Published online in http://ijam.co.in ISSN No: 0976-5921

International Journal of Ayurvedic Medicine, Vol 14 (4), 2023; 1099-1105

Analytical Study of Dashamoolakatu Treyam Kvatha Churna

Research Article

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Abstract

Aim: For the purpose of analyzing the active principle in the obtained extracts of the herbs, GC-MS is considered as an important tool. This study includes two techniques where the components is separated by Gas chromatography and the mass spectroscopy does the analysis of the active component. As a part of such an analytical aspect, *Dashamoolakayu treyam Kvatha Churna* is subjected to GC-MS to study their bioactive compounds. Methodology: 1g of sample was extracted with 10mL of methanol and was filtered through a syringe filter (Nylon 13 mm 0.2um) and injected to GCMS. The components of the oil were identified by comparison of their mass spectra with those of the spectrometer/ mass spectral database using NIST library (NIST -08 SPECTRAL DATA). The identifications were confirmed by comparison of the fragmentation pattern and their retention indices with those reported in the literature. Result: Result shows three peaks with R.T mins as 7.689, 7.964, 8.841 and the compounds are identified are α-Copaene (Tricyclo [4.4.0.02,7] dec-3-ene, 1,3-dimethyl- 8- (1-methylethyl)-, stereoisomer), Caryophyllene, Caryophyllene oxide with peak heights as 17768, 104199, 39458 respectively. Discussion: Identified components has antioxidant, Analgesic and anti-inflammatory activity and are used in the therapy of inflammatory diseases of the upper airways and also in the management of acute and chronic infectious and inflammatory diseases of different localization. Conclusion: Thus, this GC-MS study indicates the scientific validation of *Dashamoolakatu treyam Kvatha Churna* as a potent medicine as claimed by ayurvedic literature and practice.

Keywords: Gas Chromatography, Drug Analysis, Mass Spectrum, *Dashamoolakatu Treyam Kvatha Churna,* α-Copaene, Caryophyllene.

Introduction

Purity testing and assessment of natural or synthetic biologically active substance is the most prime aspect of studying the chemical analysis in pharmaceutical preparations (1). In any such type of compound, chromatographic methods are an excellent tool for the analysis of bioactive compounds present in them. Systematic assessment of the qualitative and quantitative compositions in a compound is done by both planar techniques, as well as high-performance liquid chromatography and gas chromatography. In recent years, in both routine analysis and in research centers chromatographic techniques have become more popular (2). In recent research aspects of Ayurveda, analysis always requires fast, highly efficient, and reliable methods. Low efficiency and sensitivity, and nonspecific interactions in the analyzes used in our routine clinical applications cause the reliability of these techniques to decrease. As a result, more specific analyzes should be preferred today than traditional methods (3). GC-MS method used for the analysis of the obtained extracts can be an interesting tool for testing the amount of some active principles in herbs used in cosmetics, drugs, the pharmaceutical or food industry, environmental and forensic applications (4). It combines two analytical techniques into a single method of analyzing mixtures of chemical compounds. This study includes two techniques where the components is separated by Gas chromatography and the mass spectroscopy does the analysis of the active component. As a part of such analytical techniques, *Dashamoolakayu treyam Kvatha Churna* is subjected to GC-MS to understand their bioactive compounds.

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Material and methods

Dashamoolakatu Treyam Kvatha Churna(5):
This *Kvatha Churna* (decoction powder) is mentioned in the Ayurvedic Formulary of India (6) (AFI) under *Kvatha Churna* – 4:9 whereas 4 denotes the *Kvatha Churna* reference and 9 reference for *Dashamoolakatu treyam Kvatha* (Table No:1).

Dashamoolakatu treyam Kvatha is an ayurvedic formulation used for treatment of *Shwasa* (asthma), *Kasa* (cough) caused due to *Vata dosha*, subsides the pain in *Parshva* (sides/flank), *Prishta* (back), *Trika* (sacral region) and *Murdha* (head) immediately.

The medicinal aspects of each plant constituent of *Dashamoolakatu treyam Kvatha churna* ingredients is given in Table 1,2.

Table 1: Showing the Ingredients of *Dashamoolakatu Treyam Kvatha Churna*

S. No.	Ingredients	Part used	Quantity
1	*Bilwa* (*Aegle marmelos*, Linn.)	Root/stem bark	1 part
2	*Syonaka* (*Oroxylum indicum* Linn)	Root/stem bark	1 part
3	*Gambhari* (*Gmelina arborea* Roxb)	Root/stem bark	1 part
4	*Patala* (*Stereospermum suaveolens* Roxb.)	Root/stem bark	1 part
5	*Agnimantha* (*Premna integrifolia* Linn.)	Root/stem bark	1 part
6	*Salaparni* (*Desmodium gangeticum* DC)	Plant (whole)	1 part
7	*Prsniparni* (*Uraria picta* (Jacq.))	Plant (whole)	1 part
8	*Brhati* (*Solanum indicum* Linn)	Plant (whole)	1 part
9	*Kantakari (Solanum xanthocarpum* Schradt.Wendl)	Plant (whole)	1 part
10	*Goksura* (*Tribulus terrestris*)	Plant (whole)	1 part
11	*Sunthi* (*Zingiber officinale* Roxb)	Rhizome	1 part
12	*Marica* (*Piper nigrum* Linn)	fruit	1 part
13	*Pippali* (*Piper longum* Linn.)	fruit	1 part
14	*Vasa* (*Adhathoda vasica* Nees.)	root	1 part

Table 6: Showing the medicinal aspects of each plant constituent of *Dashamoolakatu treyam Kvatha churna*

S.No.	Ingredients	Action
1	*Bilwa* (*Aegle marmelos*, Linn.)	Antihistamine activity(7), Immunomodulatory activity(8), Anti-inflammatory activity(9).
2	*Syonaka* (*Oroxylum indicum* Linn)	Anti-Allergic, Anti-asthmatic Effect(10,11)
3	*Gambhari* (*Gmelina arborea* Roxb)	Immuno-stimulant activity (12) Immuno-modulatory activity (13).
4	*Patala* (*Stereospermum suaveolens* Roxb.)	Anti-inflammatory activity (14).
5	*Agnimantha* (*Premna integrifolia* Linn.)	Antihistaminic, anti-kinin, COX-inhibitory and antioxidant action (15,16).
6	*Salaparni* (*Desmodium gangeticum* DC)	Anti-inflammatory activity (17,18,19,20)
7	*Prsniparni (Uraria picta (Jacq.))*	Anti-inflammatory activities (21,22,23,24,25,26)
8	*Brhati* (*Solanum indicum* Linn)	Anti-histamine and mast cell stabilizing efficiency (27,28), anti-asthmatic potentiality (29), bronchodilator effect (30).
9	*Kantakari (Solanum xanthocarpum Schradt.Wendl)*	Anti-allergic properties (31), antihistaminic activity (32,33), smooth muscle relaxation (34).
10	*Goksura* (*Tribulus terrestris*)	Antihistaminic, anti-kinin(35).
11	*Sunthi* (*Zingiber officinale* Roxb)	Bronchodilator effect(36).
12	*Marica* (*Piper nigrum* Linn)	Immune-modulatory activity(37, 38).
13	*Pippali (Piper longum Linn.)*	Antiasthmatic activity (39), Immunomodulatory action (40).
14	*Vasa* (*Adhathoda vasica* Nees.)	Active compound (44) Antiasthmatic, Bronchodilator activity (41) Antitussive activity (42), Broncho-dilatory activity (43).

GCMS Introduction

The present study envisages finding the active biomolecules present in *Dashamoolakatu treyam Kvatha Churna* by GC-MS analysis and tries to understand the medicinal efficacy of this drug due to these bio molecules. It is assumed that during the processing of this drug the chemical constituent that contributed various phytochemicals must have interacted to produce the biomolecules as observed by GC MS analysis.

Plant material

All the ingredients of *Dashamoolakatu treyam Kvatha Churna* were purchased from authorized Ayurveda shop at Kanyakumari and was identified by expert botanist from Tamil Nadu Agricultural University, Coimbatore (letter dated 2/01/2022). All the ingredients were dried in shade, coarsely powdered, and stored for future reference.

Preparation of Extracts

1g of sample was extracted with 10mL of methanol and was filtered through a syringe filter (Nylon 13 mm 0.2um) and injected to GCMS.

GC-MS analysis

GC-MS analysis was performed on the Instrument Model -7890 A GC 5975C TAD Series Gas Chromatograph/Mass Selective Detector (GC/MSD) with the Triple-Axis High Energy Diode (HED) Electron Multiplier (EM) Detector. Column - DB 5MS 30 m x 0.250mm Diameter x 0.25 Micro Meter Thickness. Analysis was performed by injecting 1 µL of the sample with a split ratio of 100:1. Helium gas (99.9995%) was used as the carrier gas at a flow rate of 1 mL/min. The analysis was performed in the EI (electron impact) mode with 70 eV of ionization energy. The injector temperature was maintained at 280°C (constant). The column oven temperature program is [Table-2].

Table No:2 showing the column oven temperature

Oven	Rate °C/min	Value °C/min	Hold time
Initial		60	2
Ramp 1	25	240	5
Ramp 2	35	280	30

The components of the oil were identified by comparison of their mass spectra with those of the spectrometer/ mass spectral database using NIST library (NIST -08 SPECTRAL DATA). The identifications were confirmed by comparison of the fragmentation pattern and their retention indices with those reported in the literature.

Result of GCMS analysis in *Dashamoolakatu treyam Kvatha Churna*

Result shows three peaks with R.T mins as 7.689, 7.964, 8.841 [Table-3] [Figure-1] and the compounds are identified are α-Copaene (Tricyclo [4.4.0.02,7] dec-3-ene, 1,3-dimethyl- 8- (1-methylethyl)-, stereoisomer), Caryophyllene, Caryophyllene oxide with peak heights as 17768, 104199, 39458 respectively. The details of each component are given in table [Table-4].

Figure No:1 showing the GCMS analysis

Table 3: Showing the values in GC-MS analysis

Peak #	R.T min	First scan	Max scan	Last scan	PK TY	Peak height	Corr. area	Corr. %max.	% of total
1	7.689	532	535	538	rBV4	17768	23036	16.57%	9.127%
2	7.964	562	567	575	rBV2	104199	139020	100.00%	55.082%
3	8.841	662	669	675	rBV8	39458	90331	64.98%	35.791%

Table 4: Showing the Details of Each Component

S. No	Compound Name	Molecular Formula	Molecular weight	10 largest peaks	Synonyms
1.(a)	Copaene	C15H24	204	161 999 | 119 938 | 105 900 | 93 507 | 41 308 | 91 308 | 92 256 | 81 254 | 120 232 | 204 199 |	Tricyclo [4.4.0.02,7] dec-3-ene, 1,3-dimethyl- 8- (1-methylethyl)-, stereoisomer, α-Copaene, Tricyclo[4.4.0.02,7]dec-3-ene, 8-isopropyl-1,3-dimethyl-, (1R,2S,6S,7S,8S)-(-)- Copaen, Tricyclo[4.4.0.0(2,7)]dec-3-ene, 1,3-dimethyl-8-(1-methylethyl)-, st Aglaiene, Tricyclo[4.4.0.0(2,7)]dec-3-ene, 8-isopropyl-1,3-dimethyl-
1.(b)	Naphthalene, 1,2,3,4,4a,7-hexahydro-1,6-dimethyl-4-(1-methylethyl)-	C15H24	204	119 999 | 105 996 | 41 622 | 161 449 | 91 384 | 55 312 | 92 265 | 120 241 | 27 238 | 121 232 |	4-Isopropyl-1,6-dimethyl-1,2,3,4,4a,7-hexahydronaphthalene, 4-Isopropyl-1,6-dimethyl-1,2,3,4,4a,7-hexahydronaphthalene
2	Caryophyllene	C15H24	204	41 999 | 69 976 | 93 937 | 133 646 | 79 614 | 91 551 | 55 432 | 81 389 | 107 389 | 105 372 |	Bicyclo[7.2.0]undec-4-ene, 4,11,11-trimethyl-8-methylene-, [1R-(1R*,4E,9S*)]- L-Caryophyllene Bicyclo(7.2.0)undec-4-ene, methylene-4,11,11-trimethyl-, (E)-(1R,9S)-(-)-. Caryophyllene , α + β mixt. Methylene-4,11,11-(trimethyl)bicyclo(7.2.0)undec-4-ene 4,11,11-Trimethyl-8-methylenebicyclo[7.2.0]undec-4-ene
3	Caryophyllene oxide	C15H24O	220	43 999 | 41 927 | 79 885 | 93 661 | 91 573 | 95 420 | 69 407 | 55 393 | 67 377 | 81 373 | 5-	Oxatricyclo[8.2.0.0(4,6)-]dodecane, 4,12,12-trimethyl-9-methylene-, [1R-(1R*,4R*,6R*,10S*)]-, Caryophylene oxide, Caryophyllene epoxide, (-)-β-Caryophyllene epoxide, β-Caryophyllene oxide, Epoxycaryophyllene

Structure of Active Compounds

Figure 2: Structure of Active compound- Copaene

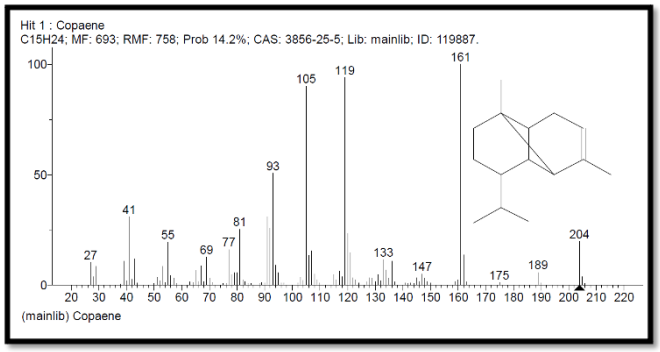


Figure 3: Structure of Active compound- Naphthalene

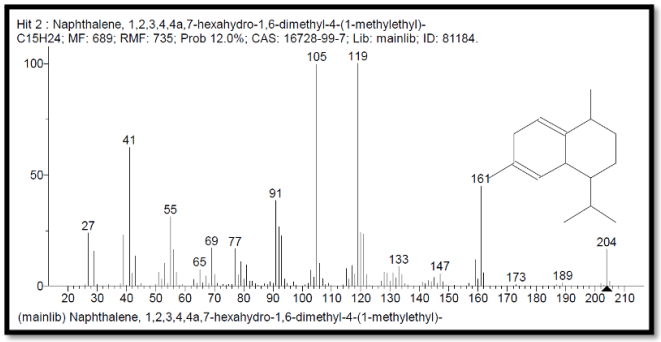


Figure 4: Structure of Active compound- Caryophyllene

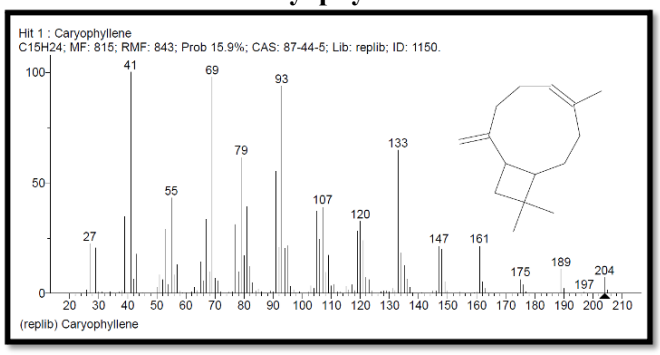
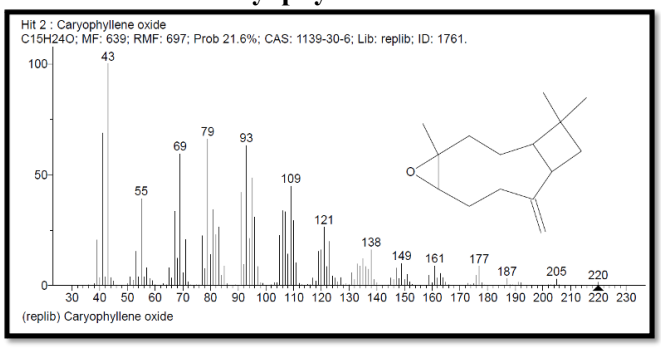


Figure 5: Structure of Active compound- Caryophyllene oxide



Discussion

Volatile components in *Dashamoolakatu treyam Kvatha Churna* are isolated through Gas Chromatography and are identified based on their mass using Mass Spectrometry. Thus, GC-MS provides improved sample identification and increased sensitivity that helps in understanding the medicinal value of given formulation (Table No:5).

Copaene (Figure 2)

Cytotoxic, genotoxic/antigenotoxic and antioxidant/oxidant activity of copaene (COP), a plant-derived tricyclic sesquiterpene, on human lymphocyte cultures was investigated and it has been reported for the first time that copaene is not genotoxic and it increases the antioxidant capacity in human lymphocyte cultures (45). Also, copaene derivatives are used in treating wide range of respiratory system infections. Valuable compounds including, α-copaene, β-element, tetradecane, E-caryophyllene (46). α-copaene of Manna gum leaves are used for the management of acute and chronic infectious and inflammatory diseases (47).

Naphthalene (Figure 3)

Naphthalene, a cytotoxic moiety, is an extensively explored aromatic conjugated system with applications in various pathophysiological conditions viz anti-inflammatory, anti-viral, anti-tubercular, anti-hypertensive, anti-diabetic, anti-neurodegenerative, antipsychotic, anticonvulsant, antidepressant. Naphthalene-containing phytoconstituents include podophyllotoxins, bis-ANS 82, Rifampicin etc(48). These major compounds have cytotoxic, antioxidant, antitumor antiangiogenic (49,50), cytotoxicity (51,52), cholesterol-lowering activity (53). 2-Naphthalene methanol, decahydro-. alpha.,. alpha., 4a-trimethyl- 8-methylene-, [2R-(2. alpha., 4a. alpha., 8a. beta.))- has a high value in the application of medicine, reduction of cough and phlegm, detoxification and diuretic and other effects.

Beta-caryophyllene (Figure 4)

BCP was found to ameliorate pulmonary inflammation in a mice model pulmonary inflammation by suppressing neutrophil accumulation, suppressing CXCL1/KC, LTB4, IL-12 and mediating the CB2R activation (54). BCP produced antispasmodic effects on the isolated tracheal smooth muscle of rats by inhibiting voltage-dependent L-type Ca2+ channels and a balance between relaxant and constrictor prostanoids exerted by BCP suggested that it may be useful in asthma-like conditions (55). Based on its therapeutic mechanisms, pharmacological potential and molecular properties of BCP, it can be considered as an effective drug in treating and in preventing the triad of infection, immunity, and inflammation (56). Other than anti-inflammatory actions, the possess analgesic, antipyretic, and platelet-inhibitory actions by inhibiting the production of prostaglandin (57). Beta caryophyllene compound are used in the management of acute and chronic infectious and inflammatory diseases of the upper airways (47).

Beta Caryophyllene and Caryophyllene Oxide (Figure 5)

Beta caryophyllene and caryophyllene oxide, isolated from *Aegle marmelos*, as the potent anti-inflammatory agents against lymphoma and neuroblastoma cells (58). Thyme (Thymus serpyllum L.) is used in the therapy of cold diseases to increase bronchial secretion and to hasten phlegm secretion; it

also has disinfecting properties. Coltsfoot (Tussilago farfara L.) leaves are used as an expectorant and emollient (47). Caryophyllene oxide has Analgesic and anti-inflammatory activity (59).

Table 5: Showing the medicinal value of each compound

Sl. No	Retention Time	peak value	Compound	Medicinal Value
1	7.689	17768	Copaene	Antioxidant capacity in human lymphocyte cultures (45), Anti-inflammatory properties (47)
2	7.964	104199	Beta Caryophyllene	Ameliorate pulmonary inflammation (54) potent Anti-inflammatory agents against lymphoma and neuroblastoma cells (58), Expectorant and emollient (47) Antispasmodic effects, acute and chronic infectious diseases, Anti-inflammatory effects (57)
3	8.841	39458	Caryophyllene oxide	Analgesic and Anti-inflammatory activity (59). potent Anti-inflammatory agents against lymphoma and neuroblastoma cells (58), Expectorant and Emollient (47)

Conclusion

GC MS analysis of the present study is shown by the retention time, peak values of the possible compounds, names of the compounds, and their reported medicinal roles. These identified compounds with the studied medicinal properties make the formulation very effective. Thus, this study indicates the scientific validation of *Dashamoolakatu treyam Kvatha Churna* as a potent medicine as claimed by ayurvedic literature and practice.

References

1. Guaadaoui, A.; Benaicha, S.; Elmajdoub, N.; Bellaoui, M.; Hamal, A. What is a bioactive compound? A combined definition for a preliminary consensus. Int. J. Food Sci. Nutr. 2014,3, 174–179.
2. Parys, Wioletta & Dołowy, Małgorzata & Pyka-Pająk, Alina. Significance of Chromatographic Techniques in Pharmaceutical Analysis. Processes. (2022). 10. 172. 10.3390/pr10010172
3. Coşkun, Ozlem & Oztopuz, Ozlem. Chromatographic Applications in Medicine. European Journal of Science and Technology. (2019). 522-529. 10.31590/ejosat.558010
4. Uma B, Prabhakar K, Rajendran S, Sarayu LY. Studies on GC/MS spectroscopic analysis of some bioactive antimicrobial compounds from Cinnamomum zeylanicum. J Med Plants. 2009; 8(31):125–131
5. Nishteswar, Vidyanath, Kvatha Preparations. Sahasrayogam, Chowkhamba Sanskrit Series Office, Varanasi, Fourth Edition 2014, p.14. PP-540
6. Anonymous, The Ayurvedic Pharmacopoeia of India Part –I. Kashaya prakana. Second edition. Government of India, Ministry of Health & Family Welfare, Department of Indian systems of medicine and homeopathy. https://dravyagunatvpm.files.wordpress.com/2009/02/afi-part-i_part_a_formulations1.pdf.Kvatha Churna 4:9. p.165, 174.
7. Nugroho AE, Riyanto S. Effects of skimmianine, a quinoline alkaloid of aegle marmelos correa roots, on the histamine release from rat mast cells. J Bas App Sci, 2010; 6: 141-148.
8. Patel P, Asdaq SM. Immunomodulatory activity of methanolic fruit extract of Aegle marmelos in experimental animals. Saudi pharm J, 2010; 18: 161-165.
9. Dinesh Kumar Sekar, A review on pharmacological and phytochemical properties of Aegle Marmelos(L.) Corr.Serr. (Rutaceae), Asian Journal of plant and research,2011, 1 (2):8-17
10. Jagetia, G.C. A Review on the Medicinal and Pharmacological Properties of Traditional Ethnomedicinal Plant Sonapatha, Oroxylum indicum. Sinusitis 2021, 5, 71–89
11. Lee, A.Y.; Kang, S.; Park, S.J.; Huang, J.; Im, D.S. Anti-allergic effect of oroxylin a from Oroxylum indicum using in vivo and in vitro experiments. Biomol. Ther. 2016, 24, 283–290
12. Punita R. Maurya, Swati R. Dhande, Yadunath M. Joshi, Vilasrao J. Kadam. A review on Gmelina arborea Roxb. Research J. Pharmacognosy and Phytochemistry 2013; 5(2): 69-76.
13. Shukla SH, Saluja AK and Pandya SS. Modulating effect of Gmelina arborea Linn. on immunosuppressed albino rats. Pharmacognosy Research. 2(6); 2010: 359-363.
14. Balasubramanian.T, Tapan Kumar Chatterjee, Mahananda Sarkar & Sundar Lal Meena Anti-inflammatory effect of *Stereospermum suaveolens* ethanol extract in rats, Pharmaceutical Biology, (2010) 48:3, 318-323.
15. Gokani RH, Lahiri SK, Santani DD, Shah MB. Evaluation of anti-inflammatory and antioxidant activity of Premna integrifolia root. [Last accessed on 2016 March 02];
16. Mali PY. Pharmacological potentials of Premna integrifolia L. Anc Sci Life. 2016 Jan-Mar;35(3):132-42. doi: 10.4103/0257-7941.179864. PMID: 27143797;
17. Rathi A, Rao ChV, Ravishankar B, De S, Mehrotra S. Anti-inflammatory and anti-nociceptive activity of the water decoction Desmodium gangeticum. J Ethnopharmacol. 2004 Dec;95(2-3):259-63. doi: 10.1016/j.jep.2004.07.009. PMID: 15507346.
18. Govindarajan R, Rastogi S, Vijayakumar M, Shirwaikar A, Rawat AK, Mehrotra S, Pushpangadan P. Studies on the antioxidant activities of Desmodium gangeticum. Biol Pharm

Bull. 2003 Oct;26(10):1424-7. doi: 10.1248/bpb.26.1424. PMID: 14519948.

19. Buddhadev, Sandip & Vaghela, Bhavesh & Shukla, Leena. Pharmacological activities of desmodium gangeticum: An overview. Pharma science monitor. (2013). 4. 264-278.
20. Subha Rastogi et al "An ethnomedicinal, phytochemical and pharmacological profile of Desmodium gangeticum (L.) DC. and Desmodium adscendens (Sw.) DC., Journal of Ethnopharmacology, Volume 136, Issue 2,2011, Pages 283-296, ISSN 0378-8741,https://doi.org/10.1016/j.jep.2011.04.031. (https://www.sciencedirect.com/science/article/pii/S0378874111002716)
21. Rahman MM, Gibbons S, Gray AI. Isoflavanones from Uraria picta and their antimicrobial activity. Phytochemistry 2007;68:1692-7
22. Saxena HO, Soni A, Mohammad N, Kakkar A, Singh N. HPLC analysis of rhoifolin in different plant parts of Uraria picta: A dashmool species. Indian J Trop Biodivers 2014;22:199-201
23. Singh NK. Anti-inflammatory and hepatoprotective activities of the roots of Uraria picta. Int J Green Pharm 2017;11:S116-73
24. Naik K, Krishnamurthy R. Anti-inflammatory activity of methanolic extracts of Pseudarthria viscida and Uraria picta against carrageenan induced paw edema in albino rat. J Med Aromat Plants 2018;9:1.
25. Olufemi AO, Omoniyi YK, Akinyinka AA, Ibitayo AM. Mechanistic evaluation of toxicity, antinociceptive, antiinflammatory and antioxidant actions of leaf extract of Uraria picta. LASU J Med Sci 2016;1:1-10.
26. Ghildiyal S, Gautam MK, Joshi VK, Goel RK. Anti-inflammatory activity of two classical formulations of Laghu Panchamula in rats. J Ayurveda Integr Med 2013;4:23-7
27. Babu NP, Pandikumar P, Ignacimuthu S. Anti-inflammatory activity of Albizia lebbeck Benth., an ethnomedicinal plant, in acute and chronic animal models of inflammation. J Ethnopharmacol, 2009;125(2):356–60.
28. Divya K, Ankit K, Tripathi JS, Tiwari SK. In-vitro and in-vivo assessment of anti-asthmatic activity of polyherbal ayurvedic drug. IOSR J Pharm Biol Sci, 2013; 6:60–70.
29. Govindan S, Viswanathan S, Vijayasekaran V, Alagappan R. A pilot study on the clinical efficacy of Solanum xanthocarpum and Solanum trilobatum in bronchial asthma. J Ethnopharmacol, 1999; 66(2):205–10.
30. Tekuri SK, Pasupuleti SK, Konidala KK, Amuru SR, Bassaiahgari P, Pabbaraju N. Phytochemical and pharmacological activities of Solanum surattense burm. f.–A review. J Appl Pharm Sci, 2019; 9(03):126–136.
31. Vadnere, Gautam Prakash and Abhay Kumar Singhai. "Evaluation of Anti-Asthmatic Property of Solanum Xanthocarpum Flower Extracts." (2008).
32. Alarcon L, Velasco J, Usubillaga A. Determinacion de la activated antibacteriana de los alcaloides presentes en los frutos verdes del *Solanum hypomalocophyllum* bitter. Rev Latinoamer Quim. (2006). 34: 13-21.
33. Chiang CT, Way TD, Tsai SJ, Lin JK. Diosgenin, a naturally occurring steroid, suppresses fatty acid synthase expression in HER2-overexpressing breast cancer cells through modulating Akt, mTOR and JNK phosphorylation. FEBS letters 2007. 581: 5735-5742.
34. Vadnere GP, Gaud RS, Singhai AK; Evaluation of Anti-Asthmatic Property of Solanum Xanthocarpum Flower Extracts. Pharmacology online, 2008; 1: 513-522.
35. Baburao B, Rajyalakshmi G, Venkatesham A, Kiran G, Shyam Sunder A, Ganga Rao B (2009) Anti-inflammatory and antimicrobial activities of methanolic extract of Tribulus terrestris Linn plant. Int J Chem Sci 16(2):209–218.
36. Jitendra H Vaghela et al., Bronchodilatory effect of Zingiber officinale Roscoe (ginger) in guinea pigs.,Tropical Journal of Pharmaceutical Research April 2020; 19 (4): 845-849 ISSN: 1596-5996 (print); 1596-9827 (electronic)
37. Ashokkumar, K., Murugan, M., Dhanya, M.K. et al. Phytochemistry and therapeutic potential of black pepper [Piper nigrum (L.)] essential oil and piperine: a review. Clin Phytosci 7, 52 (2021). https://doi.org/10.1186/s40816-021-00292-2
38. Pal Neha, Joshi M. D. Piper nigrum: An Overview of effects on Human Health. Research J. Science and Tech. 2020; 12(4):331-337. doi: 10.5958/2349-2988.2020.00050.9
39. Rajendra and Meenal, In-vivo Antiasthmatic Activity of Nava Pippali and Purana Pippali (Piper longum Linn.) Int J Ayu Pharm Chem, 2017 Greentree Group © IJAPC, 2017 Vol. 6 Issue 1 http://ijapc.com/volume6-first-issue/v6-i1-30-p-213-223.pdf (last accessed on 11/7/2023)
40. Satyapal et al., Clinical Appraisal of Immunomodulators In Ayurveda in the Light of Recent Pharmacological Advances., *World Journal of Pharmaceutical Research* ISSN 2277– 7105, Volume 4, Issue 4, 678-692.
41. Dorsch W, Wagner H. New antiasthmatic drugs from traditional medicine. Int Arch Allergy Appl Immunol 1991; 94(1-4):262-5.
42. Dhuley JN. Antitussive effect of Adhatoda vasica extract on mechanical or chemical stimulation-induced coughing in animals. J Ethnopharmacol 1999; 30; 67(3):361-5.
43. Lahiri PK, Pradhan SN. Pharmacological investigation of vasicinol, an alkaloid from Adhatoda vasica Nees. Indian Journal of Experimental Biology 1964; 2:219
44. Jain MP, Sharma VK. Phytochemical investigation of roots of Adhatoda vasica. Planta Medica 1982; 46:250.
45. Türkez H, Celik K, Toğar B. Effects of copaene, a tricyclic sesquiterpene, on human lymphocytes cells in vitro. Cytotechnology. 2014 Aug;66(4):597-603.

doi: 10.1007/s10616-013-9611-1. Epub 2013 Nov 28. PMID: 24287609; PMCID: PMC4082788.

46. Parsaeimehr, Ali & Chen, Yi-Feng & Sargsyan, Elmira. Bioactive Molecules of Herbal Extracts with Anti-Infective and Wound Healing Properties.2014. 10.1016/B978-0-12-411629-0.00012-X.
47. Kuzmenko, Natalie & Pashkova, E. & Pirogov, Andrey & Razzhivin, R. & Reshetnyak, V.. Study of a composition of officinal herb mixtures using gas-liquid chromatography with mass-spectrometric detection. Moscow University Chemistry Bulletin. (2010). 65. 106-113. 10.3103/S0027131410020100.
48. Makar S, Saha T, Singh SK. Naphthalene, a versatile platform in medicinal chemistry: Sky-high perspective. Eur J Med Chem. 2019 Jan 1;161:252-276. doi: 10.1016/j.ejmech.2018.10.018. Epub 2018 Oct 15. PMID: 30366253.
49. Robinson TP, Hubbard RB 4th, Ehlers TJ, Arbiser JL, Goldsmith DJ, Bowen JP. Synthesis and biological evaluation of aromatic enones related to curcumin. Bioorg Med Chem. *2005* Jun 2;13(12):4007-13. doi: 10.1016/j.bmc.2005.03.054. PMID: 15911313.
50. Robinson TP, Ehlers T, Hubbard IV RB, Bai X, Arbiser JL, Goldsmith DJ, Bowen JP. Design, synthesis, and biological evaluation of angiogenesis inhibitors: aromatic enone and dienone analogues of curcumin. Bioorg Med Chem Lett. 2003 Jan 6;13(1):115-7. doi: 10.1016/s0960-894x(02)00832-6. PMID: 12467629.
51. Dimmock JR, Padmanilayam MP, Zello GA, Nienaber KH, Allen TM, Santos CL, De Clercq E, Balzarini J, Manavathu EK, Stables JP, Cytotoxic Analogues of 2,6-Bis(Arylidene)Cyclohexanones. European Journal of Medicinal Chemistry, 38, 169-177.
52. Modzelewska A, Pettit C, Achanta G, Davidson NE, Huang P, Khan SR, Anticancer Activities of Novel Chalcone and Bis-Chalcone Derivatives. Bioorganic & Medicinal Chemistry, 14, 3491-3495. https://doi.org/10.1016/j.bmc.2006.01.003Bioorg. Med. Chem 2006; 14:3491-3495.
53. Piantadosi C, Hall IH, Irvine JL, Carlson GL, Cycloalkanones. 2. Synthesis and biological activity α,α′-dibenzylcycloalkanones. Journal of Medicinal Chemistry, 16, 770–795. DOI: 10.1021/jm00265a006
54. Silva, Magaiver & Correa, Luana & Candéa, André & Cavalher-Machado, Simone & Barbosa, Helene & Rosas, Elaine & Henriques, Maria. The cannabinoid 2 receptor agonist β-caryophyllene modulates the inflammatory reaction induced by Mycobacterium bovis BCG by inhibiting neutrophil migration. Inflammation research : official journal of the European Histamine Research Society (2016) 65. 10.1007/s00011-016-0969-3.
55. Pinho-da-Silva, L.; Mendes-Maia, P.V.; Teófilo, Barbosa, R.; Ceccatto, V.M.; Coelho-de-Souza, A.N.; Santos Cruz, J.; Leal-Cardoso, J.H. trans-Caryophyllene, a Natural Sesquiterpene, Causes Tracheal Smooth Muscle Relaxation through Blockade of Voltage-Dependent Ca2+ Channels. Molecules 2012, 17, 11965-11977. https://doi.org/10.3390/molecules171011965
56. Jha NK, Sharma C, Hashiesh HM, Arunachalam S, Meeran MFN, Javed H, Patil CR, Goyal SN and Ojha S., β-Caryophyllene, A Natural Dietary CB2 Receptor Selective Cannabinoid can be a Candidate to Target the Trinity of Infection, Immunity, and Inflammation in COVID-19. Front. Pharmacol. (2021) 12:590201. doi: 10.3389/fphar.2021.590201.
57. Bethesda (MD): National Library of Medicine (US), National Center for Biotechnology Information; 2004-. PubChem Compound Summary for CID 5281515, Caryophyllene; [cited 2023 July 11]. Available from: https://pubchem.ncbi.nlm.nih.gov/compound/Caryophyllene
58. Sain S, Naoghare PK, Devi SS, Daiwile A, Krishnamurthi K, Arrigo P, Chakrabarti T. Beta caryophyllene and caryophyllene oxide, isolated from Aegle marmelos, as the potent Anti-inflammatory agents against lymphoma and neuroblastoma cells. Anti-inflamm, Antiallergy Agents Med Chem. 2014 Mar;13(1): 45-55. doi: 10.2174/ 18715230113129990016. PMID: 24484210.
59. Chavan MJ, Wakte PS, Shinde DB. Analgesic and anti-inflammatory activity of Caryophyllene oxide from Annona squamosa L. bark. Phytomedicine. 2010 Feb;17(2):149-51. doi: 10.1016/j.phymed.2009.05.016. Epub 2009 Jul 2. PMID: 19576741.

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