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Pharmaceutical Study of *Aconitum heterophyllum* (*Ativisha*) Shodhana and Analysis Based on High-Performance Thin-Layer Chromatography and High-Performance Liquid Chromatography

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ABSTRACT

Ativisha (*Aconitum heterophyllum*), a prominent drug in the Ranunculaceae family, is extensively cited in the *Charaka* and *Sushruta Samhitas* for its therapeutic efficacy. However, its potent diterpene alkaloids necessitate “*Shodhana*” (systematic purification) to ensure clinical safety and enhance pharmacological action. This review examines classical *Shodhana* protocols and their impact on the plant’s chemical profile, specifically evaluating how these traditional processes mitigate toxicity. This work highlights the importance of classical *Shodhana* process in optimizing the bioavailability of phytochemical metabolites and its impact on the plant’s chemical profile, ensuring the safety of herbal interventions in primary healthcare.

1. INTRODUCTION

A vital component of the Ayurvedic system is *Ativisha* (*Aconitum heterophyllum*), a Himalayan herb revered by practitioners as *Balanam Sarvada Pathya*^[1] (always wholesome for children) for its pediatric efficacy. While classical texts like the *Charaka Samhita*^[2] quote it as *Deepaniya*, *Pachaniya*, *Sangrahai*, and *Sarv dosha harananam*, explaining its use for obesity and gastrointestinal disorders, its potent phytochemical profile requires cautious *Shodhana* (purification). *Shodhana* of *Ativisha* is a critical pharmaceutical intervention to refine its immune-modulatory properties while ensuring its clinical safety and to perform a qualitative analysis for secondary metabolites, high-performance thin-layer chromatography (HPTLC) and high-performance liquid chromatography (HPLC).

The *Shodhana* process is essential to transform the raw tuber into a safe therapeutic agent. This introduction explores how systematic purification protocols optimize the herb’s pharmacological benefits.

1.1. Aims and Objectives

1.1.1. Aim

To carry out the *Shodhana* (classical purification) of *Ativisha* (*A. heterophyllum*) and to evaluate its phytochemical and chromatographic profile before and after purification.

1.1.2. Objectives

1. To procure and authenticate raw *Ativisha* (*A. heterophyllum*) tubers from an authentic source
2. To perform *Shodhana* of *Ativisha* as per the method described in classical Ayurvedic texts
3. To evaluate and compare the organoleptic characteristics of *Ativisha* before (*Ashodhita*) and after (*Shodhita*) purification

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4. To carry out qualitative phytochemical screening of *Shodhita Ativisha* for the presence of secondary metabolites (alkaloids, glycosides, tannins, flavonoids, etc.)
5. To perform HPTLC profiling of *Shodhita Ativisha*
6. To perform HPLC analysis of *Shodhita Ativisha* for identification and quantification of its active constituents.

1.2. Description of *Ativisha*^[3]

Latin name: *A. heterophyllum* Wall.

Family: Ranunculaceae

Synonyms: *Aruna*, *Ghunapriya*, *Visha*, *Shishu*, *Bhaishajya*, *Shuklakanda*, *Bhangura*.

1.3. Classification According to *Bhruhatrayi*, *Laghutrayi* and *Nighantu*^[4]

- *Charka* - *Lekhaniya*, *arsoghna*, *tikta skandha*, *sirovirecana*
- *Sushruta* - *Pippalyadi*, *mustadi*, *vacadi*
- *Ashtang Sangraha* - *Lekhaniya*, *Arsoghna*, *Pippalyadi*, *Mustadi*, *Vacadi*
- *Astanga Hrdaya* - *Pippalyadi*, *Mustadi*, *Vacadi*
- *Dhanvantari Nighantu* - *Guducyadi Varga*
- *Sodhala Nighantu* - *Guducyadi Varga*, *Anekārtha Varga*
- *Kaiyadeva Nighantu* - *Osadhi Varga*
- *Bhav Prakasa Nighantu* - *Haritakyadi Varga*
- *Raja Nighantu* - *Pippalyadi Varga*, *Upavisa Gana*
- *Priya Nighantu*^[5] - *Shatapushpadi Varga*.

1.4. Properties of *Ativisha*^[6]

- *Rasa*: *Tikta* (bitter), *Katu* (pungent)
- *Guna*: *Laghu*, *Ruksha*
- *Virya*: *Ushna* (Hot potency)
- *Vipaka*: *Katu*
- *Karma*: *Dipana*, *Pachana*, *Sanghrahika*, *Kaphapittahara*.

1.5. Definition of *Shodhana*^[7]

In Ayurvedic Formulary of India, *Shodhana* is explained as the process of eliminating impurities and increasing drug potency. *Shodhana* is a purification process in which *kshalana* (washing), *Mardana* (pounding), *Bhavana* (Levigation), *Swedana* (boiling), *Bharjana* (frying), and *Nirvana* (Heating and dipping in specified liquids) are carried out on drugs to eliminate impurities and enhance their therapeutic use.

1.6. Importance of *Shodhana*

1. Eliminate physical and chemical impurities from raw drug
2. Detoxifies drug by neutralizing toxins
3. Enhance therapeutic effects of drug
4. Impart desirable organic properties
5. Make drugs safe and suitable for administration.

2. MATERIALS AND METHODS

Shodhana of *Ativisha* is not described in *Bhruhatrayi*, *Laghutrayi* and *Nighantus* except *rajnighantu*.

2.1. *Shodhana* of *Ativisha*^[8]

- Reference - *Raj Nighantu* (*Pippalyadi Varga*)
- Principle method - *Swedana*

- Equipment - Heater, weighing machine, container, Spatula.
- Ingredients - *Ativisha* (120 g), *Gomaya* (1 kg).

2.2. Pre-procedure

1 kg of *Gomaya* was taken in stainless-steel vessel and 4 times of water was added to it. It was heated over medium flame until the liquid was reduced to 1/4th and then filtered through cotton cloth. 1 L of *Gomaya kwatha* was prepared (Figure 1).

2.3. Procedure

Ativisha was dried in sunlight to remove moisture and all the physical impurities were removed. *Ativisha* was pounded in *Ulukhala Yantra* and tied into *Pottali* using clean cotton cloth. Then, it was subjected to *Swedana* using *Gomaya kwatha* in *Dolayantra* for 3 h (Figures 2 and 3).

After 3 h, *Pottali* was untied, and *Ativisha* was washed with hot water and dried in sunlight until completely dry. *Shuddha Ativisha* was collected and stored in an airtight container (Figures 4-6) (Table 1 and 2).

2.4. Precautions

- Physical impurities need to be eliminated
- Medium heat is used for *Dolayantra Swedana*
- Properly dried under sunlight
- Proper gloves in hand and face mask should be used.

3. RESULTS

Alkaloids, carbohydrates, proteins and amino acids, saponins, glycosides, quinones, flavonoids, and terpenoids are present in the leaf, root and stem of *A. heterophyllum*.

- HPTLC is a widely utilized analytical technique that offers significant advantages in the qualitative and quantitative analysis of a variety of chemical substances.
- HPLC is a type of liquid chromatography, meaning the mobile phase is a liquid. The information that can be obtained using HPLC includes identification, quantification, and resolution of a compound.
- HPTLC and HPLC are used to determine qualitative analysis of secondary metabolites in *A. heterophyllum* (*Atis*) roots.

3.1. Qualitative Analysis of Secondary Metabolites in *A. heterophyllum* (*Atis*) Roots (Table 3)

Mention in table 3.

3.2. HPTLC Fingerprinting of *A. heterophyllum* Roots

3.2.1. Preparation of sample solution

10 g of sample grounded and then 100 mL of methanol is added. The sample is sonicated for 30 min. After that, the sample solution is filtered and the clear solution is used for analysis.

Chromatographic condition

- Stationary phase: TLC Silica gel 60 F₂₅₄ Aluminum sheet
- Mobile phase: Chloroform: Methanol (8:2)
- Saturation time: 20 min
- Band length: 8.0 mm
- Injection volume: 10 µL
- Migration distance: 70 mm
- Visualization: At 254 nm and 366 nm UV

3.2.2. Interpretation

In HPTLC fingerprints, bands of phyto-chemicals were observed under UV 254 nm and 366 nm. This is a specific fingerprint for roots of *A. heterophyllum* in the mentioned chromatographic conditions.

Peak profiling of *A. heterophyllum* RM by HPLC-PDA.

3.2.3. Experimental methodology

Analysis was performed on a Prominence-i HPLC system (Shimadzu, Japan). Separation was achieved using a Shodex C18-4E (5 μ m, 4.6 $\times$ 250 mm) column subjected to binary gradient elution. The two solvents used for the analysis were water containing 0.1% Acetic acid in water (solvent A) and Acetonitrile (solvent B). Column temperature was kept 30°C and the flow was set to 1.0 mL/min during the analysis. 10 μ L of standard and test solution were injected. Wavelength was set at 345 nm.

3.2.4. Gradient program (Table 4)

A binary gradient elution was used with solvent A (0.1% acetic acid in water) and solvent B (acetonitrile) over a total run time of 75 minutes. Solvent B was increased stepwise from 5% to 80% between 0 and 65 minutes, then reduced back to 5% by 70 minutes to re-equilibrate the column. This gradual increase in organic solvent helped separate both polar and non-polar phytoconstituents in the sample.

3.2.5. Sample preparation

2.0 g sample was dissolved in 25 mL of (Water: methanol: 5:5) by sonicating for 30 min then centrifuged at 9000 rpm for 10 min and filtered using 0.45 μ nylon filters then used for the analysis.

3.2.6. Chromatogram

The HPLC-PDA chromatogram of *Aconitum heterophyllum* showed well-resolved peaks for the different phytoconstituents in the root sample, recorded at 345 nm. This chromatographic pattern serves as a characteristic fingerprint of the drug for its identification and quality assessment.

3.2.7. Contents (area %)

HPLC analysis revealed thirteen peaks with varying retention times and area percentages. Two major peaks were observed at 26.305 minutes and 31.055 minutes, contributing 28.165% and 20.419% of the total area respectively, together accounting for nearly half the total content. Other notable peaks appeared at 13.062 minutes (13.139%) and 12.001 minutes (8.207%), while the remaining peaks contributed minor percentages (0.966%–2.522%) (Table 5).

3.2.8. Interpretation

Thirteen peaks are detected in the given sample. Out of which two major peaks are observed at RT 26.305 min and 31.055 min with areas 28.165% and 20.419%, respectively.

4. DISCUSSION

The analysis revealed a rich phytochemical profile that supports the therapeutic claims discussed in our previous conversations regarding *Ativisha*'s potency.

- Qualitative screening: The roots tested positive for Alkaloids, Flavonoids, Phenols, and Saponins. However, the sample tested negative for Tannins. The presence of alkaloids is particularly significant, as these often correspond to the diterpene alkaloids responsible for the herb's analgesic and anti-inflammatory properties.
- HPTLC fingerprinting: Using a mobile phase of Chloroform

and Methanol (8:2), the researchers observed specific bands of phytochemicals at 254 nm and 366 nm. These bands serve as a specific diagnostic "fingerprint" that can be used to authenticate the roots of *A. heterophyllum* and distinguish them from substitutes like *Mustaka*.

- HPLC-PDA peak profiling: The HPLC analysis detected thirteen distinct peaks in the sample. Two major peaks were identified, which dominate the chemical composition:
 - Peak 1: Observed at a retention time (RT) of 26.305 min, accounting for 28.165% of the total area
 - Peak 2: Observed at an RT of 31.055 min, accounting for 20.419% of the total area
 - Other notable peaks appeared at RT 13.062 (13.139%) and RT 12.001 (8.207%).

5. CONCLUSION

This data suggests that nearly 50% of the detectable phytochemical content is represented by just two major compounds, which likely play a central role in the plant's medicinal efficacy.

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7. AUTHORS' CONTRIBUTIONS

All authors give equal contribution to the making of this manuscript.

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9. ETHICAL STATEMENT

Ethical approval was not required for this study as it is an experimental study.

10. CONFLICT OF INTERESTS

The authors declare no conflicts of interest regarding the publication of this paper.

11. DATA AVAILABILITY STATEMENT

The data analyzed in this review were obtained from publicly available sources, including peer-reviewed articles, observational studies, and surveys accessible through databases.

12. DISCLAIMER/VIEW AND OPINIONS

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13. AI-USE DECLARATION

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REFERENCES

1. Gyanendra P. Shodhala nighantu by Acharya Shodhala. Varanasi: Chaukhamba Krishnadas Academy; 2012. p. 141.
2. Kashinatha, S Charak samhita of agnivesha. Vol 1., Ch. 25/40. Varanasi, India: Chaukhambha Bharati Academy; 2022. p. 407.
3. Gyanendra P. Dravyaguna vijyana. 1st ed., Vol. 1. Varanasi, India: Chaukhambha Krishnadas Academy; 2001. p. 272-8.
4. Sastry JL. Dravyaguna vijyana. Vol. 2. Varanasi India: Chaukhamba Orientalia; 2017. p. 23.
5. Sharma PP. Hindi commentry 'Padmakhya'. Varanasi: Chaukhambha Surbharati Prakashan; 2004. p. 108.
6. The Ayurvedic Pharmacopoeia of India. Vol. 1. Part 1. Delhi, India: The Controller of Publications, Government of India, Ministry of Health and Family Welfare, Department of Ayush; 2001. p. 22-3.
7. Magesh R, Jayalekshmi R. Review article of Shodhana in rasashastra. J Ayurveda Integr Med Sci 2023;8(4):156.
8. Indradev T. Rajanighantu of pandit narahari. 5th ed. Varanasi: Chaukhamba Krishnadas Academy; 2010. p. 163.

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Figure 1: Gomaya Kwatha preparation



Figure 4: Shudha Ativsha in Pottali



Figure 2: Ashudha Ativisha and Kwatha



Figure 5: Shuddha Ativisha dried



Figure 3: Daulayantra Swedana



Figure 6: Pounded in ulukhala for churna Nirmana

Table 1: Observation after *Shodhana* process

Color	Brown
Taste	Bitter
Odor	Gomaya Sadrishya

Table 2: Result of *Ativisha Shodhana*

Initial weight of <i>Ativisha</i> (g)	After removing impurities and pounding (g)	After <i>Swedana</i> (g)	After drying (g)	Loss (%)
120	95	124	92	23.3

Table 3: Secondary metabolites in *Aconitum heterophyllum* (*Atis*) roots

S. No.	Sample name	Alkaloids	Flavonoid	Phenol	Saponin	Tannin
1	<i>Aconitum heterophyllum</i> roots	Positive	Positive	Positive	Positive	Negative

Table 4: Gradient program

Time (min)	Solvent B (%)
0	5
5	10
15	15
25	15
30	20
50	30
60	60
65	80
70	5
75	5

Table 5: Retention time with area (%)

S. No.	Retention time	Area	Area %
1	3.12	22502	1.09
2	3.95	19936	0.966
3	11.15	150966	7.316
4	12.001	169346	8.207
5	13.062	271120	13.139
6	13.605	47138	2.284
7	26.305	581195	28.165
8	31.055	421354	20.419
9	36.664	31192	1.512
10	37.217	25121	1.217
11	43.642	26982	1.308
12	44.068	29536	1.431
13	55.919	52043	2.522