STUDY ON THE CHEMICAL CONSTITUENTS OF THE ESSENTIAL OIL FROM NYCTANTHES ARBOR-TRISTIS AND ITS MOLECULAR DOCKING STUDIES

KARTHICK V, VENKATAREDDY G, DHARANI J, RAVI S*
Department of Chemistry, Karpagam Academy of Higher Education, Coimbatore, Tamil Nadu, India. Email: ravisubban@rediffmail.com
Received: 01 September 2018, Revised and Accepted: 04 April 2019

ABSTRACT

Objectives: The objectives of this study were to determine the chemical composition of the essential oil obtained from the flowers of Nyctanthes arbor-tristis (NAT) and to carry out molecular docking studies against three bacterial proteins to study the mechanism of the antibacterial activity.

Methods: The essential oil was obtained from the flowers of NAT by hydrodistillation and the chemical composition was determined by gas chromatography–mass spectrometry analysis. Docking study was carried out for 14 compounds identified from NAT against three bacterial proteins 1UAG, 3TYE, and 3UDL.

Results: Fourteen compounds were identified in the essential oil. 1-octanol (74.81%) is the predominant compound followed by phytol (6.80%), bis (2-ethylhexyl) phthalate (5.88%), and eucarvone (4.23%). Many compounds are similar to that of the essential oil from jasmine. Among the 14 compounds identified, 7,9-di-tert-butyl-1-oxaspiro (4,5) deca-6,9-diene-2,8-dione interacted well with 1UAG and 3TYE and showed binding scores of −8.9 and −7.2 K Cal/mol, respectively, involving hydrophilic and hydrophobic interactions. With the protein 3UDL, the compound eucarvone exhibited a binding score of −7.1 K Cal/mol.

Conclusion: The similarities between the essential oil constituents from the flowers of the two plants NAT and jasmine were highlighted. Therefore, it could be concluded that NAT flowers of Coimbatore are a good source of fragrance for cosmetic industry and as an antibacterial agent.

Keywords: Nyctanthes arbor-tristis, Oleaceae, Essential oil, Gas chromatography–mass spectrometry, Antibacterial, Molecular docking.

INTRODUCTION

Nyctanthes arbor-tristis (NAT) Linn. is one of the well-known and most useful medicinal plant and belongs to Oleaceae. It is commonly called night jasmine in English, due to fact that its flowers emit a very strong and pleasant fragrance during whole night. NAT plant has been screened for antimalarial [1], antihistaminic, antiarthritic, local anesthetic, antihypnotic, analgesic [2], antioxidant, anti-inflammatory, antileishmaniasis, anti-cancer [4], anti-inflammatory, antitussive, antiviral [5], immunomodulatory, anthelmintic [6], antidiuretic activity, and as central nervous system modulators. NAT is said to have a wide range of medicinal benefits to humankind. The flowers of NAT are used in India, Indonesia (Java), and Malaysia to provoke menstruation while the bitter leaves are used as chologogue, laxative, diaphoretic, and diuretic (Agroforestry tree database). The iridoid glucosides from NAT and identified the increased reactive oxygen species and cellular redox homeostasis imbalance in Leishmania parasite [7], to treat loss of appetite, piles, liver disorders, chronic fever, malarial fever, obstinate redness of sciatrica, rheumatism, and as a diaphoretic [1]. NAT is also known in Indian traditional medicine to possess immune toxic, antiallergic, antihistaminic, pungent, antibacterial, and ulcerogenic activities. Conventionally, the flowers of the plant are known to be effective as stomachic, carminative, astrigent, antibilious, expectorant, and hair tonic and are used in the treatment of piles and various skin diseases. The bark is used to treat bronchitis and snakebite [8]. The present study is to identify the chemical constituents of the essential oil of the flowers of NAT Linn. and to carry out the molecular docking studies against the bacterial proteins.

MATERIALS AND METHODS

Plant material

NAT flowers were picked from the ground early in the morning before sunrise from Coimbatore District, Tamil Nadu, and taken to the laboratory for distillation. The plant was identified at the Department of Botany, Karpagam Academy of Higher Education, Coimbatore.

Hydrodistillation of flowers

Fresh flowers were hydrodistilled for 3 h using a Clevenger-type apparatus (200 g × 5 times). The obtained essential oil was collected in a test tube. From the aqueous layer, petroleum ether was used to trap the essential oil. The trapped essential oil was dried using anhydrous Na2SO4 and the essential oil was recovered and stored at 4°C.

Analysis of the essential oil using gas chromatography–mass spectrometry (GC–MS)

GC–MS analysis was performed on Agilent 5973 instrument using Restek Carbowax column 30 m 0.25 mm i.d., 0.25 µm film thickness coated with polyethylene glycol and coupled with a 5973 network mass selective detector (Agilent). Chromatographic conditions: Helium was used as carrier gas at 1.0 mL/min split less injection of 1.0 µL of oil. Injector temperature was 230°C; oven temperature program: Initial temperature 40°C (held for 5 min), rose to 220°C at 60°C/min and held for 17 min. The similarities between the essential oil constituents from the flowers of the two plants NAT and jasmine were highlighted. Compounds were identified using Wiley NIST database library. The percentages of constituents were calculated leaving out the solvent peak as well as background peaks.

Molecular docking

The molecular docking was carried out using AutoDock software which is most commonly available software used to perform the virtual screening. All the parameters used in AutoDock were selected by default. The three-dimensional crystal structure of the 1UAG (UDP-N-acetylmuramoyl-L-alanine: D-glutamate ligase [MurF]), 3UDL (Acinetobacter baumannii in complex with penicillin G), and 3TYE (dihydropteroate synthase) was retrieved from the protein database.

References

[1] K. V. Karthick, V. Venkatareddy, and S. Ravi, “Studies on the chemical constituents of the essential oil from Nyctanthes arbor-tristis and its molecular docking studies,” Asian J Pharm Clin Res, vol. 12, no. 5, pp. 29458, 2019.

© 2019 The Authors. Published by Innovare Academic Sciences Pvt Ltd. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/4.0/) DOI: http://dx.doi.org/10.22159/ajpcr.2019.v12i5.29458

Keywords: Nyctanthes arbor-tristis, Oleaceae, Essential oil, Gas chromatography–mass spectrometry, Antibacterial, Molecular docking.
| Ligands | Docking details | 1UAG | 3UDI | 3TYE |
|---------|----------------|------|------|------|
| 1-octanol | Binding score | −3.7 | −4.6 | −4.5 |
| | Conventional H-bond | SER: 415 | ARG: 482 (unfavorable-donor-donor) | ARG: 148 |
| | Alkyl and pi-alkyl | - | LYS: 73, TYR: 103 | (unfavorable-acceptor-acceptor) |
| | Others | - | ASN: 147 | |
| 1-(3,6,6-trimethyl-1,6,7,7a-tetrahydrocyclo | Binding score | −6.5 | −6.3 | −6.3 |
| | Conventional H-bond | ASN: 211 | THR: 670, LYS: 669, SER: 487 | ARG: 68,254 |
| | Alkyl and pi-alkyl | HIS: 267 | TYR: 707 | PRO: 69, LYS: 220, HIS: 256 |
| | Others | - | ARG: 482, SER: 485, VAL: 649 | PHE: 189 (pi-sigma) |
| 2,5,5,8a-tetramethyl-3,5,6,7,8,8a-hexa | Binding score | −6.0 | −6.3 | −6.5 |
| | Conventional H-bond | ASN: 138 | THR: 707 | LYS: 147, ARG: 148 |
| | Alkyl and pi-alkyl | HIS: 183, LYS: 319, ALA: 414, PHE: 422 | ARG: 482 | PHE: 71,189, TRP: 123 |
| | Others | - | - | |
| Bis (2-ethylhexyl) phthalate | Binding score | −6.4 | −6.3 | −6.4 |
| | Conventional H-bond | LYS: 115, SER: 116, ASN: 138, HIS: 183 | THR: 670,672 | LYS: 220, ARA: 68,254 |
| | Alkyl and pi-alkyl | PHE: 161, LYS: 348 | LEU: 486 | VAL: 231, PRO: 69, PHE: 189 |
| | Others | - | ARG: 485 (pi-sigma), TYR: 707 (pi-pi stacked) | HIS: 256 (carbon hydrogen bond) |
| Dibutylphthalate | Binding score | −5.1 | −6.4 | −6.4 |
| | Conventional H-bond | ASN: 138, HIS: 183 | GLY: 708,709 | ARG: 68,254, LYS: 220, HIS: 256 |
| | Alkyl and pi-alkyl | LYS: 319, PHE: 422 | TYR: 707 | PRO: 69, MET: 145 |
| | Others | - | ARG: 482 | |
| Eucarvone | Binding score | −5.0 | −7.1 | −5.4 |
| | Conventional H-bond | ARG: 302, LYS: 319 | THR: 707 | LEU: 197, MET: 200, ILE: 223, PHE: 222, LEU: 227, VAL: 226 |
| | Alkyl and pi-alkyl | PHE: 422 | LYS: 319 | |
| | Others | - | - | |
| Heneicosane | Binding score | −4.2 | −4.2 | −4.5 |
| | Conventional H-bond | - | - | |
| | Alkyl and pi-alkyl | PHE: 422,161, PRO: 72 | TYR: 707,485, LEU: 486 | |
| | Others | - | - | LEU: 197, Ala: 240, LEU: 227 |
| Hexahydrofarnesyl | Binding score | −5.1 | −5.3 | −5.4 |
| | Conventional H-bond | LYS: 319 | SER: 487, THR: 670, LYS: 669 | ARG: 68,254 |
| | Alkyl and pi-alkyl | ALA: 414, LEU: 416 | TYR: 707, LEU: 486 | LYS: 220, HIS: 256, VAL: 231, PRO: 69 |
| | Others | - | TYR: 485 (pi-sigma) | - |
| Methyl anthranilate | Binding score | −5.5 | −5.3 | −5.0 |
| | Conventional H-bond | HIS: 267, THR: 270, ASN: 271 | ASP: 648 | ARG: 68,254 |
| | Alkyl and pi-alkyl | - | ARG: 481,482 | |
| | Others | ASN: 211, ASP: 213 (carbon hydrogen bond) | TYR: 485 (pi-pi stacked) | LYS: 220 (pi-cation), PRO: 69 (pi-sigma) |
| Methyl palmitate | Binding score | −4.5 | −4.4 | −5.4 |
| | Conventional H-bond | ASN: 178 | SER: 487, LYS: 669, THR: 670,672 | ALA: 190 |
| | Alkyl and pi-alkyl | HIS: 267, ALA: 328 | TYR: 707 | LYS: 126, TRP: 123 |
| | Others | ASN: 271 (carbon hydrogen bond) | GLY: 671 (carbon hydrogen bond) | - |
| n-hexatricontane | Binding score | −4.8 | −4.7 | −4.7 |
| | Conventional H-bond | THR: 321, ARG: 302 | THR: 670,672 | ARG: 254, HIS: 256, ARG: 68 |
| | Alkyl and pi-alkyl | LYS: 328, LEU: 416 | LYS: 220, ALA: 240, LEU: 227 | PRO: 69, PHE: 71 |
| | Others | - | THR: 672 (carbon hydrogen bond) | PHE: 189 (pi-sigma) |
RESULTS AND DISCUSSION

The hydrodistillation of the flowers of NAT yielded 0.06% (w/w, wet basis) or 0.76% (w/w, dry basis) of fragrant essential oil. The essential oil obtained, had a light yellow color with a strong floral odor. The compounds present in the essential oil were identified using GC–MS analysis. The chromatogram of the essential oil is presented in Fig. 1 and the results of GC–MS analysis are summarized in Table 2. Around 14 compounds were identified in the essential oil. 1-octanol (74.81%) is the predominant compound which is a long chain primary alcohol and is used in the manufacture of perfumes and esters. This was followed by phytol (6.80%), bis (2-ethylhexyl) phthalate (5.88%), and eucarvone (4.23%). The compounds present in the essential oil from flowers of NAT such as methyl palmitate, phytol, methyl anthranilate, eucarvone, and hexahydrofarnesyl acetone were also present in jasmine oil. This strongly suggests that NAT could be used instead of jasmine for fragrant purposes. Therefore, it could be concluded that NAT flowers of Coimbatore are a good source of fragrance for cosmetic industry. This is the 1st time we are reporting that 1-octanol is the predominant compound in the essential oil of NAT. Further, the study carried out on flowers of Bangladesh reported phytol and 2-methyloctadecane as major constituents from the essential oil of the flowers [10]. Some similarities exist between the essential oil obtained from the flowers of NAT from India and Bangladesh.

Molecular Docking

Docking study was carried out for 14 bioactive compounds identified from the GC–MS analysis of NAT against three bacterial proteins. The results are presented in Table 1 and Figs. 2–4. The binding score, amino acids involved in the conventional H bond, alkylation, and pi-alkyl and other forms of interactions were presented. Among the 14 compounds identified, 7,9-di-tert-butyl-1-oxaspiro (4,5) deca-6,9-diene-2,8-dione interacted well with 1UAG and showed a binding scores of −8.9 K Cal/mol. Conventional H bonds were formed with LYS:191, ASN:322, and SER:415 and were formed with the amino acids ALA:414, LEU:416, PHE:422, and ASN:421; alkyl and pi-alkyl bonds were formed with LYS:319,348, AS 421; alkyl and pi-alkyl bonds were formed with the amino acids ALA:414, LEU:416, PHE:422, and ASN:421 of 1UAG. The same compound when docked with the protein 3TYE showed a binding affinity of −7.2 K Cal/mol and formed conventional H bonds with LYS:191, SER:221, and ASN:196. With the same protein, the compound bis (2-ethylhexyl) phthalate showed a binding score of −6.4 K Cal/mol and conventional H bonds with ARG:319,348, HIS:183, and ALA:414. With the protein 3UDI, the compound eucarvone exhibited a binding score of −7.1 K Cal/mol and showed conventional H bond with ARG:482, alkyl and pi-alkyl bonds with TYR:485, ARG:482, and VAL:649.

CONCLUSION

The essential oil was obtained from the flowers of NAT and chemical composition was determined by GC–MS analysis. 1-octanol, phytol, bis (2-ethylhexyl) phthalate, and eucarvone were found to be present in appreciable quantities. There are some similarities in the composition of the essential oil obtained from flowers India and Sri Lanka and it fairly resembles the constituents from jasmine. Therefore, it could be concluded that NAT flowers of Coimbatore are a good source of fragrance for cosmetic industry. Molecular docking studies were carried out for the identified compounds of this plant against three

---

Table 1: (Continued)

| Ligands | Phytol | 7,9-di-tert-butyl-1-oxaspiro (4,5) deca-6,9-diene-2,8-dione | Tetradecane |
|---------|--------|-------------------------------------------------|-------------|
| Docking details | 1UAG | 3TYE | 3TYE |
| Binding score | −4.9 | −4.4 | −4.9 |
| Conventional H-bond | Alkylation and pi-alkyl | Alkylation and pi-alkyl | Alkylation and pi-alkyl |
| Others | | | |
Table 2: The chemical composition of the essential oil obtained from the flowers of N. arbor-tristis

| Compound name                              | Molecular formula | Molecular weight | Retention index | Percentage |
|--------------------------------------------|-------------------|------------------|-----------------|------------|
| 1-octanol                                  | C₈H₁₈O            | 130              | 1059            | 74.81      |
| 2-hexadecen-1-ol, 3,7,11,15-tetra          | C₂₀H₄₀O           | 296              | 2045            | 6.80       |
| Bis (2-ethylhexyl) phthalate                | C₉H₁₄O₂           | 390              | 2704            | 5.88       |
| 2,4-cycloheptadione-1-one, 2,6,6-trimethyl  | C₁₀H₁₄O           | 150              | 1199            | 4.23       |
| Hexadecanoic acid methyl ester             | C₁₆H₃₂O₂          | 270              | 1878            | 2.07       |
| Benzoic acid, 2-amino-, methyl ester       | C₈H₇NO₂           | 151              | 1372            | 1.21       |
| 2,5,5,8a-tetramethyl-3,5,6,7,8a-hexa        | C₁₂H₁₆O           | 206              | 1552            | 0.92       |
| 2-pentadecane, 6,10,14-trimethyl-          | C₁₄HaO₂           | 268              | 1754            | 0.75       |
| Tetradecane                                | C₁₄HaO₂           | 198              | 1413            | 0.68       |
| 1-(3,6,6-trimethyl-1,6,7,7a-tetrahydrocyclo| C₁₃H₂₈O₂          | 206              | 1484            | 0.62       |
| Heneicosane                                | C₂₁H₄₄O           | 296              | 2109            | 0.51       |
| n-hexatriacontane                          | C₁₇H₃₄             | 506              | 3600            | 0.42       |
| 7,9-di-tert-butyl-1-oxaspiro (4,5) deca-6,9-d| C₁₃H₂₈O₃          | 276              | 2081            | 0.33       |
| Dibutyl phthalate                          | C₁₆H₃₂O₄          | 278              | 2037            | 0.32       |

N. arbor-tristis: Nyctanthes arbor-tristis

Fig. 1: The gas chromatography–mass spectrometry chromatogram of the essential oil obtained from the flowers of Nyctanthes arbor-tristis

Fig. 2: 7,9-di-tert-butyl-1-oxaspiro (4,5) deca-6,9-diene-2,8-dione with 1UAG

antibacterial target proteins 1UAG, 3UDI, and 3TYE. The compound 7,9-di-tert-butyl-1-oxaspiro (4,5) deca-6,9-diene-2,8-diones showed very good docking score with 1UAG and 3TYE proteins involving hydrophilic and hydrophobic interactions.

AUTHORS’ CONTRIBUTIONS

V. Karthick and G. Venkatareddy have carried out the work and prepared the manuscript. Dharani J helped in the docking analysis and S. Ravi has guided and done modification and editing of the manuscript.
CONFLICTS OF INTEREST
The authors declared that they have no conflicts of interest.

REFERENCES
1. Sah AK, Verma VK. Phytochemical and pharmacological potential of Nyctanthes arbor-tristis: A comprehensive review. Int J Res Pharm Biomed Sci 2012;3:420-7.
2. Nirmal SA, Pal SC, Mandal SC, Patil AN. Analgesic and anti-inflammatory activity of β-sitosterol isolated from Nyctanthes arbor-tristis leaves. Inflammopharmacology 2012;20:219-24.
3. Rani C, Chawala S, Mangat M, Mangal AK, Kajla S, Dhawan AK. Nyctanthes arbor-tristis Linn (Night Jasmine): A scared ornamental plant with immense medicinal potentials. Ind J Traditional Knowledge 2012;11:427-35.
4. Kumari TD, Madhuri TD, Charya MA, Rao K. Antioxidant and anticancer activities of Nyctanthes arbor-tristis. Int J Pharm Pharm Sci 2012;4:452-4.
5. Vyas S, Kachhwa S, Kothari SL. Comparative analysis of the in vitro antioxidant activity and polyphenolic content of successive extracts of Nyctanthes arbor-tristis Linn. Int J Pharm Pharm Sci 2014;6:373-6.
6. Sandhar HK, Kaur M, Kumar B, Prasher S. An update on Nyctanthes arbor-tristis Linn. Int Pharm Sci 2011;1:77-86.
7. Kumari TD, Charya MA. Docking studies on bioactive compounds of Nyctanthes arbor-tristis. Int J Pharm Pharm Sci 2016;8:361-5.
8. Agrawal J, Pal A. Nyctanthes arbor-tristis Linn a critical ethnopharmacological review. J Ethnopharmacol 2013;146:645-58.
9. Goodsell DS, Lauble H, Stout CD, Olson AJ. Automated docking in crystallography: Analysis of the substrates of aconitase. Proteins 1993;17:1-10.
10. Rahman MM, Roy SK, Hussain M, Shahjahan M. Chemical constituents of essential oil of petals and corolla tubes of Nyctanthes arbor-tristis flower. J Essent Oil Bearing Plants 2011;14:717-21.