GC-MS analysis of n-Hexane Extract of Fruits of *Trichopus zeylanicus* ssp. *travancoricus* Burkill ex K. Narayanan

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ABSTRACT

Aim: The present study focused to analyse the bioactive compounds present in the fruits of *Trichopus zeylanicus* ssp. *travancoricus* Burkill ex K. Narayanan by using GC-MS. **Methods:** The dried and pulverized fruit materials were extracted with n-hexane for 6-8 h. The phytochemical constituents were analysed by GC-MS. **Results:** Totally 23 bioactive compounds were identified with RI and SI factors. Of the twenty-three compounds detected from n-hexane extract, Tetradecane (27.87%), Hexadecane (27.26%), Nonadecane (22.25%) and Pentacosane (8.13%) were detected as major compounds. **Conclusion:** The obtained bioactive compounds were reported as potentially active in various medicinal treatments and can be used for the treatment of various diseases.

Key words: Agasthyamalai, Hexadecane, Kani tribe, Nonadecane, Tetradecane.

INTRODUCTION

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Trichopus zeylanicus ssp. travancoricus Burkill ex K. Narayanan is a rhizomatous herb belongs to the family Dioscoreaceae locally known as Arogyapachai (Tamil) and Arogyapacha (Malayalam) and literally known as the green that gives strength. In India, the species have reported as endemic to the Southern Western Ghats with a restricted distribution in Agasthyamalai Biosphere Reserve. It is reported as one of the important ethnomedicinal plants which grow near to the wet banks of streams and rivulets of the dense forests. The Kani tribe of Agasthyamalai has contributed to bring the multifarious uses of this wild plant to the present medical world. They also claim that to remain healthy, agile, young and resistant to the various diseases on infections, one should consume the fresh fruits of Aroghyapacha regularly.1 According to ethnobotanical knowledge, the unripened fruits are immersed in honey for ten days and then taken internally to get relief from asthma and the powder of leaves of T. zeylanicus ssp. travancoricus along with stem bark of Mangifera indica has taken orally to treat venereal diseases.² The powder of whole plant of Diospyros ebenum has taken along with the leaves and fruits of Trichopus zeylanicus, rhizome of Curculigo orchioides and fruits of Phyllanthus emblica, Terminalia bellirica and honey to strengthen the body and these combinations of herbals contains phytochemical constituents such as ceryl alcohol, lupeol, betulin, sitosterol, diospyric acid, triterpene and carboxylic acid.3 Arogyapachhai also possess several pharmacological activities and medicinal properties such as choleretic, aphrodisiac, hepatoprotective, mast cell stabilization.45,6 adaptogenic,7

cardioprotective,⁸ anxiolytic and antidepressant activity, hepatoprotective activity, immunomodulatory activity and anti-ulcer activity.^{9,10,11} The phytochemical studies have found that, the seeds and leaves extracts were reported with rich in saponins. The chemical investigations showed that, the leaf contains flavonoid glycosides, glycolipids and some other non-steroidal compounds.¹² The chemical investigation of aerial parts of *T. zeylanicus* ssp. *travancoricus* resulted in the isolation and characterization of five compounds namely, 6-acetyl-7-hydroxy, 8-methoxy-2,2- dimethyl-3,4-dihydro-1H-1-benzopyran, β -sitosterol, Triacontanol, apigenin-8-C glucoside (vitexin) and apigenin-6,8-di-C glucoside (Vicenin-2).¹³

Being an important ethnomedicinal herb, so far, no study has been carried out on phytochemical analysis of fruits of *T. zeylanicus* ssp. *travancoricus*. With this note, the present study was carried out to identify the bioactive compounds present in the n-hexane extract of the fruits of *T. zeylanicus* ssp. *travancoricus*.

MATERIAL AND METHODS

Plant Materials

The fruits of *T. zeylanicus* ssp. *travancoricus* were collected from Agasthyamalai Biosphere Reserve, India. The specimens collected from the forest areas were identified with the standard literature and authenticated with valid voucher specimens. The voucher specimen of T. *zeylanicus* ssp. *travancoricus* (GU 511) was submitted in the Herbarium

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Preparation of Extract

The shade dried and coarsely powdered fruit of *T. zeylanicus* ssp. *travancoricus* was extracted with n-hexane by using soxhlet apparatus for 6-8 h. The extract was filtered through Whatman No. 1 filter paper and concentrated in vacuum rotary evaporator to get a constant weight and stored at 4°C dark place prior to analysis. The n-hexane extract of the fruit was used for GC-MS analysis.

GC-MS analysis

Gas chromatography combined with mass spectroscopy is a preferable for analysis of compounds. n-hexane extract was performed using a Shimadzu GC-MS QP 2010 Ultra model and Gas Chromatograph interfaced to a mass spectrometer (GC-MS) equipped with a Rxi-5Sil MS, fused silica capillary column (30 m × 0.25 mm ID × 1× df, composed of 100% Dimethyl polysiloxane). For GC/MS detection, an electron ionization system with ionizing energy of 70 eV was used. Helium gas (99.999%) was used as the carrier gas at constant flow rate 1 ml/min and an injection volume of 1 μ l was employed (Split ratio of 50:1) injector temperature 250°C; ion-source temperature 280°C. The oven temperature was programmed from 100°C (isothermal for 2 min) with an increase of 5°C/min to 2000°C, then 10°C/min to 280°C, ending with a 2-min isothermal at 280°C. Mass spectra were taken at 70eV; a scan interval of 0.3 sec and fragments from 40 to 800 Da. Total GC running time was 32 min. Software adopted to handle mass spectra and chromatograms was a Lab Solutions.

Identification of components

The components of the extracts were identified by the calculation of their retention indices (RI) under temperature- programmed conditions for n-alkanes (C7 to C33). The identification of individual compounds was made by comparison of their relative retention time and their identity was confirmed by comparing the mass spectra with the database from the Library of National Institute of Standard and Technology 11 and Wiley8.

RESULTS AND DISCUSSION

The n-hexane extract was obtained from fruits of *T. zeylanicus* ssp. *travancoricus* through soxhlet method and the chemical composition of the extract was analysed using GC-MS. The fruits of T. *zeylanicus* ssp. *travancoricus* afforded brownish-yellow coloured semi solid mass (3.7%). Through GC-MS, a total of twenty-three compounds were detected and depicted in Table 1. All the twenty-three compounds were identified by comparison with the authentic spectra obtained from GC-MS library (NIST - 11 and WILEY 8) with SI factor. Of the twenty-three compounds identified from n-hexane extract of fruit, Tetradecane (27.87%) was found as a major compound which is an acyclic alkane hydrocarbon with 1858 structural isomers (CH₃(CH₂)₁₂CH₃). The second major compound was detected as Hexadecane (27.26%) is an alkane hydrocarbon reported from the extract of the fruit with the chemical

Table 1: GC-MS analysis of n-hexane extract of the fruits of Trichopus zeylanicus ssp. travancoricus Burkill ex K Narayanan

ex K. Narayanan.				
Rt	Name	Area (%)	RI	SI factor (%)
4.531	Dodecane	0.40	1214	95
5.252	Pentadecane, 7-methyl-	0.28	1548	91
5.360	9-methylheptadecane	0.78	1746	92
5.481	2,6-dimethyl decane	2.19	1086	92
5.786	Undecane, 3-methyl-	1.48	1150	93
6.388	Tetradecane	27.87	1413	96
8.568	Heptadecane, 2,6,10,15-tetramethyl-	0.22	1852	94
8.946	Hexacosane	0.30	2606	93
9.349	Heptadecane, 7-methyl-	0.26	1746	91
9.626	Eicosane, 10-methyl-	0.28	2045	92
9.712	Decane, 5,6-dipropyl-	0.92	1483	94
9.793	5-Methyltetradecane	1.30	1448	91
10.219	2-bromo dodecane	1.13	1446	92
10.945	Hexadecane	27.26	1712	95
13.308	Heneicosane	0.40	2109	94
14.384	Tetratetracontane	1.17	4395	91
14.505	Nonadecane, 9-methyl-	0.92	1945	92
14.954	Pentadecane, 8-hexyl-	0.82	2045	93
15.650	Nonadecane	22.25	1910	94
17.880	Hexadecane	1.18	1612	94
18.832	Tritetracontane	0.25	4295	91
20.035	Pentacosane	8.13	2506	94
23.018	Dibutyl phthalate	0.22	2037	97

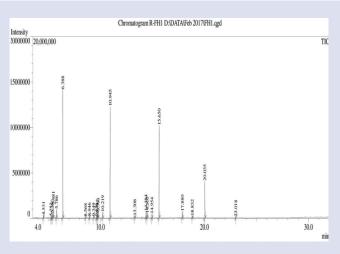


Figure 1: GC-MS Chromatogram for hexane extract obtained from dried fruits of *Trichopus zeylanicus* ssp. *travancoricus* Burkill ex K. Narayanan.

formula C₁₆H₃₄ Figure 1. Tetradecane have antimicrobial, wound healing, antiviral and anti-tumour properties whereas Hexadecane possess antibacterial and antioxidant activities.14 The third major chemical constituent was Nonadecane (CH₂(CH₂)₁₇CH₂) which comes under the saturated aliphatic hydrocarbon which occupied 22.25% of the total extract. The other compounds viz. Pentacosane (8.13%), 2,6-dimethyl decane (2.19%), Undecane, 3-methyl- (1.48%), 5-Methyltetradecane (1.30%), Hexadecane (1.18%), Tetratetracontane (1.17%), 2-bromo dodecane (1.13%), Decane, 5,6-dipropyl- (0.92%), Nonadecane, 9-methyl- (0.92%), Pentadecane, 8-hexyl- (0.82%), 9-methylheptadecane (0. 78 %), Dodecane (0.40 %), Heneicosane (0.40%), Hexacosane (0.30%), Pentadecane, 7-methyl- (0.28%), Eicosane, 10-methyl- (0.28%), Heptadecane, 7-methyl- (0.26%), Tritetracontane (0.25%), Dibutyl phthalate (0.22%) and Heptadecane, 2,6,10,15-tetramethyl - (0.22%) were found as a meagre amount in the fruit extract. Huang et al.¹⁵ stated that, n-dodecane, tetradecane, heptadecane, octadecane and dibutyl phthalate are generally considered to smell faint. The plant has the promising bioactive compounds which are helpful in the production of novel pharmaceuticals and in addition a nutraceutical. It is beneficial to further separate the compounds and determine their specific activity and to understand the synergistic impact of compounds for their therapeutic roles.

CONCLUSION

In the present investigation, twenty-three chemical constituents have been identified from n-hexane extract of the fruit of *T. zeylanicus* ssp. *travancoricus* by Gas Chromatogram Mass Spectrometry (GC-MS) analysis. The presence of various phytochemicals contributes to the various bioactive compounds have different medicinal properties which can be useful for the treatment of several diseases.

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CONFLICT OF INTEREST

The authors have no conflict of interest

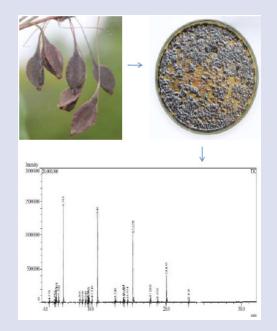
ABBREVIATION USED

GC-MS: Gas Chromatography – mass spectrometry; Rt: Retention time; RI: Retention index; SI: Super Impossibility.

REFERENCES

- Pushpangadan P, Rajasekharan S, Rathesh Kumar PK, Jawahar CR, Nair VV, Lakshmi N, et al. Arogyappacha (*Trichopus zeylanicus* Gaertn.) The ginseng of Kani tribes of Agashtyar Hills (Kerala) for Ever Green. Ancient Sci Life. 1988;8(1):13-16.
- Ayyanar M, Ignacimuthu S. Traditional knowledge of Kani tribals in Kouthalai of Tirunelveli hills, Tamil Nadu, India. J Ethnopharmacology. 2005;102(2):246-55.
- Ayyanar M, Ignacimuthu S. Ethnobotanical survey of medicinal plants commonly used by Kani tribals in Tirunelveli hills of Western Ghats, India. J Ethnopharmacology 2011;134(3):851-64.
- Subramoniam A, Madhavachandran V, Rajasekharan S, Pushpangadan P. Aphrodisiac property of *Trichopus zeylanicus* extract in male mice. J. Ethnopharmacology. 1997;57(1):21-7.
- Subramoniam A, Evans DA, Rajasekharan S, Pushpangadan P. Hepatoprotective activity of *Trichopus zeylanicus* extract against paracetamol-induced hepatic damage in rats. Indian J Experimental Biol. 1998;36(4):385-9.
- Subramoniam A, Evans DA, Rajasekharan S, Pushpangadan P. Inhibition of antigen-induced degranulation of sensitized mast cells by *Trichopus zeylanicus* in mice and rats. J. Ethnopharm.1999;68(1):137-43.
- 7. Singh B, Chandan BK, Sharma N, Singh S, Khajuria A, Gupta DK. Adaptogenic activity of *glyco-peptido-lipid* fraction from the alcoholic extract of *Trichopus zeylanicus Gaerten*. (Part II). Phytomedicine 2005;12(6):468-81.
- Velavan S, Selvarani S, Adhithan A. Cardioprotective effect of *Trichopus zeylanicus* against myocardial ischemia induced by isoproterenol in rats. Bangladesh J Pharm. 2009; 4(2):88-91.
- Rishikesh RS, Sambathkumar R, Vaibhav B. Hepatoprotective mechanism of the flavonoid fraction of *Trichopus zeylanicus Gaertn* on CCl₄ induced liver damage in rats. Inter J Med Pl. 2013;(105):269-76.
- Bachhav RS, Sambathkumar R. Evaluation of immunomodulatory activity of the alkaloid fraction of *Trichopus zeylanicus Gaertn* on experimental animals. Indian J Pharma Sci. 2016; 78(1):161-6.
- Rishikesh BS, Kumar SR, Ravindranath SB, Vaibhav BV. Anti-ulcer potential of saponin fraction of *Trichopus zeylanicus* on a various experimental animal models. Inter J Green Pharm. 2017;11(1):11-6.
- George V, Ijinu TP, Chithra MA Pushpangadan P. Can local health traditions and tribal medicines strengthen Ayurveda? Case study 2-*Trichopus zeylanicus ssp. travancoricus Burkill ex Narayanan.* J Trad Folk Practices. 2016;4:3-16.
- Chacko S, Sethuraman MG, George V, Pushpangadan P. Phytochemical constituents of *Trichopus zeylanicus ssp. travancoricus*. J Med Aromatic Plant Sci. 2002;24:703-6.
- Velmurugan G, Anand SP. GC-MS analysis of bioactive compounds on ethanolic leaf extract of *Phyllodium pulchellum L*. Desv. Inter J Pharm Phytochemical Res. 2017;9:114-8.
- Huang ZX, Jin QZ, Luo SG, Chen LH. Flavour chemistry and technology. Beijing: Chinese Light Industry Press. 1991;207-14.

GRAPHICAL ABSTRACT



SUMMARY

- Trichopus zeylanicus ssp. travancoricus is one of the ethnomedicinal herb
- By using GC-MS, totally twenty three compounds were detected from the fruits of n-hexane extract of *T. zeylanicus* ssp. *travamcoricus*
- The fruits have the potential phytochemical constituents which are useful in the production of novel pharmaceuticals

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