ISSN 2063-5346 *Section A-Research paper*

FORMULATION AND EVALUATION OF POLYHERBAL FORMULATION CONTAINING INDIGENOUS MEDICINAL PLANTS

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Abstract

Background and objectives: The goal of the current study was to create and assess a polyherbal formulation that contained hydroalcoholic extracts of the entire plants of *Boerhavia diffusa, Eclipta prostrata, Phyllanthus amarus,* and *Solanum nigrum* in the same concentration.

Materials and Methods: Isopropyl alcohol was used in the wet granulation procedure to prepare the granules. To assess the pre-compression properties of powder mixes, preformulation research was conducted. Polyherbal tablets were prepared by wet granulation technique by using starch, talc, magnesium stearate, acacia and lactose as excipients and then precompression and post compression parameters were evaluated.

Results and Discussion: Preformulation studies were performed according to the standard procedure. Angle of repose, loose bulk density (LBD), tapped bulk density (TBD), compressibility index and loss on drying (LOD) showed satisfactory micrometric properties. Tablets are evaluated for color and appearance, weight variation, hardness test, friability test, pH of the tablet and disintegration test for tablets.

Conclusion: The observations from the formulation support the ideal properties of compressed tablets and their feasibility for large-scale commercial production. Finally, stability testing was performed on the produced tablets in order to determine their shelf life.

Keywords: Hdroalcoholic extracts, Preformulation studies, Polyherbal formulation, Accelerated stability studies, FTIR analysis.

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Introduction

Plants have a tremendous impact on disease prevention, disease treatment, and decreasing the side effects of conventional drugs. The creation of contemporary herbal dosage forms can help meet the pressing requirement for standardised and repeatable herbal remedies on the current world wide market. Although medicinal plants have been used for centuries to treat human illnesses, their use has significantly grown in recent years [1]. To obtain the benefits of synergism or to avoid adverse effects resulting from the principal herb, herbal formulations are typically made with combinations of individually extracted single herbs. Unit dose forms, which are safe, practical, easy to handle, and transportable, are advantages [2]. Almost 80% of people around the world still rely on traditional medicines to maintain their health, according to the WHO [3].

Boerhaavia diffusa, a member of the Nyctaginaceae family and also known as Punarnava in Ayurveda and "Hog weed" in English, is also called. According to Ayurveda, a rejuvenator is someone who helps us recover our youth by renewing our bodies. Over time, herbal therapy has altered and evolved. Similar to this, it has been utilised for a variety of illnesses in both raw and extract form. A number of phyto contituents flavonoids, alkaloids, glycosides, triterpenoids, lignans, have been reported from the herb. The medicinal, pharmaceutical, and antibacterial characteristics of the roots assist treat a wide range of illnesses, including diabetes, liver conditions, and renal issues [4, 5]. *Eclipta alba*, often referred to as Bhringraj and a member of the Asteraceae family, is a plant that may be found all throughout India. The plant contains the alkaloid ecliptine; additional compounds include wedelolactone, demethylwedelolactone, wedelic acid, apigenin, and luteolin. *Eclipta alba* were reported different pharmacological activities like Diuretic, hypotensive, hypocholesterolemic effect Antihyperglycemic activity and Analgesic studies [6]. A tiny herb from the phyllanthaceae family, *Phyllanthus amarus* is utilised all over the world for its medical benefits. It contain variety of phytoconstituents like lignans namely phyllanthin, hypophyllanthin, nirphyllin and phyllnirurin; flavanone glycosides like niranthin, nirtetralin, phyltetralin and lintetralin; a steroidal hormone estradiol; flavanoids like quercetin, quercitrin, and astragalin; triterpenes like phyllanthenol, phyllanthenone and phyllantheol are some of the important constituents. Antiviral, antibacterial, antiplasmodial, anti-inflammatory, antimalarial, antimicrobial, anticancer, antidiabetic, hypolipidemic, antioxidant, hepatoprotective, nephronprotective, and diurectic qualities were just a few of the pharmacological effects the plant displayed [7]. *Solanum nigrum*, which is a member of the solanaceae family. Alkaloids, flavonoids, tannins, saponins, glycosides, proteins, carbohydrates, coumarins, and phytosterols are said to be present in the entire plant. It treats gastrointestinal problems, fever, dysentery, and hepatitis. The plant's juice is used to skin conditions like ulcers. The fruits are used to cure asthma, as an appetite stimulant, and as a laxative [8].

Materials and Methods

Authentication and Collection of plant materials

Boerhavia diffusa, *Eclipta prostrata*, *Phyllanthus amarus*, and *Solanum nigrum* entire plants were collected in Paruthikulam and nearby villages in the Kanchipuram District of Tamil Nadu, India. These four plants were authenticated by Dr. M.U. Sharief, Scientist "E" and Director of the Southern Regional Center of the Botanical Survey of India, Coimbatore. The voucher specimen carries the designation BSI/SRC/5/23/2021/Tech.

Extraction of plant material

The collected whole plant materials were cleaned dried under shade, and coarsely powdered (40 mesh size) by a mechanical grinder. The coarsely powdered materials were macerated with ethanol and water (70:30v/v) for seven days with intermittent stirring and filtered after seven days and concentrated at an appropriate temperature (40 ^{o}C) on a rotary evaporator and dried [9].

Preformulation studies

Carr's, Hausner ratio, and the angle of repose were used to calculate the formulation's flowability [10, 11].

Angle of repose

The fixed funnel method was used to determine the angle of repose. The funnel's height was set such that the tip of the funnel barely touched the top of the mound of precisely weighted grains. Onto the surface, the granules were allowed to freely flow through the funnel. The powder cone's diameter was measured, and the angle of repose was computed using the formula below.

$\text{Tan } \Theta = h/r$

Loose bulk density

A graduated cylinder is used to measure the weight and volume of loose bulk density (LBD), which is calculated using a weighted quantity of granules.

LBD = Weight of powder/Volume of the packing

Tapped bulk density
Tapped bulk density was determined by placing a graduated cylinder containing a known mass of granules in the furnace. The cylinder was allowed to fall under its own weight on a hard surface from a height of 10 cm. After drying, the granules were again screened through sieve no. 18 to remove larger granules and stored in desiccators.

Compressibility index
The compressibility index of the blends was determined by the carrier's compressibility index.
Compressibility index (%) = (TBD-LBD) x 100/TBD

Loss on drying (LOD)
A shallow, glass-stoppered, dried weighing bottle was filled with 2 gm of granules. In the drying chamber, the materials were arranged uniformly. In order to determine a constant weight, the bottle's stopper was removed out and the contents were allowed to dry for a predetermined period of time.
Loss on drying = Initial weight – Final weight / Initial weight x 100

Formulation of polyherbal tablets
The Polyherbal tablet formulation contains dried hydroalcoholic extracts of *Boerhavia diffusa*, *Eclipta prostrata*, *Phyllanthus amarus*, and *Solanum nigrum*. Polyherbal tablets were prepared by wet granulation technique by using starch, talc, magnesium stearate, acacia and lactose as excipients. The tablet compression process uses a single-punch tablet machine (Figure 1). The weight of the tablet was adjusted to 500 mg and the dose adjusted to a 200 mg concentration [12].

Evaluation of polyherbal tablets
For the evaluation of polyherbal tablets, the post compression parameters mentioned below were used [13-15].

Colour and appearance
The color, odor, and texture of the compressed tablets were examined.

Weight variation
For weight variation analysis, the average weight and standard deviation (SD) of 20 tablets were chosen randomly and weighed individually.

Hardness test
A tablet must have a certain level of strength and resistance to friability. The hardness was tested for the tablets using a calibrated hardness tester (Monsanto hardness tester and Pfizer hardness tester).

Friability test
Using a calibrated Roche friabilator (4 minutes at 25 rpm), the tablets' friability was evaluated, and the weight loss percentage was calculated.

pH of the tablet
The pH of the tablet was tested by using a pH meter. Standardize the electrode using pH 7 buffer by changing to pH mode and doing precise adjustment using the calibration control knob. Then switch to standby mode for a change of solution. Rinse the electrodes with distilled water and then with the solution to be measured. Check the pH of the tablet, it was dissolved in 100 ml of distilled water.

Disintegration test for tablets
Six tablets were put inside the tube, which was then raised and lowered so that the entire up and down motion was repeated 28 to 32 times every minute. When there are no particles above the gauge, which easily pass through the mesh, the tablets are broken up (10 mesh screen). Distilled water was the disintegrating agent for the polyherbal tablet. The equipment was used and kept at a temperature of 37±2ºC.

Accelerated stability studies
The environmental factors of storage, temperature, light, air, and humidity can affect the stability study of tablets. The formulation conducted accelerated stability testing for six months.
As per ICH guidelines, accelerated stability studies were carried out. The tablets were kept at 40°C/75% RH 2°C/5% RH for up to six months [16, 17].

FT-IR analysis of polyherbal formulation
By scanning the sample on potassium bromide discs, an infrared spectrum was recorded. To confirm the presence of the triterpenoid group, the vibrational spectra of the standards of ursolic acid compound and formulation were scanned separately on each sample of the formulation and standard [18].

Total alkaloid contents
Take 5gm sample in a separating funnel and add 20mL 10% NaOH and shake. Add 20mL chloroform shake and separate chloroform layer and repeat this step for 2times.and combine the chloroform extract. Wash with 20mL water for 2times in the separating funnel each time discard the water layer and collect the chloroform layer. Combine the chloroform layer and add 20mL 1N HCL and shake. Collect the chloroform layer and

discard the acid layer Repeat the step 2times.Combine the chloroform layer and add 15mL ammonia and shake. The chloroform layer is collected evaporated to dryness and % weight is calculated.

Alkaloid = Weight of residue x 100
Weight of sample

Total flavonoid contents

Standard preparation

Take 10 mg Quercetin standard in 10mL standard flask. Make up to volume with methanol to get 1000ppm. 1, 2, 3, 4ppm were prepared by serial dilution. From this 0.5mL standard+1.5mL ethanol+0.1ml 10% Aluminium chloride+0.1mL sodium acetate +2.8mL distilled water.

Sample preparation

26.1 mg sample taken and add 25mL methanol sonicate for 20 minute. From this 2mL sample taken and add 1.5mL ethanol,0.1ml 10% Aluminium chloride,0.1mL sodium acetate .Then make up to 5 ml with distilled water. Incubate both STD and sample 30 minute at room temperature. Read the absorbance of both standard and sample at 415 nm [19].

Flavanoid % = Observed concentration x Purity of standard
Sample concentration

Results and Discussion

Table: 1 Preformulation study

S.no	Parameters	Observation
1	Angle Of repose	28.7 ± 1.53
2	Bulk Density	0.427 ± 0.003g/cc
3	Tapped Density	0.515 ± 0.005g/cc
4	Compressibility Index	17.15 ± 0.2 %
5	Loss On Drying	5.5 ± 0.05% w/w

Preformulation studies were performed on combined hydroalcoholic extracts, starch, talc, magnesium stearate, acacia and lactose as excipients according to the standard procedures. Precompression parameters Angle of repose, loose bulk density (LBD), tapped bulk density (TBD), compressibility index and loss on drying (LOD) showed good flow characteristics of the powders. The acceptable limit of good flow indicating low interparticle friction (Table 1).

Formulation of polyherbal tablet

Table 2: Working formula for formulation of PHT

Boerhavia diffusa	50
Eclipta prostrata	50
Phyllanthus amarus	50
Solanum nigrum	50
Starch	40
Talc	10
Magnesium stereate	10
Acacia	10
Lactose	230

Fig 1: PHT 500mg

Polyherbal tablet containing equal quantities of *Boerhavia diffusa, Eclipta prostrate, Phyllanthus amarus, and Solanum nigrum* whole plant hydroalcoholic extracts. The formula was applied

to make the polyherbal tablets. A rotary press punching machine was used to create the tablets, which were then submitted to post compression evaluation criteria including colour and appearance, weight variation, a hardness test, a friability test, a tablet's pH, and a disintegration test for tablets. (Table 2).

Table 3: Evaluation of polyherbal tablet formuation

S.no	Post compression parameters	Result
1	Color	Light brown
2	Odor	Characteristic
3	Shape	Circle
4	Texture	Smooth
5	Average weight	499 ± 0.71mg
6	Thickness	4.18 ± 0.03 mm
7	Hardness test	5.8 ± 0.15 kg/cm^2
8	Friability test	0.72 ± 0.37%
9	Disintegration time	4.31 ± 0.26 min
10	pH	6.3

After development, tablets were subjected to standard pharmacopeial procedures for assessing factors such physical appearance, uniformity of weight, thickness, hardness, friability, and disintegration time. (Table 3). All of the test product parameters have been established to comply with pharmacopeial standards. The multiple evaluation criteria and the outcomes were within the pharmacopoeia's limits. The average weight variation of tablets was 263mg. The acceptable weight variation range is 5%. Hence, the entire formulated tablet passed the weight variation test. The hardness values also had higher friability values. The hardness of tablets was in a range of 6.5 kg/cm^2 to 7 kg/cm^2, which showed appreciable hardness. The minimum and maximum friability of the formulation were obtained as 0.50% and 0.50%, respectively. The present friability was less than 1% in all formulations, ensuring that the tablets were mechanically stable. Disintegration time is one of the most important factors in the quality control of drugs, especially in the formulation of natural products. The time required to disintegrate the tablets was in the range of 3 to 5 minutes, and the range was within the pharmacopoeia limit.

Table 4: Accelerated stability studies of the developed tablets. (Storage condition: 40 ± 2ºC/75 ± 5% RH)

Time months	Physical appearance	Average weight (mg)	Hardness (cm^2)	Friability (%)	Disintegration time (min)
I	Light brown, circle shaped tablets with characteristic odor	500 ± 1.68	5.2 ± 1.28	0.83 ± 1.11	4.30 ± 0.83
III	Light brown, circle shaped tablets with characteristic odor	501 ± 1.21	5.4 ± 0.29	1.01 ± 0.65	4.36 ± 0.86
VI	Light brown, circle shaped tablets with characteristic odor	501 ± 0.25	5.4 ± 0.15	1.11 ± 1.24	5.30 ± 1.69

The environmental factors of storage include temperature, light, air, and humidity, as well as the elements of the package, can have an impact on the stability parameters of a medication dosage form. The formulation were subjected to accelerated stability for a period of 6 months at accelerated temperature conditions, room temperature 40 ± 2ºC/75 ± 5% RH. In accelerated temperature settings, uniformity of weight, hardness, friability, and disintegration time were all observed with no discernible change in the physical appearance (Table 4). The developed tablets are stable under the designated storage conditions, it was determined.

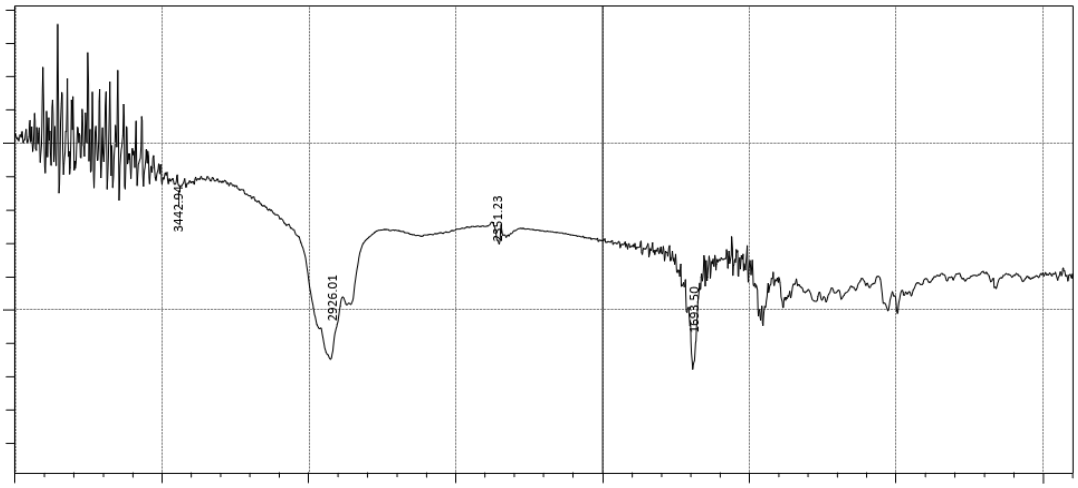

Figure 2: IR analysis of ursolic acid

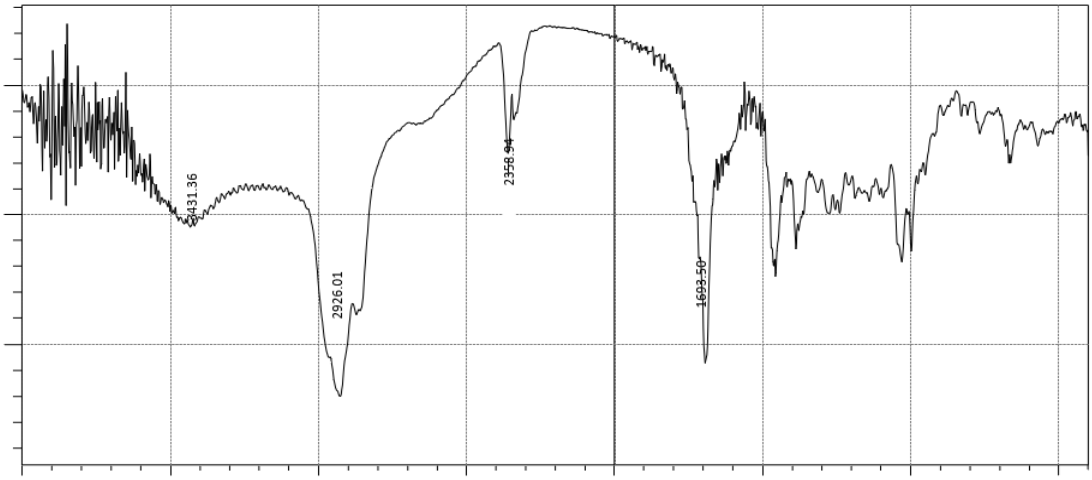

Figure 3: IR analysis of polyherbal formulation

FTIR studies

For comparison investigations, IR spectra of standard ursolic acid were used. Using the KBr disc method, the IR spectra of a polyherbal formulation were compared to standard IR spectra. The result of FTIR spectra of polyherbal formulation were shown that the presence of alcoholic group at 3431/cm, methylene group at 2926/cm & 2358/cm and 1963/cm for C=O. It was compared with standard ursolic acid and the reports suggested that presence of alcoholic group, methylene group and carboxylic acid. From the results, the polyherbal formulation may consist of ursolic acid (Figure 2&3).

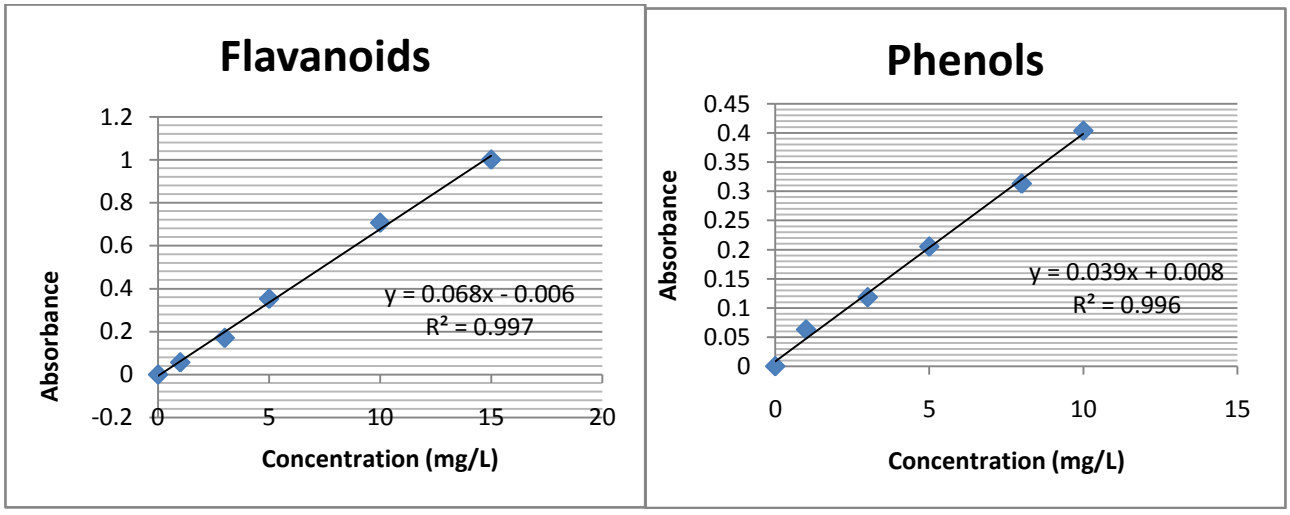

Figure 4: Linearity data for flavanoids and phenols

Table 5: Total alkaloids, flavanoids and phenolic content of polyherbal formulation

Parameter	Result
Alkaloids	4.63% w/w
Flavanoids	1.01% w/w
Phenols	3.68 % w/w

Phytochemical quantification

Polyherbal formulation contained maximum total alkaloid content (4.63%w/w), flavanoid content (1.01%w/w) and phenolic content (3.68%w/w). The hydroalcoholic extract of polyherbal formulation contained alkaloids, flavanoids and phenols were reported (Figure 4). Quantitative assessment of phytoconstituents also supports it. (Table 5).

Conclusion

The prepared formulation of polyherbal tablets on a laboratory scale may be employed as a stable solid dosage form, and the results of stability testing may aid in the development of shelf life estimation. The results of the current investigation showed that the stability characteristics were unaffected by the composition ratio of the constituents in polyherbal tablets. According to the findings of this study, it is possible to prepare medicinal plants in the form of affordable tablet formulations in order to increase their stability, consumer compliance, and acceptance. The traditional usage of *Boerhavia diffusa*, *Eclipta prostrata*, *Phyllanthus amarus*, and *Solanum nigrum* for treating liver problems is supported by all of these scientific findings. The newly prepared formulation is safer and more efficient.

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