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Research Article

High-performance thin-layer chromatography (HPTLC) fingerprinting and analytical evaluation of *Vajra Guggulu*: a Herbo-mineral formulation

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Abstract:
Introduction: *Vajra Guggulu* is a classical Ayurvedic formulation in which every medication comprises a combination of chemical components with proven neuroprotective properties. The study's goals were to perform HPTLC to identify phytochemicals in *Vajra Guggulu* and physiochemically analyze the formulation of *Vajra Guggulu* according to the Ayurvedic Pharmacopoeia of India (API) standards.
Methods: *Vajra Guggulu* was prepared according to the reference provided in the *Ayurvedic* classical text *Ras Ratnakar*, and its quality was evaluated using organoleptic characteristics and physiochemical specifications as described in API, followed by Standard HP-TLC analysis on the samples.
Results: The physicochemical evaluation of *Vajra Guggulu* confirms its authenticity and quality. HPTLC analysis reveals the presence of phytochemicals that can delay the progression of inflammatory, peripheral vascular, and neurological illnesses.
Conclusion: This study will serve as a reference standard for medicine manufacturing, quality analysis, and assessing Vajra Guggulu's future therapeutic potential.

Keywords- Ayurveda, Bioactivity, Neuroprotective, Peripheral neuronal Disorders, Peripheral Vascular Disorders

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Introduction:
Medicine analysis is required to ensure that the ingredients, process, and final product are all standardized. Herbal formulations have several challenges in terms of quality analysis and standardization. Herbal formulations undergo a variety of quality-related obstacles resulting from the characteristics of the herbal ingredients and the numerous secondary metabolites they possess. In addition, growing, harvesting, storing, and drying procedures are examples of the internal and extrinsic factors that contribute to alterations in the chemical composition of herbs [1, 2].
Good manufacturing processes in the production units are ensured by a physiochemically standardized product. To make sure that the formulations include active marker compounds, a variety of chromatographic methods are employed. Thin layer chromatography (TLC) is the foundation of high-performance thin-layer chromatography (HPTLC), an analytical method that

has been improved to enable quantitative analysis of the chemicals and improve the resolution of the compounds to be separated. Better resolution is made possible by several improvements, such as the use of better TLC plates in the stationary phase with smaller sizes of particles. HPTLC is employed in the identification and quantification of active compounds as well as the identification and detection of contaminants.[3]

Vajra Guggulu is a traditional Ayurvedic polyherbal formulation mentioned in Ayurvedic classical text *Rasratnakar* under management of gouty arthritis (~*Vaatrakta*) [4], in which each drug contains a combination of chemical constituents that are known to have neuroprotective properties, as well as antioxidants, anti-inflammatory agents, anti-diabetics, analgesics, antinociceptives, fibrinolytics, vasodilators, and compounds with other potential actions. These actions may help in the management of peripheral neuronal and vascular disorders, inflammatory disorders, and pain management for symptomatic relief. No prior attempt is documented in the analysis of *Vajra Guggulu*.

The goal of the current study was to analyse *Vajra Guggulu* based on physiochemical parameters – weight variation, hardness, friability, disintegration time, moisture content, total ash value, alcohol-soluble extractive value, water-soluble extractive value, Ph value, and total ash value as per the norms of Ayurvedic Pharmacopoeia of India (API) [5] and Indian Pharmacopoeia (IP)[6]. The study also focused on identifying the phytochemicals in Vajra Guggulu using HPTLC and evaluating the therapeutic potential of Vajra Guggulu.

MATERIAL AND METHODS:

The raw drugs for the Vajra Guggulu formulation were obtained from a reputable local raw herbal medicine supplier. An in-house specialist botanist authenticated all of the crude drugs, and the voucher for the specimen sample was stored in the department. Table 1 describes the individual ingredients in detail.

Table No 1: Ingredients and Composition of Vajra Guggulu

S. no	Drug Name	Latin Name	Part Used	PROPORTION	Chemical Constituent
1.	Shunthi	*Zingiber Officinale Rosc.*	Rhizome	1 Part	(6)-gingerol and (6)-paradol, shogaols, zingerone, and Galanals A and B.
2.	Maricha	*Piper Nigrum L.*	Fruit	1 Part	Piperine
3.	Pipalli	*Piper Longum L.*	Fruit/ Roots	1 Part	Piperine/ Piperlongumine/ Piperlonguminine
4.	Haritki	*Terminalia Chebula Retz.*	Fruit	1 Part	Gallic/Ellagic Chebulic/Chebulagic Ascorbic
5.	Bibhitaka	*Terminalia Belerica Roxb.*	Fruit	1 Part	Gallic acid, ellagic acid, ethyl gallate, chebulagic acid
6.	Amalki	*Phyllanthus Emblica L.*	Fruit	1 Part	Phembembaic Acid, Gallic Acid, Mymic Acid
7.	Danti	*Baliospermum Montanum Muell.*	Roots	1 Part	Terpenoids, Glycosides, Saponins, Alkaloids
8.	Chitraka	*Plumbago Zeylanica L.*	Roots-Bark	1 Part	Plumbazeylanone, Plumbagic Acid, B Sitosterol, Lupeol
9.	Trivrata	*Operculina Turpethum Linn.*	Leaves- Fruit	1 Part	α-andβ-Turpethin, glycosides, coumarins, Scopoletin
10.	Shathi	*Hedychium Spicatum Sm.*	Rhizome	1 Part	Hedychenone, B- Furaldehyde
11.	Vidanga	*Embelia Ribes Burm F.*	Seeds	1 Part	Embelin/ Embeliaribyl Ester, Benzoxadiazine
12.	Mustaka	*Cyperus Rotundus L.*	Rhizome	1 Part	Sugetriol Triacetate, Patchoulenone, Isopatchoulenone, Sugeonyl Acetate
13.	Haridra	*Curcuma Longa Linn.*	Rhizome	1 Part	Curcuminoids , Curcumin, Desmethoxycurcumin
14.	Bakuchi	*Psoralea Corylifolia* Linn.	Seeds	1 Part	Coumarins, Flavonoids, And Meroterpenes
15.	Indrayava	*Holarrhena Antidysentrica L.*	Bark-Seeds	1 Part	Coumarins, Ergosterol, Saponins, Triterpenoids
16.	Vacha	*Acorus Calamus L.*	Roots-Rhizome	1 Part	Saponins/Flavonoids, β-Asarone, Z-isoelemicin
17.	Ankola	*Alangium Salvifolium L.f.*	Roots- Bark	1 Part	Alangine/Alangicine, kaempferol, and kaempferol 3-O-b-D-glucopyranoside.
18.	Kushta	*Sassurea Lappa Decne.*	Roots	1 Part	Dehydrocostus Lactone, Costunolide and Cynaropicrin
19.	Aragvadha	*Cassia Fistula L.*	Flower- Leaves/ Bark/ FruitPulp	1 Part	Flavon-3-ol/Anthroquinones, Leucine, Arginine.
20.	Guggulu	*Commiphora Mukul Arnott.*	-	19 Parts	Guggulsterones , Terpenoids, Flavonoids
21.	Bhallataka taila	Oil extracted from *Semecarpus Anacardium Linn.*	-	2 Parts	Semecarpetin, Nall flavanone, Semecarpuflavanone, Anacarduflavone
22.	Tamra Bhasma	Copper Calx	-	1 Part	-
23.	Taala Bhasma	Arsenic Trioxide	-	1 Part	-

Authentication of Bhasma:
The Samples of Tamra Bhasma and Taala Bhasma were evaluated for Bhasma Standards according to the parameters of Pharmacopoeial standards for Ayurvedic formulations (PSAF) and Ayurvedic Pharmacopoeia of India (API).

Table 2: Authentication parameters of Tamra and Tala Bhasma

S.No	Parameters	Tamra Bhasma	Tala Bhasma
1.	Heavy Metals	Not detected other than Cu	Not detected other than As
2.	Moisture Content	0.3%	Not Present
3.	pH Value	5.2	6.8
4.	Fineness	Passes through 160 mesh size	Passes through 160 mesh size
5.	Limit Test	CuS is Assessed	Arsenic sulphate with trace of oxide found.

Method of Preparation of Vajra Guggulu.

- Vajra Guggulu was prepared in accordance with Ayurvedic Pharmacopoeia of India (API) and Indian Pharmacopoeia (IP-2018) standards at a study center pharmacy.
- Vajra Guggulu was prepared using the reference from Ras-Ratnakar. [4]
- All of the crude medications [1–19] listed in Table 1 were processed for drug preparation after initial cleaning, shade drying, powdering, and sieving. The Shodhan of Guggulu with Triphala decoction was followed.
- Following that, one part of each of the previously created crude medications (1–19) and nineteen parts of Shodhit Guggulu were mixed thoroughly in an end-runner machine with two parts Bhallataka Oil, one part Tamra Bhasma, and one part Taala Bhasma.
- The mixture was dried for 45 minutes using a hot air tray dryer. Next, using a Granulator, the dried mixture was transformed into granules or sizable agglomerates.
- The binding agent Shankha Jiru was mixed into the granules in a 1:10 ratio. The mixture was then formed into 720 mg tablets using a multi-punch tablet punching machine.

Physiochemical Analysis of Vajra Guggulu:
Physiochemical parameters utilized in accordance with API (Ayurvedic Pharmacopoeia of India)[5] and IP (Indian Pharmacopoeia)[6] standards for the analysis of the tablet (Vajra Guggulu).

Chromatograph profiling by HPTLC:[7]
Preparation of Test solution: Weigh 5 g of the sample in a beaker and add 100 mL of Ethanol to it. Sonicate for 16 hours and filter with the help of simple filter paper. Filtrate is used as a Test solution thus obtained for HPTLC fingerprinting.

Table 3: Chromatographic conditions for HPTLC of Vajra Guggulu

Chromatographic Conditions:	
Application Mode	CAMAG Linomat 5 (S/N: 280008) Applicator
Filtering System	Simple filter
Stationary Phase	MERCK - HPTLC Silica gel 60 F_{254} on Aluminium sheets
Application (Y axis) Start Position	15 mm
Development End Position	80 mm from the plate base
Sample Application Volume	10 µL, 15 µL, 20 µL
Distance Between Tracks	21.4 mm
Development Mode	CAMAG TLC Twin Trough Chamber
Chamber Saturation Time	20 minutes
Mobile Phase (MP)	Toluene Ethyl acetate (9:1 v/v)
Visualization	@ 254 nm and @366 nm
Drying Mode, Temp. & Time	Room Temperature for 5 minutes.

Identification of Compounds:
For HPTLC interpretation,the unknown component's R_f Value , Relative R_f Value and % area at 254nm and 366nm was compared to the R_f Value , Relative R_f Value of known components listed in the Phytochemical fingerprint and chemometrics online database[8] and IMPPAT 2.0[9].

Results and Discussion :
The Organoleptic characteristics of Vajra Guggulu is mentioned in Table 4. The Physico-chemical parameters result along with its normal range and limit as per the norms of Ayurvedic Pharmacopoeia of India (API) [5] and Indian Pharmacopoeia (IP)[6]. is mentioned in table 5.

Table No 4: Organoleptic characteristics of Vajra Guggulu

Colour	Brownish Green
Odour	Pleasant
Taste	Astringent
Consistency	Tablet

Table No 5: Physio-Chemical Parameters of Vajra Guggulu

S.no	Parameters	Result	Normal Limit[5,6]
1.	Loss on Drying at 110 c (%w/w)	2.7	≤ 15%
2.	Total Ash Value (%w/w)	6.2	≤ 10%
3.	Acid insoluble Ash (%w/w)	1.75	(Variable)
4.	Water soluble extractive value (%w/w)	30.0	15-30%
5.	Alcohol soluble extractive value (%w/w)	32.9	20-40%
6.	P^H Value (10% Aqueous)	7.0	5.5-7.5
7.	Tablet Avg. Weight Variation (Mg)	720	-
8.	Tablet Disintegration Time (mins)	58	(Variable)
9.	Tablet Hardness (Kg/cm^2)	5.0	4-12 kg/cm²
10.	Tablet Friability (%)	1.6	≤ 2%

The HPTLC Chromatograph of Vajra Guggulu developed at wavelength of 254nm with volume of 10.0 µL, 15.0 µL, 20.0 µL is depicted in figure 1, 2, 3 respectively. Chromatograph developed at wavelength of 366nm with volume 20.0 µL is depicted in figure 4, showing the R_f Values, Chromatograph fingerprint, peak height and percentage area. By comparing the results (Fig 1-4) at Phytochemical fingerprint and chemometrics online database[8] and IMPPAT 2.0[9] chemical substances were then identified is mentioned in Table 6-9 along with their Bioactivity [Source: PubChem, Google Scholar].

Figure 1: HPTLC Chromatograph of Vajra Guggulu @254 nm and 10.0 µL volume (A=Fingerprint, B= Peak height, C= R_f Value and Area percentage)

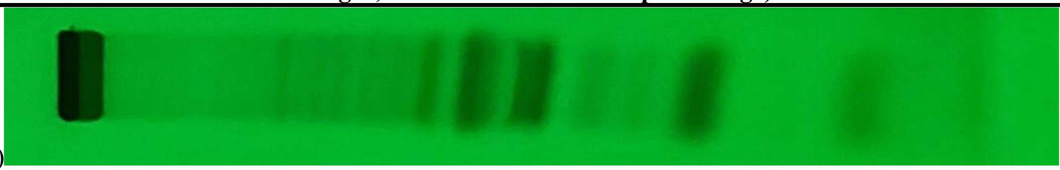


Fig 1(A)

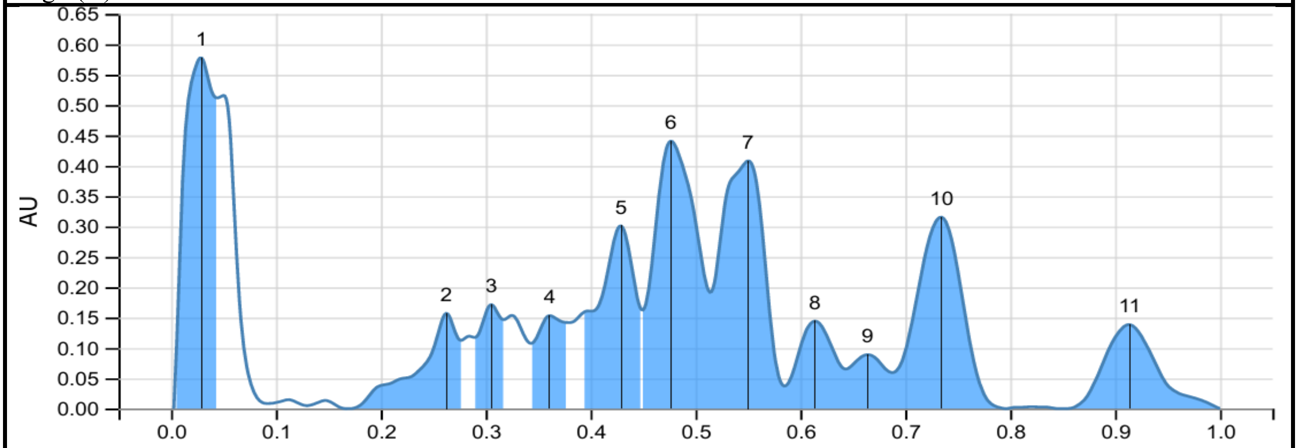


Fig 1(B)

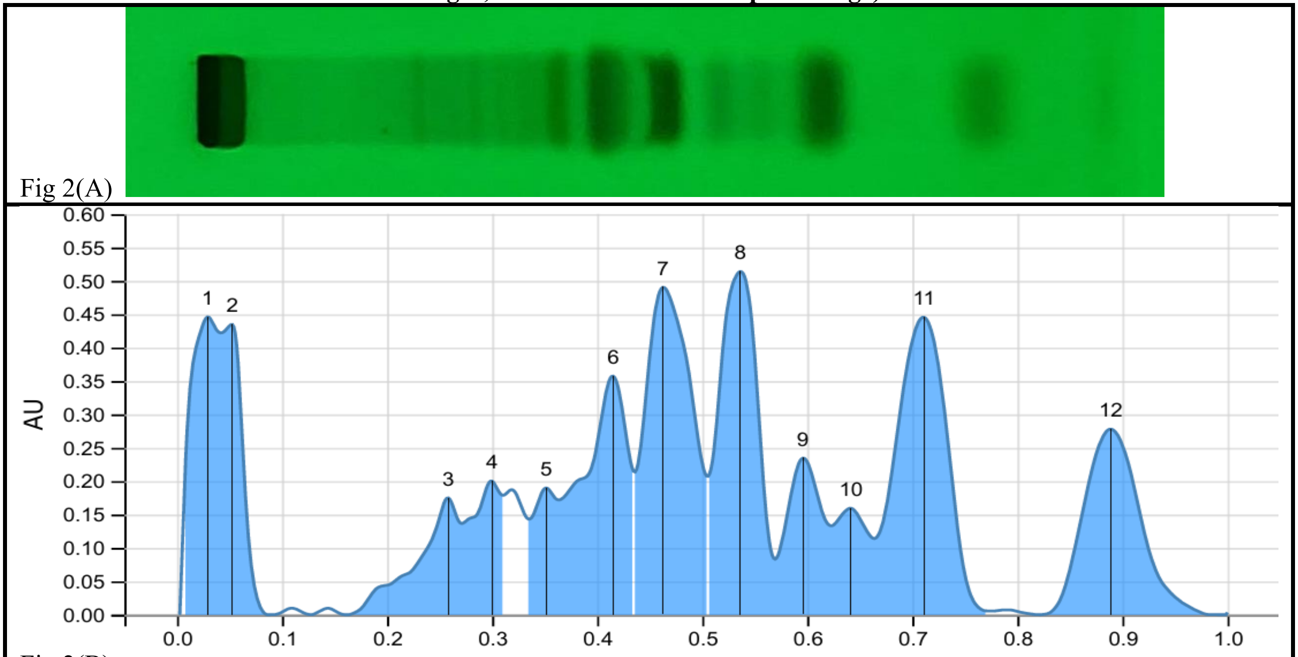
Peak #	Start		Max			End		Area	
	R_F	H	R_F	H	%	R_F	H	A	%
1	0.003	0.0000	0.029	0.5776	19.95	0.044	0.5118	0.01921	15.91
2	0.171	0.0000	0.263	0.1569	5.42	0.276	0.1124	0.00676	5.60
3	0.289	0.1173	0.306	0.1712	5.91	0.317	0.1466	0.00414	3.43
4	0.343	0.1074	0.361	0.1531	5.29	0.379	0.1417	0.00499	4.13
5	0.394	0.1595	0.429	0.3008	10.39	0.449	0.1630	0.01190	9.85
6	0.450	0.1625	0.476	0.4405	15.22	0.514	0.1917	0.02075	17.19
7	0.514	0.1917	0.550	0.4080	14.09	0.585	0.0372	0.01971	16.33
8	0.585	0.0372	0.614	0.1441	4.98	0.643	0.0652	0.00583	4.83
9	0.643	0.0652	0.664	0.0889	3.07	0.688	0.0594	0.00340	2.81
10	0.688	0.0594	0.735	0.3152	10.89	0.796	0.0000	0.01599	13.24
11	0.853	0.0000	0.914	0.1383	4.78	1.000	0.0006	0.00806	6.68

Fig 1(C)

Table No 6: Substance name and their Bioactivity based on R_f Value@254 nm and 10.0 µL volume

Peak No.	R_f Value	% Area	Substance Name	Bioactivity (Source: PubChem, Google Scholar)
1.	0.029	19.95	Chelerythrine	Protein kinase C inhibitor, Antibacterial, Antineoplastic Agent. [PubChem], Anti-Inflammatory effect. [10]
2.	0.263	5.42	Glycosides	Analgesics, Anti-Inflammatory. [11]
3.	0.306	5.91	Phenols	Anti-Oxidant, Anti-Cancer Activity. [12]
5.	0.429	10.39	Tetrahydroharmine	Anti-Microbial. [13]
6.	0.476	15.22	Berberine	Vasodilator (Potassium Channel Blocker), Anti-Neoplastic, Anti-Fungal, Neuroprotective (Acetylcholinterase Inhibitor), Anti-Lipidemic, Inflammatory disorders (Kappa B Kinase Inhibitor), Anti-Diabetic, Anti-Oxidant. [PubChem]
7.	0.550	14.09	Hydroxylamine	Anti-Oxidant, Anti-Fungal, Anti-Bacterial [PubChem]
8.	0.614	4.98	Ergotamine	Vasodilator Agent, Smooth muscle relaxant, Anti-Inflammatory agent, Non-Narcotic Analgesics, Serotonergic Agonist [PubChem]
9.	0.664	3.07	Alkaloids	Type II Diabetes Mellitus, Cardioprotective Agent, Dyslipidemia, Analgesics, Anti-Inflammatory, Antiplaque Agent, Vascular disorders. [14]
10.	0.735	10.89	Rescinnamine	Vasodilator and Anti-Hypertensive agent (Angiotensin-Converting enzyme) [PubChem]

Figure 2: HPTLC Chromatograph of Vajra Guggulu @254 nm and 15.0 µL volume (A=Fingerprint, B= Peak height, C= R_f Value and Area percentage)



Peak #	Start R_F	Start H	Max R_F	Max H	Max %	End R_F	End H	Area A	Area %
1	0.003	0.0000	0.029	0.4460	11.37	0.042	0.4221	0.01412	8.49
2	0.042	0.4221	0.053	0.4345	11.07	0.086	0.0000	0.00999	6.01
3	0.163	0.0000	0.258	0.1748	4.45	0.271	0.1366	0.00801	4.82
4	0.271	0.1366	0.300	0.2001	5.10	0.311	0.1789	0.00677	4.07
5	0.335	0.1432	0.351	0.1898	4.84	0.364	0.1719	0.00501	3.01
6	0.364	0.1719	0.415	0.3573	9.11	0.435	0.2148	0.01780	10.70
7	0.436	0.2138	0.463	0.4903	12.49	0.504	0.2077	0.02519	15.15
8	0.506	0.2073	0.536	0.5139	13.10	0.569	0.0832	0.02223	13.37
9	0.569	0.0832	0.596	0.2344	5.97	0.622	0.1332	0.00910	5.47
10	0.622	0.1332	0.642	0.1594	4.06	0.662	0.1145	0.00573	3.44
11	0.664	0.1140	0.711	0.4454	11.35	0.775	0.0055	0.02508	15.08
12	0.822	0.0000	0.889	0.2780	7.08	0.983	0.0000	0.01727	10.38

Fig 2(C)

Table No 7: Substance name and their Bioactivity based on R_f Value@254 nm and 15.0 µL volume

Peak No.	R_f Value	% Area	Substance Name	Bioactivity (Source: PubChem, Google Scholar)
1.	0.029	11.37	Chelerythrine	Protein kinase C inhibitor, Antibacterial, Antineoplastic Agent. [PubChem], Anti-Inflammatory effect. [10]
2.	0.053	11.07	Glycosides	Analgesics, Anti-Inflammatory. [11]
5.	0.351	4.84	1° Derivate of Guggulsterones	Anti-Inflammatory, Anti- Oxidant. [15]
6.	0.415	9.11	Glycosides	Analgesics, Anti-Inflammatory. [11]
7.	0.463	12.49	Berberine	Vasodilator (Potassium Channel Blocker), Anti-Neoplastic, Anti-Fungal, Neuroprotective (Acetylcholinterase Inhibitor), Anti-Lipidemic, Inflammatory disorders (Kappa B Kinase Inhibitor), Anti-Diabetic, Anti-Oxidant. [PubChem]
8.	0.536	13.10	Recinnin alpha Hydrase	Pyridine Alkaloid (Analgesics, Anti-Inflammatory, Neuroprotective (Peripheral nervous System)). [PubChem]
9.	0.596	5.97	Phenol	Anti-Oxidant, Anti-Cancer Activity. [12]
11.	0.711	11.35	Rescinnamine	Vasodilator and Anti-Hypertensive agent (Angiotensin-Converting enzyme) [PubChem]

Figure 3: HPTLC Chromatograph of Vajra Guggulu @254 nm and 20.0 µL volume (A=Fingerprint, B= Peak height, C= R_f Value and Area percentage)

Fig 3(A)

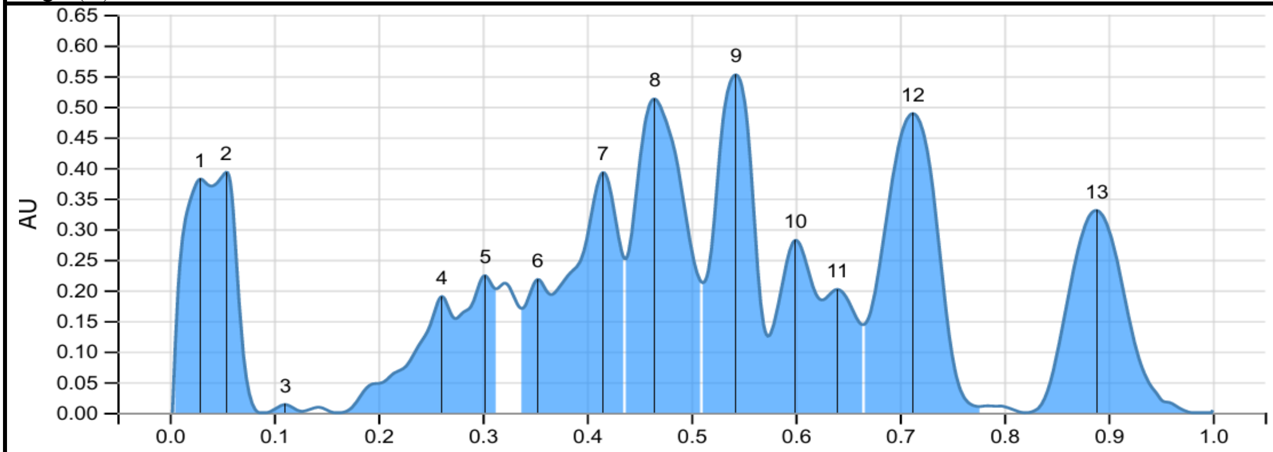


Fig 3(B)

Peak #	Start R_F	Start H	Max R_F	Max H	Max %	End R_F	End H	Area A	Area %
1	0.003	0.0000	0.029	0.3815	9.14	0.040	0.3701	0.01162	6.33
2	0.040	0.3701	0.054	0.3927	9.40	0.087	0.0000	0.01027	5.60
3	0.092	0.0000	0.111	0.0132	0.32	0.128	0.0023	0.00025	0.13
4	0.164	0.0000	0.261	0.1897	4.54	0.274	0.1537	0.00906	4.93
5	0.274	0.1537	0.303	0.2237	5.36	0.313	0.2023	0.00736	4.01
6	0.338	0.1702	0.353	0.2175	5.21	0.365	0.1930	0.00551	3.00
7	0.365	0.1930	0.415	0.3921	9.39	0.436	0.2517	0.02003	10.91
8	0.438	0.2514	0.465	0.5124	12.27	0.510	0.2136	0.02807	15.29
9	0.511	0.2123	0.543	0.5519	13.21	0.574	0.1250	0.02351	12.81
10	0.574	0.1250	0.600	0.2816	6.74	0.625	0.1841	0.01118	6.09
11	0.625	0.1841	0.640	0.2014	4.82	0.664	0.1438	0.00709	3.86
12	0.665	0.1437	0.713	0.4886	11.70	0.776	0.0099	0.02899	15.79
13	0.822	0.0000	0.889	0.3303	7.91	0.981	0.0000	0.02066	11.25

Fig 3(C)

Table No 8: Substance name and their Bioactivity based on R_f Value@254 nm and 20.0 µL volume

Peak No.	R_f Value	% Area	Substance Name	Bioactivity (Source: PubChem, Google Scholar)
1.	0.029	11.37	Gelsemine	GABA Antagonist, Spinal cord synaptic Inhibitor. [PubChem]
5.	0.351	4.84	Glycosides	Analgesics, Anti-Inflammatory. [11]
7.	0.463	12.49	Tetrahydroharmine	Anti-Microbial. [13]
8.	0.536	13.10	Berberine	Vasodilator (Potassium Channel Blocker), Anti-Neoplastic, Anti-Fungal, Neuroprotective (Acetylcholinterase Inhibitor), Anti-Lipidemic, Inflammatory disorders (Kappa B Kinase Inhibitor), Anti-Diabetic, Anti-Oxidant. [PubChem]
10.	0.642	4.06	Phenols	Anti-Oxidant, Anti-Cancer Activity. [12]
12.	0.711	11.35	Rescinnamine	Vasodilator and Anti-Hypertensive agent (Angiotensin-Converting enzyme) [PubChem]
13.	0.889	7.08	Phyto steroid	Inflammatory Disorders, Cardiovascular Disorders. [16]

Figure 4: HPTLC Chromatograph of Vajra Guggulu @366 nm and 20.0 µL volume (A=Fingerprint, B= Peak height, C= R_f Value and Area percentage)

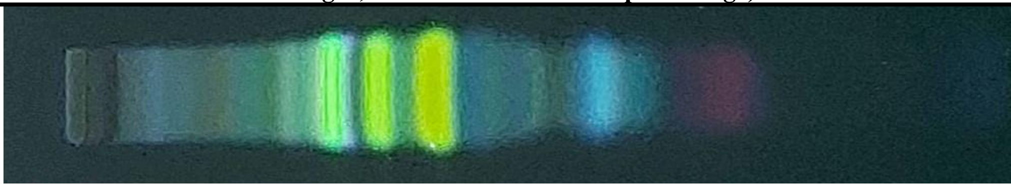


Fig 4(A)

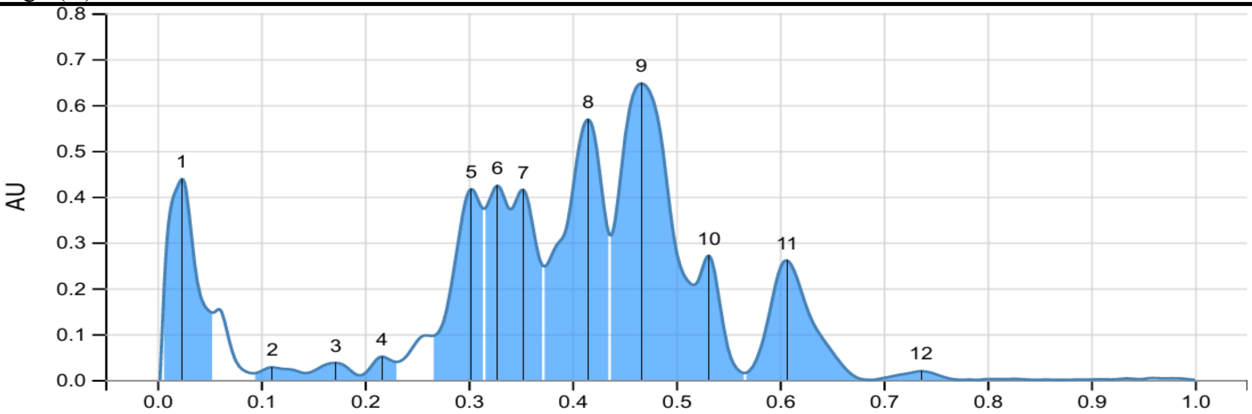


Fig 4(B)

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Peak #	Start R_F	Start H	Max R_F	Max H	Max %	End R_F	End H	Area A	Area %
1	0.003	0.0000	0.024	0.4378	12.26	0.053	0.1468	0.01426	10.58
2	0.093	0.0142	0.111	0.0274	0.77	0.143	0.0148	0.00108	0.80
3	0.143	0.0148	0.172	0.0371	1.04	0.196	0.0099	0.00136	1.01
4	0.196	0.0099	0.217	0.0507	1.42	0.231	0.0391	0.00125	0.93
5	0.265	0.0960	0.303	0.4161	11.65	0.314	0.3741	0.01317	9.77
6	0.315	0.3734	0.328	0.4236	11.86	0.340	0.3721	0.00998	7.40
7	0.340	0.3721	0.353	0.4146	11.61	0.372	0.2482	0.01136	8.43
8	0.374	0.2476	0.415	0.5679	15.90	0.436	0.3164	0.02531	18.78
9	0.438	0.3154	0.467	0.6468	18.11	0.517	0.2071	0.03630	26.94
10	0.517	0.2071	0.532	0.2706	7.57	0.565	0.0150	0.00738	5.48
11	0.567	0.0150	0.607	0.2604	7.29	0.686	0.0000	0.01251	9.28
12	0.689	0.0000	0.736	0.0191	0.54	0.775	0.0000	0.00080	0.59

Fig 4(C)

Table No 09: Substance name and their Bioactivity based on R_f Value@366 nm and 20.0 µL volume

Peak No.	R_f Value	% Area	Substance Name	Bioactivity (Source: PubChem, Google Scholar)
1	0.029	11.37	Eugenol	Neuro-Protective agent, Anti-Oxidant, Anti-Inflammatory ,Analgesics, Antiseptic Agent. [PubChem]
2	0.053	11.07	Phenol	Anti-Oxidant, Anti-Cancer Activity. [12]
3	0.258	4.45	Saponin	Anti-Inflammatory, Anti-Microbial, Anti-Ulcer, Anti- Tumour. [17]
8	0.536	13.10	Berberine	Vasodilator (Potassium Channel Blocker), Anti-Neoplastic, Anti-Fungal, Neuroprotective (Acetylcholinterase Inhibitor), Anti-Lipidemic, Inflammatory disorders (Kappa B Kinase Inhibitor), Anti-Diabetic, Anti-Oxidant. [PubChem]
9	0.596	5.97	Commiferin	Hypoglycemic, Antioxidant, Analgesic, Antithrombotic, Spasmolytic, Anti-Tumour and Antibacterial Effects [18]
11	0.711	11.35	Terpenoid	Anti-Inflammatory and Anticancer Activities. [19]
12	0.889	7.08	Phyto Steroid	Inflammatory Disorders, Cardiovascular Disorders. [16]

Discussion:

HPTLC (High-Performance Thin-Layer Chromatography) and physicochemical testing are crucial in Ayurveda medicine because they ensure quality control by confirming the authenticity, purity, and potency of herbal components. Additionally, they aid in the detection of adulteration by spotting contamination or the substitution of harmful or subpar ingredients. Physicochemical assays are also useful in monitoring for harmful heavy metals, pesticides, and microbiological contamination. Additionally, HPTLC also helps clinical trials by providing standardized products for research, ensuring consistent findings.

Such studies contribute to patient care by ensuring therapeutic efficacy with consistent, high-quality products, which leads to better treatment outcomes while additionally minimizing adverse effects by eliminating the likelihood of contamination and adulteration. It also contributes to the advancement of Ayurvedic research by providing consistent information to facilitate the development of evidence-based medicines.

Here, the high-performance liquid chromatography (HPTLC) of the methanolic extract of Vajra Guggulu showed the presence of multiple phytoconstituents in various concentrations, as illustrated in figures (1-4) and tables (7-10). The chromatogram scanned at 254 nm and 366 nm shows a variety of peaks and their obtained Rf Values (Figures 3-6). The Rf Value, Relative Rf, and peak area % were then compared to standard references to estimate the compound name, which is the primary derivative of the phytochemicals, as illustrated in tables 7-10.

According to high-performance thin-layer chromatography, the phytochemicals extracted from Vajra Guggulu have anti-inflammatory and antioxidant properties. They also include inhibitors of protein kinase C and Kappa B kinase (Table 6,7), which can be utilized to regulate inflammatory markers in the circulatory system and oxidative stress that are responsible for inflammatory illnesses. It additionally includes active components that function as analgesics, non-narcotic analgesics for skeletal muscle pain, smooth muscle relaxants and spasmolytic drugs for smooth muscle pain, and GABA agonists and spinal cord synaptic inhibitors (Table 8,9) for neuronal pain management.

The chromatogram also reveals compounds that act as Hypoglycemic agents, anti-lipidemic agents, anti-thrombolytic agents, vasodilators (potassium channel blockers), angiotensin-converting enzymes (Table 6-9), and anti-plaque agents, all of which can act as cardioprotective agents and manage peripheral vascular disorders. With respect to the compounds already described, it also exhibits the presence of compounds that function as Acetylcholinterase inhibitors, which can be used to treat neurodegenerative illnesses (Table 6-9).

The HPTLC chromatogram also displays many chemicals that demonstrate the following actions: antiulcer, anti-tumour, anti-bacterial, anti-fungal, and anti-neoplastic.

In light of the discussion that followed, we can conclude that the primary derivatives or metabolites of phytochemical elements detected by HPTLC analysis of Vajra Guggulu could be useful in the treatment of neurodegenerative disorders, peripheral neuronal disorders, peripheral vascular disorders, chronic venous insufficiency, and inflammatory disorders.

Limitations and Future Scope of the Study

The phytochemicals found in Vajra Guggulu using HPTLC with Rf Value are only primary derivatives of phytochemical components, which limits the knowledge of the drug's pharmacological effect and therapeutic potential. Analytical methods like GCMS (gas chromatography-mass spectrometry) and LCMS (liquid chromatography-mass spectrometry) can be used to determine bioactive phytochemical components in Vajra Guggulu.

CONCLUSION

This study discusses the preparation and quality of Vajra Guggulu using physicochemical evaluation as mentioned in API (Ayurvedic Pharmacopoeia of India) followed by high-performance liquid chromatography (HPTLC). The Chromatogram reveals the presence of phytochemicals capable of managing pain in skeletal muscles, smooth muscles, and neuronal pain, as well as preventing the progression of inflammatory, peripheral vascular, and neuronal disorders at various stages. The current study will also serve as a reference standard for medicine manufacturing, quality analysis, and assessing Vajra Guggulu's therapeutic potential through GC-MS pharmaceutical analysis and further pre-clinical research.

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PG-STAR SCHEME- 1.0

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Conflict of Interest

There is no conflict of interest, according to the authors.

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