decomposition of the formulation. Citric acid helps to stimulate salivary secretions and hence leading to disintegration of the granules in the oral cavity, thereby obviating the need to consume water along with the formulation. Hence, it can be conveniently used by travelling patients. Sucralose and the flavoring agent help to mask the bitter taste of Vasaka. As they are triturated with the drug at the very beginning of the preparation before the addition of other excipients, they form a coating over the drug particles and hence in spite of disintegration within the oral cavity it makes the formulation highly palatable.

3.3 Angle of repose, bulk and tapped density, % LOD and disintegration time

The values of angle of repose are below 30°thereby indicating excellent flow properties. Lower values of bulk and tapped density indicate higher porosity implying the time required for disintegration would be lower. % LOD test values comply with the official limits and indicatelower moisture content in the formulation [19]. This test cannot be performed on F2 as the chocolate will melt at higher temperature and granules would be converted to a sticky mass. The disintegration test implies that the granules can disintegrate within 20 sec, thereby leading to quicker absorption and onset of action of the drug as compared to that in its other dosage forms such as tablets and lozenges.**(Table 3)**

3.4 Microbial limit test

No viable bacterial and fungal count was found. Thereby, indicating that the formulation is free from microbial contamination.

3.5 Total alkaloidal content:

The total alkaloid content was found to be 1.056% per gram of the formulation with reference to vasicine **(Figure 1 and 2)**. The alkaloids present in the plant are responsible for the bronchodilatory and mucolytic effects hence, evaluation of their content is crucial. The results prove that one gram of the formulation has sufficient amount of alkaloids to evoke a response.

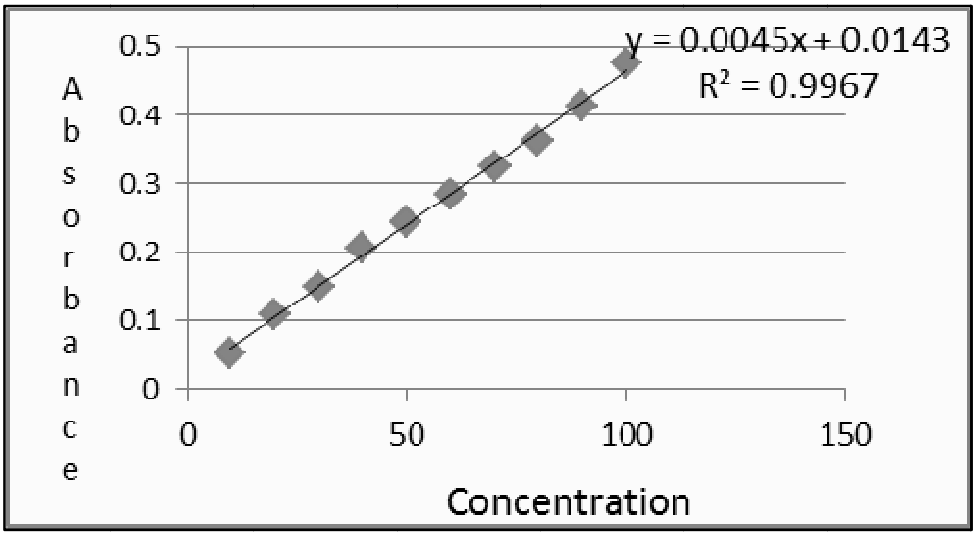


Figure 1: Calibration curve to estimate total alkaloidal content in the formulation

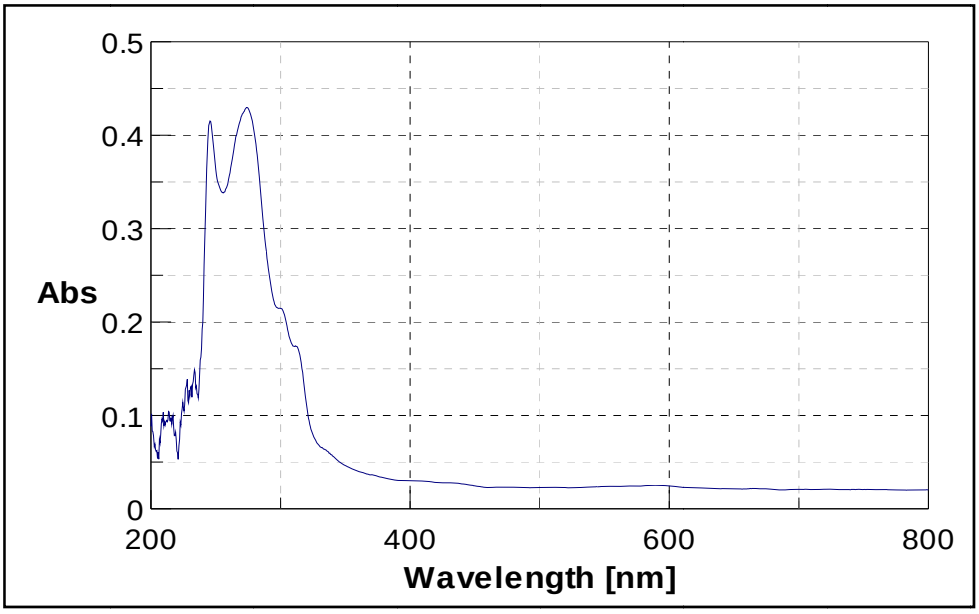


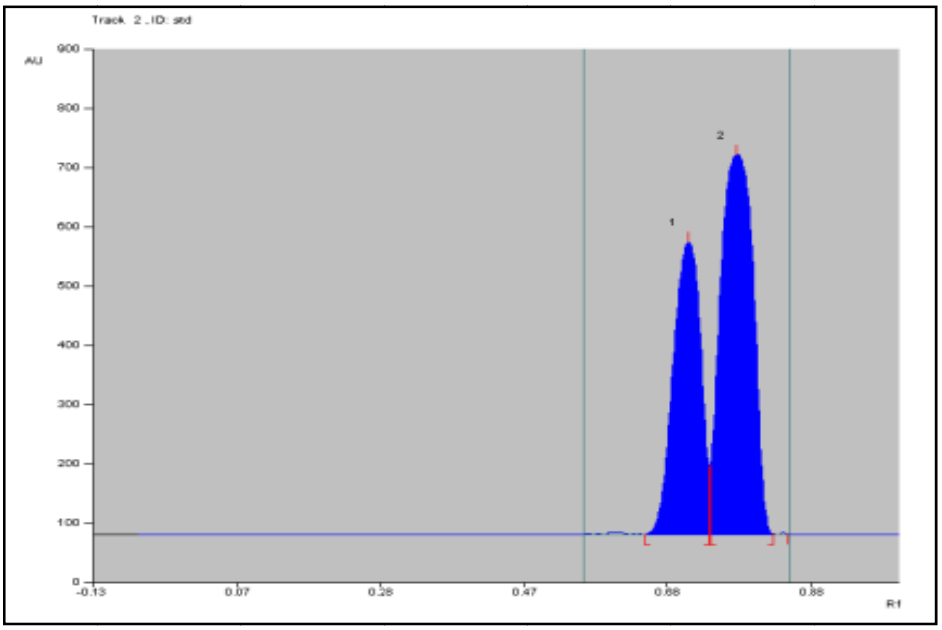
Figure 2: UV estimation of total alkaloidal content in the formulation.

3.6 HPTLC of Aqueous Extract of *Justicia adhatoda* (Vasaka)

The peaks appearing at 0.7 and 0.82 Rf values confirm the presence of vasicine and vasicinone respectively which are responsible for the bronchodilatory and mucolytic effects.(**Table 4 and Figure 3,4)**

Table 4: HPTLC results

Test extract	Solvent system	Rf values
Justicia adhatoda	Ethyl acetate: Methanol: Ammonia 8: 2 : 0.2	0.7 and 0.82



Rf values of 0.7 and 0.82 confirm presence of Vasicine and Vasicinone respectively.

Figure 3: HPTLC analysis to confirm the presence of Vasicine and Vasicinone.

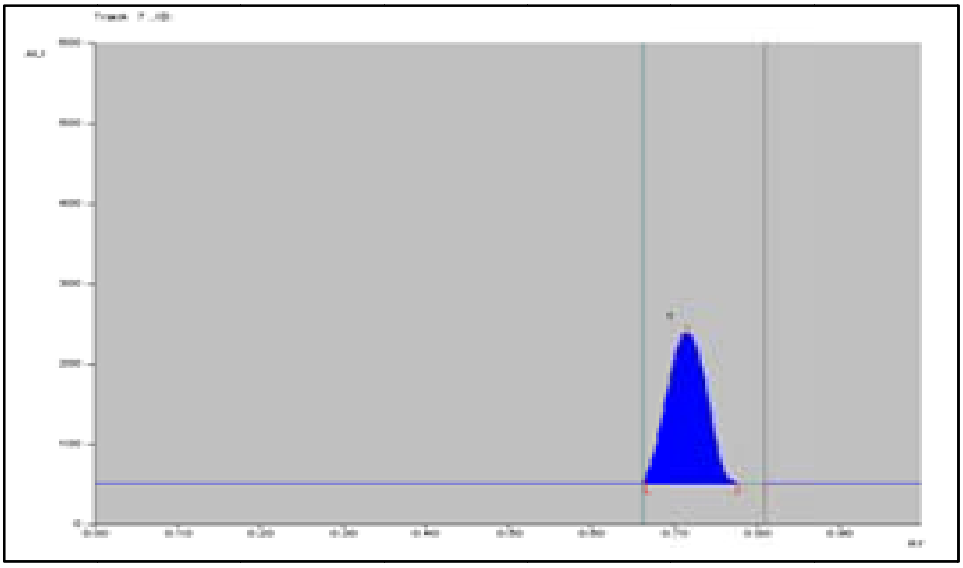


Figure 4: HPTLC analysis of standard Vasicine

3.7 Qualitative phytochemical examination of extract of Vasaka

Decoction was subjected to preliminary phytochemical screening for the evaluation of secondary metabolites.**Table 5** shows the result of the phytochemical screeningof leaf extract. The result shows the presence of saponins, alkaloids, tannins, flavonoids, steroids and carbohydrates.

Table 5: Preliminary phytochemical screening for the evaluation ofsecondary metabolites.

Sr. No	Name of test	Result
1	**Test for saponin glycosides**	
A	Foam Test	+
B	Hemolytic Test	+
2	**Test for Alkaloids**	
A	Dragendorff's test	+
B	Mayer's test	+
3	**Test for Tannins**	
A	Drug +5% $FeCl_3$ solution	+
B	Drug + Lead acetate solution	+
4	**Test for Flavonoids**	
A	Shinoda test	+
B	Drug + Lead acetate solution	+
5	**Test for Steroids**	
A	Salkowski test	+
B	Libermann-Burchad test	+
6	**Test for Carbohydrates**	
A	Molish's test	+
7	**Test for cardiac glycosides**	
A	Baljet's test	-

4. Conclusion

Herbal drugs like Vasaka have proven bronchodilatory activity. According to Ayurveda, the swarasa or juice of Vasaka leaves is administered for respiratory conditions. Many liquid oral formulations such as syrups and other formulations such as tablets, capsules and lozenges of Vasaka are available in market. The solid oral formulations are not readily accepted by pediatric and geriatric patents due to the fear of chocking whereas the variation in dose and sugar content in liquid formulations is of concern.Thus, herbal granules formulated from dried aqueous extract of Vasaka have good flow properties, greater palatability and disintegrate within 20 seconds in the oral cavity without the use of water. Hence, it can be widely accepted by pediatric and geriatric patients. It can also be an ideal choice for travelling patients as it obviates the need of water for its administration.As Vasaka is a single component in the formulation, the standardization is easier. The cost of the formulation is reduced as it is easy to manufacture this formulation on a large scale.

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Conflicts of interest

The authors confirm that this article content has no conflicts of interest.

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