Indian Journal of Traditional Knowledge
Vol 25(1), January 2026, pp 32-39
DOI: 10.56042/ijtk.v25i1.20700

Formulation and evaluation of polyherbal Asava to treat polycystic ovarian syndrome

Prathyusha Madireddy[a,b], Sheetu Wadhwa[b,*], Saurabh Singh[b] & Neelima Aligireddy[c]

[a]Department of Pharmaceutics, Sri Sai Jyothi College of Pharmacy, Hyderabad 500 075, Telangana, India
[b]School of Pharmaceutical Sciences, Lovely Professional University, Phagwara 144 411, Punjab, India
[c]Post Graduate Institute of Ayurveda, Dr. Sarvepally Radhakrishnan Rajasthan Ayurved University, Jodhpur 342 037, Rajasthan, India
[*]E-mail: sheetupharma@gmail.com, sheetu.21001@lpu.co.in

Received 02 July 2025; revised 06 November 2025; accepted 09 December 2025

The purpose of the study is to formulate and evaluate the polyherbal asava for the treatment of polycystic ovarian syndrome (PCOS). The asava was formulated by taking reference from the ancient literature to cure the disease which has a complex pathophysiology. Herbs such as *Salix caprea* L., *Syzygium cumini* (L.) Skeels., *Saraca asoca* (Roxb.) Willd., *Terminalia arjuna* (Roxb.) Wight and Arn, *Madhuca indica J.F. Gmel., Woodfordia fruticosa* (L.) Kurz, *Cinnamomum zeylanicum* Blume, *Santalum album* L., *Saccharum officinarum* L. (jaggery) were used in preparation of asava, using madhuca, woodfordia and jaggery as fermentating agents, and santalum and cinnamomum as additives. The physicochemical, phytochemical, quantification of markers by HPTLC were performed for the crude herbs. The physicochemical evaluations for the crude herbs were performed and complied with the standards. The phytochemical analysis of the crude drugs revealed the presence of various phytoconstituents. The quantification of markers in the crude drugs and optimized formulation by HPTLC was found as *S. caprea* (0.16, 0.86%), *S. cumini* (0.11, 0.60%), *S. asoca* (0.04, 0.29%), *T. arjuna* (0.008, 0.08%), *M. indica* (0.002, 0.02%). Five formulations, F1, F2, F3, F4, F5 were prepared and evaluated for organoleptic parameters, pH, viscosity, refractive index, alcohol content, sugar content, total solids, total acidity by standard methods. Amongst all the formulations F1 was considered optimum. Biostatistical analysis was performed by using one-way ANOVA and found that there is no significant difference between the evaluation parameters. Heavy metal analysis, microbiological analysis and accelerated stability studies of the optimized formulation were performed and found stable.

Keywords: Asava, Ayurveda, Fermentating agent, Herbal drug technology, Marker, *Madhuca indica*

IPC Code: Int Cl.[26]: A61K 36/00

In the context of the concept of polycystic ovarian syndrome (PCOS), still evolving with indeterminate etiology, complex pathophysiology, and multiple diagnostic criteria, symptom-based approach to its treatment is inadequate to manage PCOS effectively, and this calls for holistic/integrated approach for its treatment. Few plants were found to have multi-potential beneficial effects and show some optimism in evolving a standard polyherbal formulation for PCOS. The selected Ayurvedic drugs, when related to different herbal drug technologies indicate a great promise for effective treatment of PCOS[1].

PCOS is a complex endocrine disorder which leads to numerous health complications such as infertility, menstrual dysfunction, metabolic syndrome, acne, hirsutism, and obesity. Symptoms of PCOS are irregular, infrequent periods, light or very heavy bleeding during period of weight gain, excessive hair growth on face, chest and lower abdomen. If the symptoms remain unattended for long, it may lead to infertility in about less than 50% PCOS cases. This being the theoretical background, the critique of PCOS do indicate the inadequacies in explaining the causative factors, variations in estimation criteria, unclear treatment strategies and serious adverse effects of the current treatments and thus, the high magnitude of prevalence, unclear etiology and heterogeneous pathophysiology pose a great challenge[1-4].

The standard therapies for PCOS in modern medicine include oral contraceptives, progestins, androgens and ovulation induction agents. Allopathy uses antibiotics that have serious side effects and provides temporary cure while surgery is a costly process. Sometimes it may recur again after cessation

[*]Corresponding author

of the drugs. All over the World there is a growing importance of traditional systems of medicine in general and in treating PCOS in particular, because of their high potential to address the syndrome in a safe and effective manner. Ayurveda is one such alternative that can offer explanation to various aspects of PCOS[5-7].

Asava is a famous ancient medicinal preparations mentioned in the Vedas. They are self generated alcoholic medicaments prepared by making the powdered herbs or their decoctions to undergo fermentation when sugars are added. Presence of alcohol in the preparation had advantages, like better stability[8], enhanced therapeutic properties, increased efficiency of drug extraction from herbs and improvement in the drug delivery into the human body[9-11].

The pathognomonic feature of PCOS *i.e.,* cyst can be considered as one of the types of swellings called Granthi. It is due to the derangement of tridoshas. The selected drugs fall into important three vargas (Classification of plants for different medicinal properties) of Madhuka and Jambu in Amradiphala, Arjuna in Vatadiphala and Vetasa in Guduchyadi. The mechanism of action of these drugs is that they have the potential of balancing vata, pitta and kapha that can address PCOS. Literature study showed that herbs have diversified medicinal values. Each herb has some of its own unique medicinal values that have an effect on PCOS. Outcome of the combined drug effect is expected to be more encouraging in the management of PCOS[12-14].

Different formulations of asava were prepared by mixing dravya (SC- *Salix caprea* L., SZC- *Syzygium cumini* (L.) *Skeels.*, SA- *Saraca asoca* (*Roxb.*) *Willd.*, TA- *Terminalia arjuna* (*Roxb.*) *Wight and Arn*) and sandhan dravya (WF- *Woodfordia fruticosa* (L.) *kurz*, MI- *Madhuca indica J.F. Gmel.*) in appropriate ratios along with madhur dravya (SO- *Saccharum officinarum* L. or jaggery), prakshepa dravya (CZ- *Cinnamomum zeylanicum Blume*, SNA- *Santalum album* L.) and drava dravya (water) in the fermentation vessel. Then close the fermenter with clay smeared cloth. After fermentation, formulation is filtered and bottled[15]. SC is useful in bleeding disorders, balances Kapha & Pitta, SZC is used in treating menstrual blood loss, SA is having a stimulating effect on endometrial and ovarian tissue, TA is used as anti inflammatory and obesity all these effects combined helps to rule out PCOS in a safe and effective manner[16-18].

Materials and Methods

Herbal material

The barks of *Saraca asoca* (FBI ii 271, belongs to family Leguminosae, is a brown colour powder with characteristic taste and odour) and *Terminalia arjuna* (FBI ii 447, belongs to the family Combrataceae, is a reddish brown powder with characteristic odour and astringent taste), and seeds of *Syzygium cumini* (FBI ii 499, belongs to the family Myrtaceae is a light brown powder with aromatic odour and bitter taste) were collected by self, flowers of *Madhuca indica* (commonly known as honey tree belongs to family Sapotaceae is a dark brown powder with characteristic odour and sweet taste) and *Woodfordia fruticosa* (FBI ii, belongs to family Lythraceae bear cream and light pink colour flowers) were obtained from Swaraj Pansari, Punjab, and bark extract of *Salix caprea* [brown fine powder with characteristic odour and taste (batch number: VH/SCBEP/VHSCBEP89)] was obtained from Vital Herbs, New Delhi. The collected herbs were authenticated by Dr. MD. Mustafa, Department of Botany, Kakatiya University, Warangal. The samples were evaluated morphologically and microscopically as per the World Health Organization (WHO) guidelines for the standardization of herbal drugs[19].

Physicochemical investigation

As per the standard procedures of the Ayurvedic Pharmacopoeia of India (API) various quantitative tests of the crude drugs were performed. They are tested for the Ash value, Acid insoluble ash, ethanol soluble extractive, water soluble extractive[20-22].

Phytochemical investigation

The extracts of the crude drugs were subjected to various qualitative tests such as test for carbohydrates, amino acids, proteins, alkaloids, flavonoids, tannins, steroids or terpenoids, phenols and glycosides to determine the presence of various phytochemical constituents using standard procedures[23,24].

Quantification of markers

The quantification of markers in the extracts was performed at Vasu Research Center, Gujarat by using High Performance thin Layer Chromatography (HPTLC), using CAMAG Linomat-5 applicator using

Merck – Thin Layer Chromatography (TLC)/ HPTLC Silica gel 60 F254 aluminium sheets considered as stationary phase. The markers used for quantification of herbs and optimized formulation were catechin for SC, gallic acid for SZC, epicatechin for SA, arjunolic acid for TA and quercetin for MI. The development of the plates was performed in CAMAG TLC Twin trough chamber containing mobile phase. The mobile phase has of Toluene: Ethyl acetate: Formic acid: Methanol (6:6:1.6:0.4, v/v) (for SC and SA); Toluene: Ethyl acetate: Formic acid (10:7:1, 5:4:1 v/v) (for SZC, TA); Chloroform: Methanol (8.5:1.5, v/v) (for TA). Densitometric HPTLC quantification was performed at 278 nm (SC, SZC, SA); 540 nm (TA); 254 nm (MI) respectivlely[25,26].

Formulation of asava- sandhana kalpana

Five different formulations were prepared namely F1, F2, F3, F4, F5 by adding coarsely powdered herbs like SC, SZC, SA, TA mixed along with additives like CZ and SNA in the porcelain container. Then fermentating agents like MI, SO, WF were used to make the formulations. All these ingredients were mixed well with the help of distilled water. The container was covered with a lid and the edges were sealed with clay smeared cloth (muslin cloth) wound in seven consecutive layers. A constant temperature is maintained for fermentation by keeping the container in a heap of paddy. After 4 weeks the lid was opened and the contents were observed to ascertain whether fermentation has been completed. The fluid was decanted and then strained after two or three days after the fine suspended particles settle down, it is strained and bottled[27].

Evaluation of asava

pH

pH was measured by immersing the digital pH meter (SKADIOO, India) with a glass electrode in the standard solution for calibration by pressing the on button, stirred gently and waited for 30 seconds till the readings gets stabilized and then the same is repeated for the test solution and the readings were noted[28].

Refractive index

The Abbes' Refractometer was used to measure refractive index. The liquid sample (2 to 3 drops) is sandwiched into a thin layer between an illuminating prism and a refracting prism which is kept in front of the light source. Focus the eyepiece on the cross section of the instrument and rotate the index arm until a coloured band is seemed through the telescope. 3 readings noted for the sample[28].

Viscosity

It was measured by using Ostwald viscometer also called as U-tube viscometer, where the liquid with known density example water was sucked through the wider limb and time required for water to flow from point A to B between the bulbs was calculated. Same procedure was repeated by taking the sample. Density of sample is calculated by using pycnometer and the readings obtained were substituted in the below equation.

$$\mu L/ \mu 2 = t1\rho 1/t2\ \rho 2,$$

where is μ1, μ2 is viscosity of sample and water, t1, t2 is time of flow of sample and water and ρ1, ρ2 are density of sample and water[29].

Sugar content

It was calculated by using digital refractometer the brix percentage is equal to the sugar content in the sample. Where few drops of sample is placed on the prism and closed and aim front end of the refractometer to the direction of light. The reading was taken observing the blue and white lines cross the graduated scale. The scale will provide a direct reading of the concentrations[29].

Total solids

10 mL of sample was taken into a china dish which was previously weighed and allowed to evaporate in the hot air oven so that only solid content remains in the dish and rest gets evaporated. Then weigh the final weight of the container which gives the solid content of formulation is calculated[29].

Total acidity

Total acidity, also called titratable acidity for the formulation was determined by titrating known amount of sample with standard solution of NaOH solution, using phenolphthalein as an indicator[29].

Alcohol content

It is determined using distillation method. 25 mL of sample was transferred to the distillation flask and its temperature is noted. It was diluted with equal volume of water. Afterwards it distilled and distillate about 2 mL less than the total volume collected. Water was added to measure exactly same volume of original test liquid and adjusted to temperature noted before. Specific gravity of this liquid was determined and

alcohol content analyzed using relative density table given in United States Pharmacopoeia[29].

Heavy metal analysis

It was performed for the presence of heavy metals like Lead, Cadmium, Arsenic and Mercury according to the Ayurvedic Pharmacopoeia of India. The standard solutions and the sample solutions of different concentrations were prepared according to the standard procedure and a calibration curve was plotted using inductively coupled plasma-mass spectroscopy (ICP-MS) (model: NexION 2000, make: Perkin Elmer) to detect metals[29].

Microbiological testing

The test was performed for the presence of different viable microorganisms like yeast, fungi and bacteria according to the API. Pour plate method was used to determine total microbial plate count by using soya bean casein digest agar medium with 1ml of sample after incubation of 3-5 days at temperature 20-25ºC, colonies were counted by the formula,

Colony forming units (cfu) =
no. of colonies*dilution/weight of sample.

In the same way the tests are also performed for the presence of different microorganisms like yeast, moulds and bacteria in the sample by placing it on different agar mediums and observed for the growth of the microorganisms[29].

Accelerated stability studies

Factors like temperature, light, air and humidity affect the storage conditions of crude drugs and finished dosage forms. The optimized formulation was subjected to the test for 6 months as per the International Conference on Harmonization (ICH) guidelines at 40±2℃/75±5% RH. The formulation is tested for pH, refractive index and alcohol content at 0, 3, 6 months intervals[30].

Results and Discussion

The study aimed to formulate and evaluate polyherbal asava to treat polycystic ovarian syndrome. 5 different formulations were prepared using different herbs such as SC, SZC, MI, SA, TA and WF in different ratios. The organoleptic evaluation of herbs and the formulations, physicochemical investigations, phytochemical investigations, quantification of markers, evaluation of formulation were performed and reported in the following tables and figures. The visual representation of the work is depicted as graphical abstract shown in (Supplementary Fig. S1) and submitted as supplementary file.

Organoleptic evaluation

The organoleptic properties of herbs and formulations were carried out using visual examinations and showed that there is no foreign organic matter or no contamination. The results of Organoleptic Evaluation are reported in Table 1.

Physicochemical investigation

These studies were the part of preformulation study which was performed according to the Ayurvedic Pharmacopoeia of India (API). The parameters like total ash value, acid insoluble ash value, alcohol soluble extractive value, water soluble extractive value were carried out for all the herbs and the results obtained were within the limits. The results of physicochemical tests are reported in Table 2.

Phytochemical investigation

The phytochemical investigations of all the herbs were carried out and there was presence of different phytochemicals such as carbohydrates, alkaloids, flavonoids, tannins, steroids, phenols and glycosides. Flavonoids, tannins, phenols are present in SC, carbohydrates, alkaloids, flavonoids, tannins, phenols, glycosides were found to be present in both SA and MI, carbohydrates, alkaloids, tannins, steroids, phenols, glycosides in SZC, carbohydrates, alkaloids, flavonoids, tannins, steroids, phenols in TA were observed. The results of phytochemical tests are reported in Table 3.

Quantification of markers

The test was performed by using HPTLC technique and the amount of catechin in SC was found to be 0.16% in raw material and 0.86% in the fermented formulation, gallic acid is 0.11% in raw SZC and

Table 1 — Organoleptic evaluation of herbs and formulations

Organoleptic Property	M*I*	*SA*	*TA*	*SZC*	*SC*	*Formulation* F1 to F5
Colour	Dark Brown	Brown	Reddish Brown	Light Brown	Brown Fine Powder	Dark Brown
Odour	Characteristic	Characteristic	Characteristic	Aromatic	Characteristic	Alcoholic
Taste	Sweet	Characteristic	Astringent	Bitter	Characteristic	Sweet

0.60% in the formulation, SA has 0.04% of epicatechin in raw material and 0.29% in the formulation, TA has 0.008% of arjunolic acid in the raw herb and 0.08% in the formulation in the same way quercetin is MI is 0.002% in raw material and 0.02 in the formulation. By the above findings it is found that the herbs in the fermented formulation have more marker constituents than compared to the raw or crude form of the herbs. The results of HPTLC analysis are shown in Table 4 and (Fig. 1 & Fig. 2).

Table 2 — Physicochemical investigation of MI, SZC, TA, SA; n=3

Parameters (a)	Observed mean values (%)	Reference values (%)
Madhuca indica (*MI*)		
Total ash value	2.92±0.90	NMT 4.0
Acid insoluble ash value	1.16±0.2	NMT 1.2
Alcohol soluble extractive value	42.4±0.05	NLT 42.0
Water soluble extractive value	75.2±0.1	NLT 74.0
Syzygium cumini (*SZC*)		
Total ash value	0.9±0.1	NMT 1.9
Acid insoluble ash value	0.28±0.05	NMT 0.4
Alcohol soluble extractive value	6.4±0.2	NLT 5.7
Water soluble extractive value	18.4±0.90	NLT 14.5
Terminalia arjuna (*TA*)		
Total ash value	25.24±0.85	NMT 27.0
Acid insoluble ash value	1.38±0.11	NMT 2.0
Alcohol soluble extractive value	16.8±0.1	NLT 16.0
Water soluble extractive value	20±0.2	NLT 17.0
Saraca asoca (*SA*)		
Total ash value	11.75±1.58	NMT 13
Acid insoluble ash value	0.5±0.05	NMT 0.6
Alcohol soluble extractive value	12±0.1	NLT 11.0
Water soluble extractive value	16±0.05	NLT 10.5

Evaluation of asava

Different formulations of asava are prepared as per Table 5a and for the obtained fermented asava formulation evaluations like pH, refractive index,

Table 3 — Phytochemical investigation of herbs

S. No.	Parameters	SC	MI	SZC	TA	SA
1	Carbohydrates	Nil	+	+	+	+
2	Amino acids	Nil	Nil	Nil	Nil	Nil
3	Proteins	Nil	Nil	Nil	Nil	Nil
4	Alkaloids	Nil	+	+	+	+
5	Flavonoids	+	+	Nil	+	+
6	Tannins	+	+	+	+	+
7	Steroids and Terpenoids	Nil	Nil	+	+	+
8	Phenols	+	+	+	+	+
9	Glycosides	Nil	+	+	Nil	+

Table 4 — Quantification of markers in herbs and optimized formulation, F1

S. No	Crude Herb	Marker Component Analyzed	% of Marker present	
			Herb	Asava
1	SC	Catechin	0.16	0.86
2	SZC	Gallic acid	0.11	0.6
3	SA	Epicatechin	0.04	0.29
4	TA	Arjunolic acid	0.008	0.08
5	MI	Quercetin	0.002	0.02

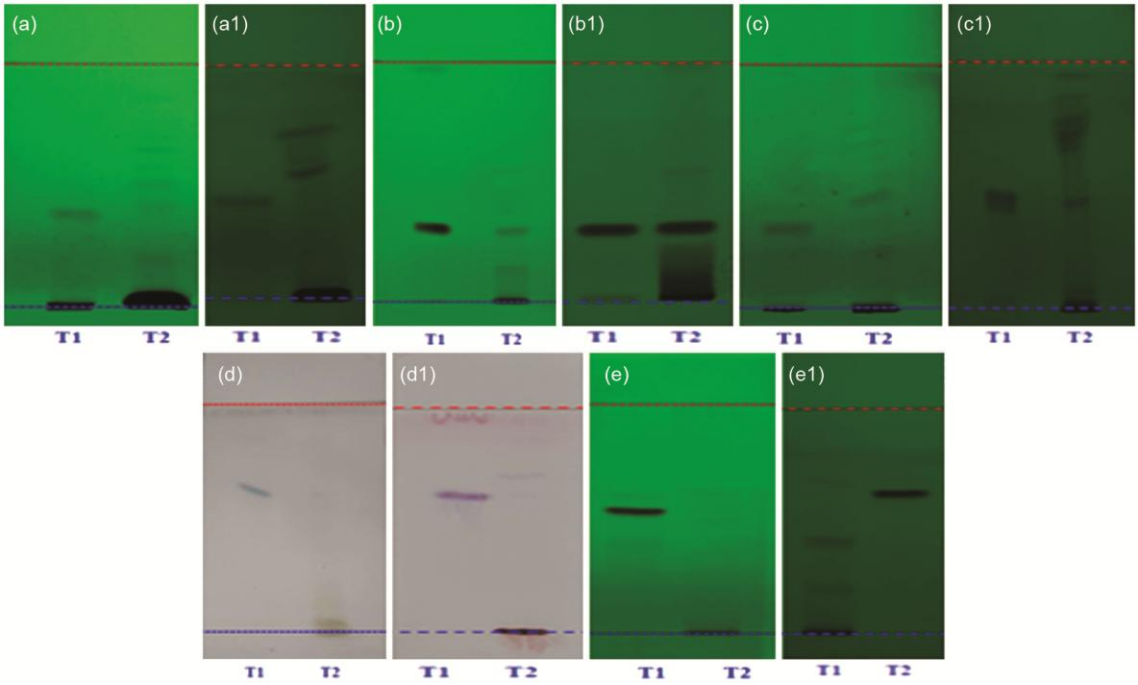

Fig. 1 — HPTLC plates of a-SC, b-SZC, c-SA, d-TA, e-MI in the herbs; a1-SC, b1-SZC, c1-SA, d1-TA, e1-MI in the optimized formulation, F1

viscosity, Sugar content, total solids, total acidity, and alcohol content, shown in Table 5b. The pH and total acidity values indicated that the asava is weakly acidic in nature. The values of refractive index, viscosity, sugar content and total solids suggest that the solution is less dense, and values were found to be within the standard limits. Alcohol content was found to be 6.01% which suggest that the self generated alcohol during the fermentation process itself acts as preservative making the formulation stable for a longer period of time. Biostatistical analysis done using One-Way ANOVA and p- value found to be >0.05 which implies that it fails to reject null hypothesis.

Table 5a — Different formulations of asava

Ingredients	F1 (g)	F2 (g)	F3 (g)	F4 (g)	F5 (g)
SA	200	200	200	200	200
SZC	100	100	100	100	100
TA	200	200	200	200	200
SC	100	100	100	100	100
MI	100	-	100	200	-
WF	-	100	100	-	200
CZ	100	100	100	100	100
SNA	100	100	100	100	100
SO	100	100	100	100	100
Distilled Water (ml)	1000	1000	1000	1000	1000

Heavy metal analysis

These studies were performed for the optimized formulation to detect the presence of lead, cadmium, arsenic and mercury which were below the mentioned limits of API. The formulation thus prepared is safe to consume without any heavy metal poisoning. The results of heavy metal tests are shown in Table 6

Table 5b — Evaluation parameters of asava

S. No	Parameter	F1	F2	F3	F4	F5
1	pH	4.00 ±0.01	4.01±0.01	3.9±0.01	4.0±0.01	3.9±0.02
2	Refractive index	1.35±0.003	1.35±0.002	1.3±0.001	1.2±0.001	1.3±0.002
3	Viscosity (Pa.s)	1.66±0.04	1.85±0.02	0.99±0.02	1.00±0.04	1.00±0.02
4	Sugar content (g/L)	14.93±0.11	12.86±0.05	17.00±0.1	18.00±0.03	18.00±0.04
5	Total solids (g)	23.61±0.11	21.65±0.13	25.00±0.20	24.00±0.10	24.00±0.10
6	Total acidity	1.84±0.14	1.24±0.06	2.00±0.10	1.90±0.2	1.90±0.20
7	Alcohol content (%)	6.01±0.20	2.53±0.20	2.0±0.10	5.0±0.02	3.00±0.10

Values expressed as Mean ± SD (n=3), biostatistical analysis (one-way ANOVA) found p>0.05, which states that the test fails to reject null hypothesis. (p value was found to be 0.999992).

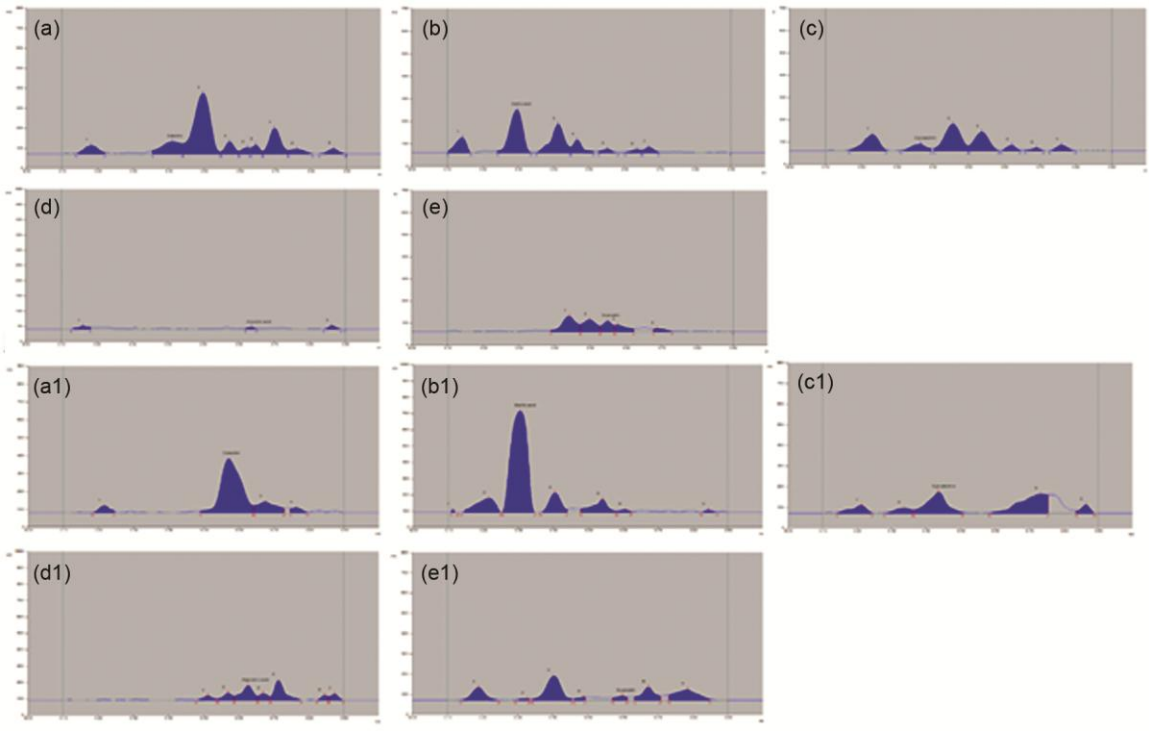

Fig. 2 — Chromatograms of a-SC, b-SZC, c-SA, d-TA, e-MI in the herbs; a1-SC, b1-SZC, c1-SA, d1-TA, e1-MI in the optimized formulation, F1

Table 6 — Heavy metal analysis of optimized formulation, F1

S. No.	Parameters	Result	Limit as per API
1	Lead	0.015 ppm	NMT 10 ppm
2	Cadmium	0.001 ppm	NMT 0.3 ppm
3	Arsenic	0.008 ppm	NMT 3 ppm
4	Mercury	0.212 ppm	NMT 1 ppm

Table 7 — Microbiological analysis of optimized formulation, F1

S. No.	Parameters	Result	Limit as per API
1	Total Microbial Plate Count (TPC)	< 10 cfu/g	10^5 cfu/g
2	Total Yeast & Mould Count (TYMC)	Nil	10^3 cfu/g
3	*Staphylococcus aureus*	Nil	Nil
4	*Salmonella* sp.	Nil	Nil
5	*Pseudomonas aeruginosa*	Nil	Nil
6	*Escherichia coli*	Nil	Nil

Table 8 — Accelerated stability studies of optimized formulation, F1

Sl. No	Parameter	Accelerated stability studies ($40\ ^0C \pm 2^0C$, 75% RH ±5%) Months		
		0	3	6
1	pH	4.01	3.99	4
2	Refractive index	1.35	1.36	1.35
3	Alcohol content (%)	6.01	6.01	6.01

Microbiological analysis

It is done for the optimized formulation to know the presence of any microbes generated during the fermentation process which will degrade the stability of the formulation. The test results concluded that no such microbial contamination was observed, and the formulation is free from contaminating yeast and bacteria. The results of microbiological analysis are shown in Table 7.

Accelerated stability studies

Optimized formulation was taken, and the stability studies were conducted which revealed that the asava formulation was stable throughout the timeline in both physical parameters like pH and refractive index and chemical parameter like alcohol content. The stability studies were conducted for 0 month, 3 months and 6 months and the results (Table 8) shown that observed values are within the acceptance range.

Conclusion

Critical evaluation of developed formulation through ayurvedic principles will help to explore innovative applications of technologies to develop better standardized and more clinically safe and effective dosage forms. Here we had made an interdisciplinary approach where traditional knowledge and modern technologies merged to formulate the asava to treat PCOS effectively with better stability and compliance. From the experimental findings it can be said that polyherbal formulations hold an important place and give promising insights in treating PCOS.

Supplementary Data

Supplementary data associated with this article is available in the electronic form at https://nopr.niscpr.res.in/jinfo/ijtk/IJTK_25(1)(2026)32-39_SupplData.pdf

Acknowledgements

The authors would like to acknowledge Chaitanya deemed to be University and Kakatiya University, Warangal, Sri Sai Jyothi College of Pharmacy, Hyderabad, Lovely Professional University, Punjab, Vasu Research Center, Vadodara, Vital Herbs, New Delhi, Swaraj Pansari, Punjab for their support and providing facilities throughout the research.

Conflict of Interest

The authors have no conflict of interest.

Author Contributions

PM, SW, SS and NA conceptualized and designed the experiments. SW, SS, NA provided the materials, supervised, reviewed and edited the manuscript. PM collected materials, performed the experiments, analysed and wrote the paper.

Ethics Approval

Not applicable

Informed Consent

Not applicable

Data Availability

Data supporting this study findings are included within the manuscript and will be made available by the authors upon reasonable request.

References

1 Ganie M A, Vasudevan V, Wani I A, Baba M S, Arif T, *et al*., Epidemiology, pathogenesis, genetics & management of polycystic ovary syndrome in India, *Indian J Med Res*, 150 (4) (2019) 333-344. DOI:10.4103/ijmr.IJMR_1937_17.

2 Teede H J, Misso M L, Costello M F, Dokras A, Laven J, *et al*., Recommendations from the international evidence-based guideline for the assessment and management of polycystic ovary syndrome, *Hum Reprod*, 33 (9) (2018) 1602-1618. DOI:10.1093/humrep/dey256.

3 Sreedharan A, Kale P, Anu M S & Dei L P, Ayurvedic management of primary infertility due to polycystic ovarian syndrome associated with multiple uterine fibroids: A case report, *Int J Ayurveda Pharm Res*, 6 (1) (2018) 19-25.

4 Siriwardene S A D, Karunathilaka L P A, Kodituwakku N D & Karunaratne Y A U D, Clinical efficacy of Ayurveda treatment regimen on subfertility with PCOS, *AYU*, 31 (1) (2010) 24-27. DOI: 10.4103/0974-8520.68203.

5 Chandran U, Mehendale N, Tillu G & Patwardhan B, Network pharmacology of Ayurveda formulation Triphala with reference to anti-cancer property, *Comb Chem High Throughput Screen*, 18 (9) (2015) 846-854. DOI:10.2174/1386207318666151119114609.

6 Lee W-Y, Lee C-Y, Kim Y-S & Kim C-E, The methodological trends of traditional herbal medicine employing network pharmacology, *Biomolecules*, 9 (8) (2019) 362. DOI: 10.3390/biom9080362.

7 Madireddy P, Wadhwa S, Singh S & Aligireddy N, Role of polyherbal formulations in the management of polycystic ovarian syndrome, *AIP Conf Proc*, 2800 (1) (2023) 020206. DOI:10.1063/5.0166080.

8 Das C & Das D, Essential parameters for preparation of Ayurvedic fermented products Asava and Arista: An overview, *Res J Pharm Technol*, 12 (3) (2019) 1473-1480. DOI:10.5958/0974-360X.2019.00244.0.

9 Patwardhan B K, Vaidya A D, Chorghade M S & Joshi S P, Reverse pharmacology and systems approach for drug discovery and development, *Curr Bioact Compd*, 4 (4) (2008) 201-212. DOI:10.2174/157340708786847870.

10 *World Health Organization*, Traditional medicine strategy 2002-2005, Geneva, *WHO*, (2002), Report No.: WHO/EDM/TRM/2002.1.

11 Athridev G, *Astanga Hridayam of Vagbhata*, (Chowkhamba Sanskrit Sansthan, Varanasi), (1993).

12 Nirmal S, Yogaratnakara: An important source book in medicine, *Indian J Hist Sci*, 27 (1) (1992) 32-40"

13 Azziz R, Carmina E, Dewailly D, Diamanti-Kandakaris E, Escobar-Morreale H F, *et al*., The androgen excess and PCOS society criteria for the polycystic ovarian syndrome: The complete taskforce report, *Fertil Steril*, 91 (2) (2009) 456-488. doi:10.1016/j.fertnstert.2008.06.035.

14 Bhingardive K B, Sarvade D P & Bhatted S, Clinical efficacy of *Vamana Karma* with *Ikshwaku Beeja* Yoga, followed by *Shatapushpa Ghanavati* in the Management of *Artava Kshaya* w.s.r to polycystic ovarian syndrome, *AYU*, 38 (3-4) (2017) 127-132. DOI: 10.4103/ayu.AYU_192_16

15 Sayyad S F, Randive D S, Jagtap S M, Chaudhari S R & Panda B P, Preparation and evaluation of fermented Ayurvedic formulation: Arjunarishta, *J Appl Pharm Sci*, 2 (5) (2012) 122-124. DOI:10.7324/JAPS.2012.2521.

16 Ram A, Lauria P, Gupta R, Kumar P & Sharma V N, Hypocholesterolaemic effects of *Terminalia arjuna* tree bark, *J Ethnopharmacol*, 55 (3) (1997) 165-169. doi: 10.1016/s0378-8741(96)01493-6.

17 Hegde S, Hegde H V, Jalalpure S S, Peram M R, Pai S R, *et al*., Resolving identification issues of *Saraca asoca* from its adulterant and commercial samples using phytochemical markers, *Pharmacogn Mag*, 13 (Suppl 2) (2017) S266-S272. DOI:10.4103/pm.pm_417_16.

18 Pohjamo S P, Hemming J E, Willfor S M, Reunanen M H T & Holmbom B R, Phenolic extractives in *Salix caprea* wood and knots, *Phytochem*, 63 (2) (2003) 165-169. DOI:10.1016/S0031-9422(03)00050-5.

19 Gamble, J S, *Flora of the Presidency of Madras*, Vol I-III, (Adlard and Son Ltd., London), (1915-1936).

20 Khandelwal K R, *Practical pharmacognosy: Techniques and experiments*, 19th ed, (Nirali Prakashan, Pune), (2005).

21 *The Ayurvedic Pharmacopoeia of India*, Part I, (Government of India, Ministry of Health and Family Welfare, Department of Indian Systems of Medicine & Homoeopathy, New Delhi), (2001).

22 Shalavadi M H, Physicochemical and phytochemical screening of *Convolvulus pluricaulis* collected from Bagalkot, Karnataka, *Int J Green Pharm*, 12 (3) (2018) S625-S632. DOI:10.22377/ijgp.v12i03.2031.

23 Sunita S, Patil Y, Joshi M, Menon S & Mhatre M, Simultaneous quantification of pharmacological markers quercetin and berberine using HPTLC and HPLC from a polyherbal formulation *Pushyanuga Churna*, *J AOAC Int*, 102 (4) (2019) 1003-1013. DOI:10.5740/jaoacint.18-0380.

24 Vasava P & Chakraborthy G S, Chromatographic analysis of polyherbal extract and formulation by HPTLC and GC-MS methods, *J Nat Remedies*, 25 (1) (2025) 113-119. DOI:10.18311/jnr/2025/43942.

25 Anonymous, *The Ayurvedic Pharmacopoeia of India*, Part I, Vol. VI, 1st ed, (Ministry of AYUSH, Government of India, New Delhi), (2008).

26 Prajapati D, Patel M & Dharamsi A, Development and evaluation of a polyherbal tablet for polycystic ovarian syndrome (PCOS), *J Nat Remedies*, 22 (2) (2022) 161-174. DOI:10.18311/jnr/2022/28557.

27 Anonymous, *Indian Pharmacopoeia*, Vol. I & II, (Indian Pharmacopoeia Commission, Ministry of Health and Family Welfare, Government of India, Ghaziabad, India), (2014).

28 Anonymous, *The Ayurvedic Pharmacopoeia of India*, 1st ed, Vol. 7, (Department of AYUSH, New Delhi), (2008), 295-303.

29 *ICH Q1A(R2), Stability Testing of New Drug Substances and Products* (2003).

30 Tamboli A K & Wadkar K A, Preparation and evaluation of fermented Ayurvedic formulation: Vasakasav, *J Pharmacogn Phytochem*, 7 (4) (2018) 1840-1842.