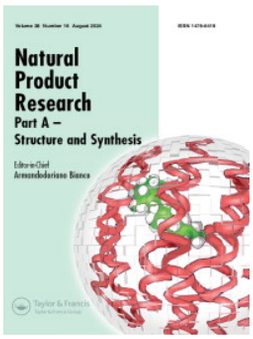


Natural Product Research

Formerly Natural Product Letters

ISSN: (Print) (Online) Journal homepage: www.tandfonline.com/journals/gnpl20

Anti-inflammatory and in silico docking studies of *Litsea wightiana* (Nees) Hook.f. (Lauraceae) leaf constituents

Kalladi Ummu Habeeba & Avanoor Ramanathan Rasmi

To cite this article: Kalladi Ummu Habeeba & Avanoor Ramanathan Rasmi (02 Aug 2024): Anti-inflammatory and in silico docking studies of *Litsea wightiana* (Nees) Hook.f. (Lauraceae) leaf constituents, Natural Product Research, DOI: 10.1080/14786419.2024.2385023

To link to this article: https://doi.org/10.1080/14786419.2024.2385023

NATURAL PRODUCT RESEARCH
https://doi.org/10.1080/14786419.2024.2385023

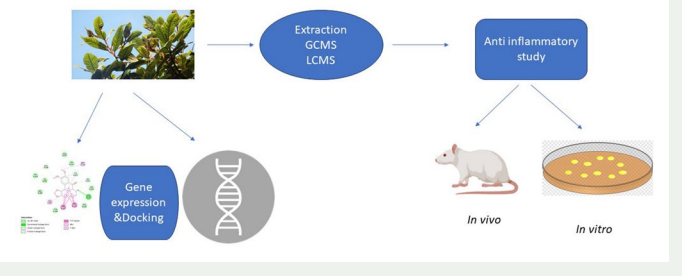
Anti-inflammatory and in silico docking studies of *Litsea wightiana* (Nees) Hook.f. (Lauraceae) leaf constituents

Kalladi Ummu Habeeba[a] and Avanoor Ramanathan Rasmi[b]

[a]Department of Botany, MES Kalladi College, Mannarkkad, Kerala, India; [b]PG & Research Department of Botany, Government Victoria College, University of Calicut, Palakkad, Kerala, India

ABSTRACT

Current study aimed to disclose the anti-inflammatory potential of the methanolic leaf extracts of *L. wightiana* (LWME). The *in vitro* studies focused on enzyme inhibition assays targeting the key enzymes such as cyclooxygenase, lipoxygenase and nitric oxide synthase and revealed that LWME effectively inhibited the activity of these enzymes. Gene expression studies confirmed the anti-inflammatory effect, demonstrating down regulation of genes associated with inflammation and key proinflammatory factors such as COX-2, TNF-α, IL-6 and NFkB. *In vivo* anti-inflammatory experiments by carrageenan-induced paw edoema method in model animals and inflammation was found to be reduced by 10% concentration of extract and significant at P<0.001 level. GCMS and LCMS analysis were conducted and the resulted compounds were docked against target proteins indicated that most of the bioactive compounds showed better binding affinity with enzymes in which the dicentrinone showed higher affinity and it may be useful in the treatment of several ailments.



ARTICLE HISTORY
Received 18 April 2024
Accepted 20 July 2024

KEYWORDS
Litsea wightiana; *in vitro* and *in vivo* anti-inflammatory; gene expression analysis; docking study

1. Introduction

The genus *Litsea*, belonging to the family Lauraceae comprised of numerous plants of significant pharmacological value, predominantly found in evergreen forests of Western Ghats. *Litsea* species are known to possess around 400 phytochemicals.

CONTACT Avanoor Ramanathan Rasmi rasmibotany@gmail.com
Supplemental data for this article can be accessed online at https://doi.org/10.1080/14786419.2024.2385023.

Crude extracts, fractions, and phytochemical constituents derived from *Litsea* have demonstrated various pharmacological attributes including anticancer, anti-inflammatory, antibacterial, antioxidant, antidiabetic, anti-HIV, insecticidal effects (Wang et al. 2016). Secondary metabolites like tannins, phenols, flavonoids, alkaloids and glycosides found in most *Litsea* plants attributes the pharmacological properties such as antioxidant, antibacterial, antihyperglycemic effects (Kamle et al. 2019). The current study endeavours to investigate the anti-inflammatory potential of *L. wightiana*, as there is no prior research on this plant's anti-inflammatory properties.

Inflammation is a type of biological response triggered by pathogens such as bacteria, fungi, or viruses, as well as various physical or chemical stimuli. Inflammatory signalling pathways most frequently NF-kB, MAPK and JAK-STAT pathways are induced by these stimuli (Chen et al. 2018). The primary cytokine to be released in response to inflammatory stimuli is tumour necrotic factor (TNF) serving as inflammatory mediator. TNF stimulates various intracellular processes, leading to the activation of NF-kB and the production of additional pro-inflammatory cytokines like IL-1 and IL-6 (Ribeiro et al. 2018). DAMP (Damage-associated molecular patterns) are produced in response to cellular stress or any damage. Metabolites including nucleic acids, glycans, proteins can acts as DAMPs and normally these molecules causes any harm to the cells. Under physical or chemical stimulus these molecules can trigger immune response and induce inflammatory diseases (Ma et al. 2024). It is also found that DAMPs can associate with Pattern recognition receptors (PRRs) and activates innate immune system and lead to chronic illness like cancer, Parkinson's, Alzheimer's etc. Hence DAMPs can be utilised as biomarkers and molecular targets in the inflammatory diseases (Roh and Sohn 2018). DAMPs associated inflammation is called sterile inflammation as it is a pathogen free type and progress to ageing. DAMPs can activate major immune effectors such as TLRs (Toll like receptors), NOD like receptors and pyrin domain containing inflammasomes and play a major role in inflammation (Feldman et al. 2015).

To mitigate the adverse effects of inflammation in the body, anti-inflammatory drugs are commonly used. However, many drugs on the market are associated with mild or significant side effects. Prolonged use of NSAIDs has been linked to adverse effects, including gastrointestinal bleeding, allergic responses, kidney problems, and heart issues (Sridevi et al. 2015). Pharmaceuticals and natural anti-inflammatory medications can effectively treat inflammation without causing side effects. An effort has been made in this area to investigate the anti-inflammatory properties of *L. wightiana* leaves. This exploration involved *in vitro* experiments utilising enzyme inhibition assays and *in vivo* studies employed by carrageenan induced paw edoema in model animals. To confirm the specific interaction of phytocompounds with target proteins, gene expression studies of pro-inflammatory cytokines and transcription factors, along with docking studies of these enzymes were conducted. This marks the primary endeavour to investigate the anti-inflammatory effect of *L.wightiana* leaves through comprehensive approach encompassing *in vitro* and *in vivo* studies, as well as molecular docking analysis of target proteins.

2. Results and discussions

2.1. In vitro anti-inflammatory assay

The anti-inflammatory property of *L.wightiana* was assessed by examining the activity of cyclooxygenase, lipoxygenase, and nitric oxide synthase. The studies demonstrated that a dose-dependent inhibition of these enzymes against standard diclofenac sodium as illustrated in (Figure S1). Additionally a decrease in the concentration of cellular nitrite levels substantiated the anti-inflammatory potential of *L.wightiana* leaf extracts. The *in vitro* anti-inflammatory studies on L. *wightiana* leaf extracts revealed significant activity, as evidenced by the inhibition of enzymes such as cyclooxygenase (IC_{50} 19.42 ± 0.08 µg/ml), lipoxygenase (16.21 ± 1.2 µg/ml) and nitric oxide synthase (36.56 ± 0.012 µg/ml). The observed IC_{50} values indicate the potency of the leaf extract in mitigating the inflammatory processes. Thus, it is considered as blocking these enzymes may aid in the treatment of diseases associated with oxidative stress and inflammation. Comparatively the leaf extract of *Litsea quinqueflora* enriched with polyphenols including gallocatechin, sinapic acid, pincocembrin, paenol and umbelliferrin, demonstrated robust anti-inflammatory action with an IC_{50} value of 23.59 g/ml (Jose and Anilkumar 2021). Similarly, anti-inflammatory studies conducted on *Psidium guajava* and *Syzygium cumini* leaf extracts confirmed the attenuation of COX-5, LOX, iNOS, and cellular nitrate levels. (Deepa et al. 2017).

2.2. Gene expression studies

The results indicated that all the cytokines demonstrated a downhill regulation of genes in LPS induced sample RAW264.7 cells as illustrated in (Figure S2). TNF-α, IL-8, IL-6, IL-1, and COX-2 are recognised as pro-inflammatory cytokines that play a crucial role in chronic inflammatory disorders (Kim et al. 2007). The transcription factor NF-kB was found to significantly contribute to intracellular signalling pathways associated with cytokine production (Ghosh et al. 1998). A study involving glycosides of dihydrostilbene isolated from *Litsea coreana* leaves revealed mild anti-inflammatory action by inhibiting TNF- and IL-1 production in RAW264.7 cells stimulated with lipopolysaccharides (LPS) (Tang et al. 2013). Furthermore, gene expression studies conducted on *L.cubeba* root extracts demonstrated the suppression of mRNA of iNOS, COX-2 in LPS-induced RAW 264.7 cells, with IC_{50} for TNF-α inhibition measured at 28.2 ± 0.9 and 15.0 ± 1.0 µM respectively (Lin et al. 2016).

2.3. In vivo anti-inflammatory activity

The *L.wightiana* methanolic leaf extracts showed remarkable reduction in size of paw edoema which is treated with carrageenan, an inflammatory substance. The effect of LWME was found to be significant and dose dependent. The drug treatment of 50 mg/kg resulted the progressive decrease in paw thickness viz., 3.67 ± 0.04 mm, 3 ± 0.26 mm, 2.33 ± 0.37 mm in third, fourth and fifth-hour periods respectively and proved statistically significant at $p<0.01$ level of significance. Similarly, the treatment of extract with 100 mg/kg exhibited decline in paw thickness during second hour after treatment and was significant at p-value <0.05. After fourth hour treatment the inflammation was reduced to 60.12% which is higher than the treatment with standard Indomethacin

is 58.02% (Table S1) &(Table S2). This results are comparable with the findings were reported in anti-inflammatory experiments in *Ocimum gratissimum* and *Moringa oleifera,* the reduction in paw thickness was observed in 100 mg/kg and 500 mg/kg respectively (Yadav and Shah 2014). The anti-inflammatory effect of methanolic extracts as compared to other solvents was reported to be higher and assumed to be the presence of bioactive constituents like terpenoids or flavonoids in plants (Lin et al. 2008).

2.4. GCMS analysis

The Gas chromatographic and Mass spectroscopic analysis were applied to find out the presence of volatile chemical compounds in leaf methanolic extracts of *L.wightiana*. The identification of detected compounds was accomplished by comparing them with spectral configurations in the database of the National Institute of Standards and Technology (NIST) GCMS analysis disclosed about 21 bioactive compounds. These compounds were submitted with their Retention Time (RT), Molecular weight (M.W) and peak area (%). The major compounds were found to be Oleic acid (65.16%) followed by n-Hexadecanoic acid(36.56%), 11-Octadecanoic acid methyl ester (13.22), Hexadecanoic acid methyl ester (10.49%),Naphthalene,1,2,4a,5,6,8a-hexahydro-4,7-dimethyl-1-(1-methylethyl)(1α,4aα,8aα)-(5.99%) Copaene (5.68%), Globulol (5.52%) (Table S3). These compounds were reported to exhibit antioxidant, anti-inflammatory, anti-proliferative, anti-microbial, anticancer, insecticidal, hepatoprotective, neuroprotective and cytotoxicity potentials (Keskes et al. 2017). Specifically hexadecanoic acid methyl ester demonstrates antioxidant, anti-inflammatory and cytotoxycytic properties, while10-octadecanoic acid methyl ester provides anti-bacterial and antifungal characteristics along with reducing blood cholesterol levels (Belakhdar et al. 2015; Ravi and Krishnan 2017).

2.5. LCMS analysis

The Liquid chromatographic and Mass spectroscopic profiling of leaf extracts by Quadrupole Time of Flight (Q-TOF) Mass spectroscopic method resulted a total of 16 compounds in both positive and negative ion modes were identified. The details of these compounds are outlined in (Table S4). The major compounds were found to be Chalcone followed by Xanthoplanine, Dicentrinone, Grandisin, (+)-,Pamitic acid, Eudesmine, Dehydroeuginol and Lurolistine.Chalcone and Corytuberine are reported to exhibit anti-inflammatory, antidiabetic, antioxidant, antituberculosis efficacies (Do santos 2018). Xanthoplanine, an isoquinone alkaloid, hinders the CrKL-STAT-5 complex in inflammatory process by diminishing the polarisation of macrophages. (Shi et al. 2020). Grandisin, a lignan compound possesses chemoprotective, cytotoxic, anti-angiogenic, anti-inflammatory anti-leishmanial potentials (Patel 2022). Dehydroeuginol, a neolignan compound possesses biological activities like antioxidant, anti-inflammatory, antimicrobial, and anticarcinogenic effects.

2.6. Molecular docking studies

The molecular docking method was employed to assess the anti-inflammatory potential of bioactive compounds in *L.wightiana* leaves. Twelve compounds were tested and

all the selected compounds were reported with anti-inflammatory activity (Singh Grewal et al. 2017). The overall result showed that notably, Dicentrinone an alkaloid compound identified by LCMS analysis, demonstrated favourable binding energy against all target proteins, including Human Myeloperoxidase,(-8.3), Human Inducible Nitric Oxide Synthase(-9.5),Cyclooxygenase II(-9.2),5-Lipoxygenase(-8) respectively. These findings positions dicentrinone as a promising anti-inflammatory agent. Analysis of interactions revealed that the compound forms a lone hydrogen bond interaction with 1dnu involving GLU102, and 4nos with ASN370. Additionally, it engages van der Waals and π interactions with all other target proteins. Notably, Xanthoplanine, Cubinol, Chalcone, a-cadinol and similar compounds displayed substantial affinity with the selected proteins. Conversely, molecules such as Linalool and hexadecenoic acid exhibited the least binding energy and affinity. The binding geometrics of the target proteins are illustrated in (Figures S3 and S4).

4. Conclusion

The current study explored the anti-inflammatory potential of *L.wightiana* leaves. Both *in vitro* and *in vivo* studies along with gene expression studies proved the anti-inflammatory efficacy of the leaf extract. The GCMS and LCMS analysis revealed the biologically active phytocompounds and is a major contribution in the understanding of chemical constituents in *L.wightiana*. Molecular docking of major phytochemicals against twelve target proteins were confirmed that dicentrinone, an alkaloid showed better binding affinity, suggesting its potential role as a primary anti-inflammatory agent in this plant. To ascertain the precise chemical compound responsible for the anti-inflammatory effect, further studies including compound isolation and purification processes are imperative.

Acknowledgment

The authors are thankful to Nanda College of Pharmacy,Coimbatore for the assistance in animal studies.The authors also grateful to the Principal,Govt. Victoria College, Palakkad for rendering the support in our research work.

Disclosure statement

No potential conflict of interest was reported by the author(s).

Funding

The author(s) reported there is no funding associated with the work featured in this article.

References

Adam OAO, Abadi RSM, Ayoub SMH. 2019. The effect of extraction method and solvents on yield and antioxidant activity of certain Sudanese medicinal plant extracts. J Phytopharmacol. 8(5):248–252. doi:10.31254/phyto.2019.8507.

Arunachalam K, Parimelazhagan T. 2013. Anti-inflammatory, wound healing and in-vivo antioxidant properties of the leaves of Ficus amplissima Smith. J Ethnopharmacol. 145(1):139–145. doi:10.1016/j.jep.2012.10.041.

Axelrod B, Cheesbrough TM, Laakso S. 1981. Lipoxygenase from soybeans: EC 1.13. 11.12 Linoleate: oxygen oxidoreductase. In: Methods in enzymology. Academic Press. Vol. 71; p. 441–451.

Belakhdar G, Benjouad A, Abdennebi EH. 2015. Determination of some bioactive chemical constituents from Thesium humile Vahl. J Mater Environ Sci. 6(10):2778–2783.

Bensalem S, Soubhye J, Aldib I, Bournine L, Nguyen AT, Vanhaeverbeek M, Rousseau A, Boudjeltia KZ, Sarakbi A, Kauffmann JM, et al. 2014. Inhibition of myeloperoxidase activity by the alkaloids of Peganum harmala L.(Zygophyllaceae). J Ethnopharmacol. 154(2):361–369. doi:10.1016/j.jep.2014.03.070.

Biswas N, Saha S, Khanra S, Sarkar A, Prasad Mandal D, Bhattacharjee S, Chaudhuri A, Chakraborty S, Roy Choudhury C. 2019. Example of two novel thiocyanato bridged copper (II) complexes derived from substituted thiosemicarbazone ligand: structural elucidation, DNA/albumin binding, biological profile analysis, and molecular docking study. J Biomol Struct Dyn. 37(11):2801–2822. doi:10.1080/07391102.2018.1503564.

Chen L, Deng H, Cui H, Fang J, Zuo Z, Deng J, Li Y, Wang X, Zhao L. 2018. Inflammatory responses and inflammation-associated diseases in organs. Oncotarget. 9(6):7204–7218. doi:10.18632/oncotarget.23208.

Deepa J, Aleykutty NA, Jyoti H. 2017. Comparative evaluation of invitroanti-inflammatory activity of psidium guajava and syzygium cumini leaves. J Ayurveda Pharma Res. 5(10):33–41.

Feldman N, Rotter-Maskowitz A, Okun E. 2015. DAMPs as mediators of sterile inflammation in aging-related pathologies. Ageing Res Rev. 24(Pt A):29–39. doi:10.1016/j.arr.2015.01.003.

Ghosh S, May MJ, Kopp EB. 1998. NF-κB and Rel proteins: evolutionarily conserved mediators of immune responses. Annu Rev Immunol. 16(1):225–260. doi:10.1146/annurev.immunol.16.1.225.

Jose SM, Anilkumar M. 2021. LCMS/MS analysis and evaluation of anti-inflammatory and antioxidant activities of the polyphenol fraction of *Litsea quinqueflora* (Dennst.) Suresh. Plant Sci Today. 8(4):865–872. doi:10.14719/pst.2021.8.4.1243.

Kamle M, Mahato DK, Lee KE, Bajpai VK, Gajurel PR, Gu KS, Kumar P. 2019. Ethnopharmacological properties and medicinal uses of Litsea cubeba. Plants (Basel). 8(6):150. doi:10.3390/plants8060150.

Keskes H, Belhadj S, Jlail L, El Feki A, Damak M, Sayadi S, Allouche N. 2017. LC-MS–MS and GC-MS analyses of biologically active extracts and fractions from Tunisian Juniperus phoenice leaves. Pharm Biol. 55(1):88–95. doi:10.1080/13880209.2016.1230139.

Kim YS, Ahn Y, Hong MH, Joo SY, Kim KH, Sohn IS, Park HW, Hong YJ, Kim JH, Kim W, et al. 2007. Curcumin attenuates inflammatory responses of TNF-α-stimulated human endothelial cells. J Cardiovasc Pharmacol. 50(1):41–49. doi:10.1097/FJC.0b013e31805559b9.

Lin B, Sun LN, Xin HL, Nian H, Song HT, Jiang YP, Wei ZQ, Qin LP, Han T. 2016. Anti-inflammatory constituents from the root of Litsea cubeba in LPS-induced RAW 264.7 macrophages. Pharm Biol. 54(9):1741–1747. doi:10.3109/13880209.2015.1126619.

Lin CT, Chen CJ, Lin TY, Tung JC, Wang SY. 2008. Anti-inflammation activity of fruit essential oil from Cinnamomum insularimontanum Hayata. Bioresour Technol. 99(18):8783–8787. doi:10.1016/j.biortech.2008.04.041.

Ma M, Jiang W, Zhou R. 2024. DAMPs and DAMP-sensing receptors in inflammation and diseases. Immunity. 57(4):752–771. doi:10.1016/j.immuni.2024.03.002.

Meenambiga SS, Rajagopal K, Durga R. 2015. In silico docking studies on the components of inonotus sp., a medicinal mushroom against cyclooxygenase-2 enzyme. Asian J Pharm Clin Res. 8(3):142–145.

Naseri N, Kalantar K, Amirghofran Z. 2018. Anti-inflammatory activity of Echium amoenum extract on macrophages mediated by inhibition of inflammatory mediators and cytokines expression. Res Pharm Sci. 13(1):73–81. doi:10.4103/1735-5362.220970.

Patel DK. 2022. Grandisin and its therapeutic potential and pharmacological activities: a review. Pharmacol Res Mod Chin. 5:100176. doi:10.1016/j.prmcm.2022.100176.

Peng W, Shen H, Lin B, Han P, Li C, Zhang Q, Ye B, Rahman K, Xin H, Qin L, et al. 2018. Docking study and antiosteoporosis effects of a dibenzylbutane lignan isolated from *Litsea cubeba* targeting Cathepsin K and MEK1. Med Chem Res. 27(9):2062–2070. doi:10.1007/s00044-018-2215-8.

Perumal V, Khatib A, Uddin Ahmed Q, Fathamah Uzir B, Abas F, Murugesu S, Zuwairi Saiman M, Primaharinastiti R, El-Seedi H. 2021. Antioxidants profile of Momordica charantia fruit extract analyzed using LC-MS-QTOF-based metabolomics. Food Chem (Oxf). 2:100012. doi:10.1016/j.fochms.2021.100012.

Rahman I, Biswas SK, Kirkham PA. 2006. Regulation of inflammation and redox signaling by-dietarypolyphenols. Biochem Pharmacol. 72(11):1439–1452. doi:10.1016/j.bcp.2006.07.004.

Rashid TS, Sijam K, Kadir J, Saud HM, Awla HK, Zulperi D, Hata EM. 2016. Screening for active compounds in Rhus coriaria L. crude extract that inhibit the growth of Pseudomonas syringae and Ralstonia solanacearum. IJARe. 50(1):15–21. doi:10.18805/ijare.v50i1.8583.

Ravi L, Krishnan K. 2017. Research article cytotoxic potential of N-hexadecenoic acid extracted from Kigelia pinnata leaves. Asian J Cell Biol. 12:20–27. doi:10.3923/ajcb.2017.20.27.

Ribeiro VP, Arruda C, Abd El-Salam M, Bastos JK. 2018. Brazilian medicinal plants with corroborated anti-inflammatory activities: a review. Pharm Biol. 56(1):253–268. doi:10.1080/13880209.2018.1454480.

Roh JS, Sohn DH. 2018. Damage-associated molecular patterns in inflammatory diseases. Immune Netw. 18(4):e27. doi:10.4110/in.2018.18.e27.

Salter M, Duffy C, Garthwaite J, Strijbos PJ. 1996. Ex vivo measurement of brain tissue nitrite and nitrate accurately reflects nitric oxide synthase activity in vivo. J Neurochem. 66(4):1683–1690. doi:10.1046/j.1471-4159.1996.66041683.x.

Sanjeewa KKA, Nagahawatta DP, Yang H-W, Oh JY, Jayawardena TU, Jeon Y-J, De Zoysa M, Whang I, Ryu B. 2020. Octominin inhibits LPS-induced chemokine and pro-inflammatory cytokine secretion from RAW 264.7 macrophages via blocking TLRs/NF-κB signal transduction. Biomolecules. 10(4):511. doi:10.3390/biom10040511.

Shi X, Pan S, Li Y, Ma W, Wang H, Xu C, Li L. 2020. Xanthoplanine attenuates macrophage polarization towards M1 and inflammation response via disruption of CrkL-STAT5 complex. Arch Biochem Biophys. 683:108325. doi:10.1016/j.abb.2020.108325.

Singh Grewal A, Lather V, Pandita D, Dalal R. 2017. Synthesis, docking and anti-inflammatory activity of triazole amine derivatives as potential phosphodiesterase-4 inhibitors. Antiinflamm Antiallergy Agents Med Chem. 16(1):58–67. doi:10.2174/1871523016666170616115752.

Sridevi G, Sembulingam P, Sembulingam K, Srividya S, Ibrahim M. 2015. Evaluation of in vivo anti-inflammatory activity of Pergularia daemia. World J Pharm Res. 4(8):1747–1756.

Tang W, Lu W, Cao X, Zhang Y, Zhang H, Lv X, Li J. 2013. Two new dihydrostilbenoid glycosides isolated from the leaves of Litsea coreana and their anti-inflammatory activity. Nat Prod Commun. 8(4):1934578X1300800. 1934578X1300800418. doi:10.1177/1934578X1300800418.

Tiwari P, Kumar B, Kaur M, Kaur G, Kaur H. 2011. Phytochemical screening and extraction: a review. Int J Pharm Sci. 1(1):98–106.

Vasudevan M, Gunnam KK, Parle M. 2006. Antinociceptive and anti-inflammatory properties of Daucus carota seeds extract. J Health Sci. 52(5):598–606. doi:10.1248/jhs.52.598.

Walker MC, Gierse JK. 2010. In vitro assays for cyclooxygenase activity and inhibitor characterization. In: Cyclooxygenases: Methods and protocols; p.644:131–144.

Wang YS, Wen ZQ, Li BT, Zhang HB, Yang JH. 2016. Ethnobotany, phytochemistry, and pharmacology of the genus Litsea: an update. J Ethnopharmacol. 181:66–107. doi:10.1016/j.jep.2016.01.032.

Winter CA, Risley EA, Nuss GW. 1962. Carrageenin-induced edema in hind paw of the rat as an assay for antiinflammatory drugs. Proc Soc Exp Biol Med. 111(3):544–547. doi:10.3181/00379727-111-27849.

Yadav JS, Shah BN. 2014. Screening of anti-inflammatory potential of some tradition Indian medicinal plants. Pharma Science Monitor. 5(3):198–204.

Yang Y, Yu T, Jang H-J, Byeon SE, Song S-Y, Lee B-H, Rhee MH, Kim TW, Lee J, Hong S, et al. 2012. In vitro and in vivo anti-inflammatory activities of Polygonum hydropiper methanol extract. J Ethnopharmacol. 139(2):616–625. doi:10.1016/j.jep.2011.12.003.

Zhang XiaoPo ZX, Jin Yan JY, Wu YouNan WY, Zhang CaiYun ZC, Jin DeJun JD, Zheng QingXia ZQ, Li You Bin LY. 2018. Anti-hyperglycemic and anti-hyperlipidemia effects of the alkaloid-rich extract from barks of Litsea glutinosa in ob/ob mice. Sci Rep. 8(1):12646. doi:10.1038/s41598-018-30823-w.