Acta Brasiliensis 7(3): 80-84, 2023 *Original Article*
ISSN 2526-4338 http://dx.doi.org/10.22571/2526-4338666

In vitro evaluation of the antioxidant potential of derivatives eugenol via ABTS radical capture assay

Josefa Aqueline da Cunha Lima [a], Jadson de Farias Silva [a], Macilio Martins de Moraes [a], Carolina Alves de Araujo [a], Claudio Augusto Gomes [a], Juliano Carlo Rufino Freitas [a,b*]

[a] *Programa de Pós-Graduação em Química, Universidade Federal Rural de Pernambuco, Recife, 52171-900, Pernambuco, Brasil.*
∗ *juliano.carlo@professor.ufcg.edu.br*
[b] *Universidade Federal de Campina Grande, Cuité, 58175-000, Paraíba, Brasil*

Received: September 5, 2023 / Accepted: December 17, 2023 / Published online: April 29, 2024

Abstract
Eugenol is a natural product present in different plant species, especially in the flower buds of *Syzygiu Aromatum*. This secondary metabolite exhibits antioxidant action, in addition to acting as an anti-inflammatory, antitumor, antibacterial, antifungal, antipyretic, anesthetic and analgesic agent. Given the biological potential of eugenol, the antioxidant activity of eugenol derivatives was evaluated. Thus, eugenol isolated from flower buds of *Syzygiu aromaticum* was subjected to different types of reactions such as *O*-alkylation. The eugenol derivatives were characterized by ^{1}H and ^{13}C NMR techniques. The evaluation of antioxidante activity was carried out using the ABTS radical capture assay. Six derivatives were synthesized with yields ranging from 75-92%, which exhibited low antioxidant potential compared to radical capture assays, with EC_{50} values from 0.085 to 0.327 mg/mL. These results motivate new studies, since the search for new antioxidant agents is extremely necessary in view of the numerous pathologies that are associated with oxidative stress.

Keywords: Organic synthesis; radical capture assays; antioxidant capacity.

Avaliação in vitro do potencial antioxidante de derivados de eugenol via ensaio de captura de radicais ABTS

Resumo
O eugenol é um produto natural presente em diferentes espécies vegetais, com destaque, nos botões florais de Syzygiu Aromatum. Esse metabólito secundário exibe ação antioxidante, além de atuar como anti-inflamatório, antitumoral, antibacteriano, antifúngico, antipirético, anestésico e analgésico. Dado o potencial biológico do eugenol, foi avaliada a atividade antioxidante dos derivados do eugenol. Assim, o eugenol isolado dos botões florais de Syzygiu Aromatum foi submetido a diferentes tipos de reações, como a O-alquilação. Os derivados de eugenol foram caracterizados pelas técnicas de RMN de 1H e 13C. A avaliação da atividade antioxidante foi realizada utilizando o ensaio de captura de radicais ABTS. Seis derivados foram sintetizados com rendimentos variando de 75 a 92%, os quais exibiram baixo potencial antioxidante em comparação aos ensaios de captura de radicais, com valores de CE50 de 0,085 a 0,327 mg/mL. Estes resultados motivam novos estudos, uma vez que a busca por novos agentes antioxidantes é extremamente necessária em face as inúmeras patologias que estão associadas ao estresse oxidativo.

Palavras-chave: Síntese orgânica; Ensaios de captura radical; Capacidade antioxidante.

Introduction

In recent decades, numerous studies have suggested that oxidative stress is associated with several pathological conditions involving chronic inflammation, which contributes to aging and chronic diseases, to mention cancer. Oxidative stress refers to the imbalance between the levels of reactive oxygen species (ROS) and their antioxidants. These species are generated in various processes involving biochemical and physiological activities in the form of by-products (Tan, Norhaizan, Liew, & Rahman, 2018). Phenolic compounds are examples of natural antioxidants. Reports indicate that they are more potent than vitamins E, C and carotenoids. Its action is closely related to the ability to donate hydrogen atoms, suppressing the formation of ROS, acting as metal chelators (which catalyze the Fenton reaction) or also as enzyme inhibitors/activators (Silva, Monte, Lemos, Nascimento, Costa, & Paiva, 2018). Eugenol, is a naturally occurring phenolic compound, found in the essential oil of

clove *Syzygium aromaticum* (L.) Merr. & amp; LM Perry, Myrtaceae which has widespread antioxidant action. In addition, eugenol has a range of biological activities, to mention, acts as anti-inflammatory, antitumor, antibacterial, antifungal, antipyretic, anesthetic and analgesic (Batiha, Alkazmi, Wasef, Beshbishy, Nadwa, & Rashwan, 2020). In view of the constant search for new effective therapeutic substances to prevent diseases caused by oxidative stress, combined with the potential of eugenol, the synthesis and evaluation of the *in vitro* antioxidant activity of different eugenol derivatives was carried out.

Material and Methods

Reagents and equipment

The reagents and solvents used were obtained in commercial form. The reactions were monitored by thin-layer chromatography (TLC) using silica gel plates (with fluorescent indicator F_{254}). TLC plates were visualized using UV light. The purification step was performed by liquid chromatography on a glass column using silica gel 60 (70-230 *mesh*) as the stationary phase, and the hexane and ethyl acetate solvents in different proportions as mobile phase. Spectral analyzes of the synthesized compounds were obtained using the Fourier transform infrared spectrophotometer (Spectrum 400 FT-IR / FT-NIR Spectrometer model PerkinElmer), the ^{1}H NMR and ^{13}C NMR spectra were obtained on a Varian Unity Plus spectrometer 300 and 400 MHz with the chloroform-*d* solvent and tetramethylsilane was used as an internal standard. The carbon and hydrogen contents of the compound 2c was determined by the Dynamic Flash Combustion technique, in a CHNS-O elementary analyzer, CE Instruments, model EA 1110.

Isolation procedure of the essential oil

Syzygium aromaticum flower bud (Clove) was purchased from a market in Recife, Brazil. The essential oils from clove (100 g) were separately isolated using a modified Clevenger-type apparatus and hydrodistillation for 2h. The oil layers were separated and dried over anhydrous sodium sulfate, stored in hermetically sealed glass containers, and kept at low temperature (-5ºC) until analysis. Total oil yields were expressed as percentages (g/100 g of fresh plant material). All experiments were carried out in triplicate.

Isolation of the eugenol

The essential oil obtained as described above (0.8 g) was chromatographed on a column of silica gel and eluted with a gradient of hexane / ethyl acetate (95:5, 85:15 and 60:40) to 100% dichloromethane. Twenty-five fractions each of 30 mL were collected and analysed by TLC eluting with hexane / ethyl acetate (7:3), fractions were grouped to yield five groups (G:1-5). Group G-2 was purified by chromatography using a mixture of hexane / ethyl acetate (80:20) as eluent, resulting in yellow oil 0.68 g (eugenol).

Chemical analysis of essential oils

Quantitative GC analysis were carried out using a PerkinElmer Clarus 500 GC apparatus equipped with a flame ionization detector (FID) and a non-polar DB-5 fused silica capillary column (30 m x 0.25 mm x 0.25 µm film thickness) (J & W Scientific). The oven temperature was programmed from 60 to 240 °C at a rate 3 °C.min^{-1}. Injector and detector temperatures were 260 °C. Hydrogen was used as the carrier gas at a flow rate of 1 mL.min^{-1} in split mode (1:30). The injection volume was 0.5 µL of diluted solution (1/100) of oil in *n*-hexane. The amount of each compound was calculated from GC-FID peak areas in the order of DB-5 column elution and expressed as a relative percentage of the total area of the chromatograms. Analyses were carried out in triplicate. The qualitative Gas Chromatography-Mass Spectrometry (GC-MS) analysis were carried out using a Varian 220-MS IT GC system with a mass selective detector, mass spectrometer in EI 70 eV with a scan interval of 0.5 s and fragments from 40 to 550 Da. fitted with the same column and temperature program as that for the GC-FID experiments, with the following parameters: carrier gas = helium; flow rate = 1 mL.min^{-1}; split mode (1:30); injected volume = 1 µL of diluted solution (1/100) of oil in n-hexane.

Identification and quantification

Identification of the components was based on GC-MS retention indices with reference to a homologous series of C8-C40 n-alkanes calculated using the Van der Dool and Kratz equation (Van, Dool & Kratz 1963) and by computer matching against the mass spectral library of the GC-MS data system (NIST 14 and WILEY 11th) and co-injection with authentic standards as well as other published mass spectra (Adams, 2007). Area percentages were obtained from the GC-FID response without the use of an internal standard or correction factors.

Synthesis of Eugenol ethers (2a-f): Synthesis of 4-allyl-1-(benzyloxy)-2-methoxybenzene ***2a:*** The synthesis of compound **2a** was carried out according to the methodology described by Kumar, Srinivas and Bettadaiah (2012). Colorless oil (yield 92%, 233.9 mg). ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.34 (d, J = 6.3 Hz, 2H), 3.89 (s, 3H), 5.12-5.08 (m, 2H), 5.14 (s, 2H), 5.90-6.04 (m, 1H), 6.69 (d, J = 2.4 Hz, 1H), 6.75 (d, J = 2.4 Hz, 1H), 6.82 (d, J = 7.5 Hz, 1H), 7.26-7.40 (m, 3H), 7.44-7.47 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 39.7, 55.9, 71.1, 112.4, 114.2, 115.5, 120.3, 127.2, 127.6, 128.4, 133.2, 137.3, 137.5, 146.5.

Synthesis of 3-(4-(benzyloxy)-3-methoxyphenyl)propan-1-ol ***2b:*** The synthesis of compound **2b** was carried out according to the methodology described by Kumar, Kuma, Srinivas and Bettadaiah (2015). Colorless oil (yield 90%, 184.2 mg). ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.45 (s, 1H), 1.86 (m, 2H), 2.63 (t, J = 5.7 Hz, 2H), 3.65 (t, J = 4.8 Hz, 2H), 3.87 (s, 3H), 5.11 (s, 2H), 6.65 (dd, J = 6.3 and 1.5 Hz, 1H), 6.75 (d, J = 1.5 Hz, 1H); 6.79 (d, J = 6.3 Hz, 1H), 7.25-7.30 (m, 1H), 7.33 (t, J = 5.7 Hz, 2H), 7.42 (d, J = 5.4 Hz, 2H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 31.72, 34.34, 55.98, 62.28, 71.23, 112.35, 114.30, 120.23, 127.27, 127.74, 128.49, 135.14, 137.41, 146.38, 149.60.

Synthesis of 4-allyl-1-(4-fluorophenoxy)-2-methoxybenzene ***2c:*** The synthesis of compound **2c** was carried out according to the methodology described by Chan, Monaco, Wang and Winters, (1998). White solid (yield 75%,193.7 mg). ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.30 (d, 1H, J = 6.9 Hz), 3.82 (s, 3H), 5.09-5.15 6(m, 2H), 5.92-6.06 (m, 1H), 6.72-7.00 (m, 7H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 39.37, 55.9, 113.1, 115.73, 116.03, 118.24, 118.35, 120.57, 120.95, 136.94, 137.20, 143.54, 151.10, 156.62, 159.79. Anal. C, 74.40%; H, 5.85 %. calcd for $C_{16}H_{15}FO_2$: C, 74.31%; H, 6.02%.

Synthesis of ethyl 2-(4-allyl-2-methoxyphenoxy)acetate ***2d:*** The synthesis of compound **2d** was carried out according to the methodology described by Spurg and Waldvogel (2008). Colorless oil (yield 90% 0.2252 g). ^{1}H NMR (400 MHz, $CDCl_3$) δ1.33 (t, 3H, J = 7.2 Hz), 3.38 (d, 2H, J = 6.4 Hz), 3.91 (s, 3H), 4.28 (q, 2H, J = 7.2 Hz), 4.70 (s, 2H), 5.10-5.15 (m, 2H), 5.95-6.03(m, 1H), 6.75 (dd, 1H, J = 8.4 and 2.0 Hz), 6.78 (d, 1H, J = 2.0 Hz), 6.86 (d, 1H, J = 8.4 Hz). ^{13}C NMR (100 MHz, $CDCl_3$) δ 14.0, 39.7, 55.8, 61.1, 66.7, 112.5, 114.6, 115.6, 120.3, 134.4, 137.3, 145.6, 149.5, 169.1.

Synthesis of 4-allyl-2-methoxy-1-(prop-2-yn-1-yloxy)benzene ***2e:*** The synthesis of compound **2e** was carried out according to the methodology described by Ferroni, Pepe, Kim, Lee, Guerriri, Parenti and Varchi, (2017). Colorless oil (yield 88%, 177.9 mg). ^{1}H NMR (400 MHz, $CDCl_3$) δ 2.49 (t, J = 2.4 Hz, 1H), 3.34 (d, J = 6.8 Hz, 2H), 3.86 (s, 3H), 4,73 (d, J = 2.4 Hz, 2H), 5.06-5.12 (m, 2H), 5.9-6.0 (m, 1H), 6.71-6.73 (m, 2H),6.97 (m, 1H). ^{13}C NMR (100 MHz, $CDCl_3$) δ39.7, 55.7, 56.8, 75.5,78.7, 112.2, 114.6, 115.6, 120.2, 134.1, 137.4, 145.0, 149.5.

Synthesis of 4-alil-2-metoxifenil acetato ***2f:***The synthesis of compound **2f** was carried out according to the methodology described by Arfa, Combes, Preziosi-Nelloy, Gontard and Chalier, (2006). Colorless oil (yield 90%, 185.6 mg). ^{1}H NMR (300 MHz, $CDCl_3$) δ 2.29 (s, 3H), 3.36 (d, J = 6.6 Hz, 2 H), 3.81 (s, 3H), 5.07-5.13 (m, 2 H), 5.90-6.02 (m, 1H), 6.74-6.78 (m, 2 H),6.93 (d, J = 7.5 Hz, 1 H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 20.6, 40.0, 55.7, 112.6, 116.0, 120.6, 122.4, 136.9, 137.9, 138.94, 150.8, 169.1.

Evaluation of the in vitro antioxidant activity

The evaluation of antioxidant activity was performed according to methodology which are based on the capture of ABTS radicals (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid). The ABTS free radical scavenging test was carried out according to the methodology described by Gupta, Kumar, Ganguly, Singh, Rana and Pandey, (2021) was used, in which (+)-6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox) as a positive control. Compounds and essential oil were tested at concentrations of 0.010 to 0.500 mg/mL and all tests were performed in triplicate. The results were expressed using the EC_{50} value, which represents the concentration of the sample necessary to sequester 50% of the ABTS radical.

Antiradical efficiency was established using linear regression analysis in the 95% confidence interval ($P < 0.05$) obtained by the GraphPad Prism 5.0 statistical program.

Results and Discussion

The GC-MS analysis of the essential oil of *S. aromaticum* enabled the identification of 98.70% of the chemical composition (Table 1). Eugenol (86.01%) was the major component identified in the oil. The eugenol was purified by column chromatography and used in synthesis. The eugenol derivatives (2a-f) were obtained through reactions of *O*-alkylation, oxidative hydroboration and hetero coupling of the eugenol as shown in Figure 1.

The eugenol derivatives (2a-f) were synthesized with yield values that varied from 70 to 92% (Table 2), these values being equal or higher when compared to those described in the literature. In addition, the spectroscopic data obtained from compounds 2a-f are in accordance with the values reported in the literature cited in Table 2. The new compound 2c showed 75% yield in 24 h.

The values of the effective concentrations of the compounds 2a-f are shown in Table 3. The synthesized eugenol derivatives exhibited the following order of antioxidant activity 2e >2f> 2c >2d>2a>2b. TROLOX was used as positive control, which had the following EC_{50} values 0.004 mg/mL.

Table 1. Percentage composition of essential oils from flower buds of *Syzygium aromaticum*.

Compounds	IR[a]	IR[b]	%	Method of identification
Eugenol	1354	1359	86.01	RI, MS
β-Caryophyllene	1411	1419	1.50	RI, MS, CI
α-Humulene	1446	1454	0.69	RI, MS
Eugenol acetate	1510	1522	10.50	RI, MS
Total				98.70

RI[a] = Retention indices calculated from retention times in relation to those of a series C_8-C_{40} of n-alkanes on a 30m DB-5 capillary column. RI[b] = Retention indices from the literature. RI = retention índice, MS = mass spectroscopy and CI = Co-injection with authentic compounds.

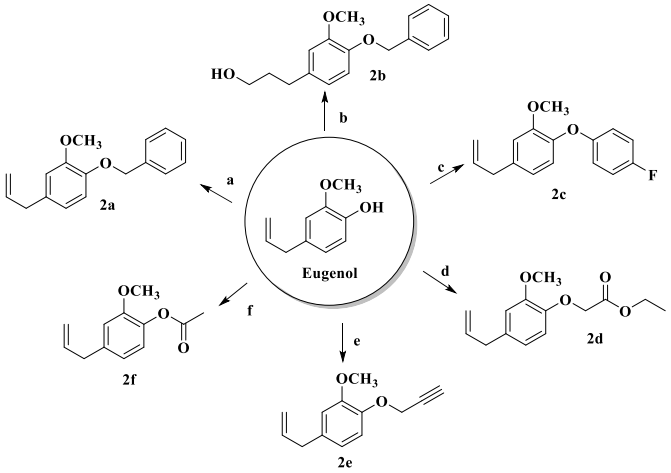

Figure 1. eugenol derivatives (2a-f) obtained through reactions of O-alkylation, oxidative hydroboration and hetero coupling of the eugenol.

Table 2. Information on the time and yield of the synthesized compounds 2a-f.

Compound	Research data		Literature data Yield (%)
	Time (h)	Yield (%)[a]	
2a	4,3	92	72% (Rahim et al., 2017)
2b	10	70	90% (Lalwani e Sudalai 2015)
2c	24	75	New product
2d	6	90	63% (Spurg e Waldvogel 2008)
2e	6	88	87% (Teixeira et al., 2018)
2f	8	90	88% (Barbosa et al., 2012)

[a]Yield of isolated product.

The EC_{50} values of the compounds 2a-f demonstrate a low antioxidant action when compared to the EC_{50} of eugenol (0.050 mg/mL). The compounds 2e and 2f had the lowest EC_{50} values from 0.085 to 0.088 mg/mL, respectively, allowing the conjecture that the incorporation of the propargyl and acetyl groups favors the capture of free radicals (ABTS) when compared to the other groups inserted in the eugenol structure (e.g. compounds 2a-d). Compound 2b showed highest EC_{50} value, allowing conjecture that the removal of the establishment of the allyl group, through oxidative hydroboration, drastically reduced the ability of this compound to capture ABTS free radicals.

Table 3. Antioxidant activity of essential oil, eugenol and derivatives (2a-f) by ABTS radical scavenging assay.

Compound	EC_{50} mg/mL
Essential oil	0.095 ± 0.005
Eugenol	0.050 ± 0.003
2a	0.206 ± 0.009
2b	0.327 ± 0.013
2c	0.128 ± 0.007
2d	0.152 ± 0.010
2e	0.085 ± 0.003
2f	0.088 ± 0.005

These results corroborate the data described in the literature, which describe that the phenolic hydroxyl and the allylic unit, present in eugenol, effectively contribute to the antioxidant activity of this natural product. Specifically, unsaturation's are considered to be very valuable for eugenol activity in relation to free radicals (Gulçin, 2011; Farias, Oliveira, Dutra, Fernandes, Pereira, Oliveira, Stefanello, Lencina and Barschaka, 2013), justifying the unsatisfactory result of compound 2b. Additionally, this study marks the first steps towards the development of new antioxidant agents, however, more studies are needed to measure the antioxidant properties of these eugenol derivatives.

Conclusion

Different eugenol derivatives were synthesized, which were obtained in good yields that varied from 75 to 92%. The evaluation of the antioxidant capacity of the eugenol derivatives provided effective concentration values in the range of 0.085 to 0.327 mg/mL, which implies a low antioxidant capacity when compared to the ABTS radical elimination test. Thus, the results obtained in this study are the first steps towards the development of new therapeutic substances that come to play an antioxidant action.

Acknowledgments

The authors are grateful to Foundation for Research Support of the State of Pernambuco (FACEPE) for funding this study (IBPG-1331-1.06/16; IBPG-0094-1.06/17 and (IBPG-0715-1.06/18) and the Foundation for Research Support of the State of Paraíba (FAPESQ, 3157/2021) for financial support.

References

Adams, R.P. (2017). Identification of Essential Oil Components by Gas Chromatography/Quadrupole Mass Spectroscopy, (4.1th ed. 809p) Allured Publishing Corporation: Carol Stream, Illinois.

Arfa, A.B., Combes, S., Preziosi-Belloy, L., Gontard, N. & Chalier, P. (2006). Antimicrobial activity of carvacrol related to its chemical structure. *Letters in Applied Microbiology*, 43(2), 149–154. doi: 10.1111/j.1472-765X.2006.01938.x

Ashok, D., Gundu, S., Amate, V.K. & Devulapally, M.G. (2018). Conventional and microwave- assisted synthesis of new indole-tethered benzimidazole-based 1,2,3-triazoles and evaluation of their antimycobacterial, antioxidant and antimicrobial activities. *Molecular Diversity*, 22(4):769-778. doi: 10.1007/s11030-018-9828-1

Barbosa, J.D., Silva, V.B., Alves, P.B., Gumina, G., Santos, R.L., Sousa, D.P. & Cavalcanti, S.C. (2012). Structure-activity relationships of eugenol derivatives against Aedes aegypti (Diptera: Culicidae) larvae. *Pest Management Science*, 68(11), 1478-1483. doi: 10.1002/ps.3331

Batiha, G.E., Alkazmi, L.M., Wasef, L.G., Beshbishy, A.M., Nadwa, E.H. & Rashwan, E.K. (2020). *Syzygium aromaticum* L. (Myrtaceae): Traditional Uses, Bioactive Chemical Constituents, Pharmacological and Toxicological Activities. *Biomolecules*, 10(2), 202. doi: 10.3390/biom10020202

Chan, D.M., Monaco K.L., Wang, R. & Winters, M.P. (1998). New N- and O-Arylations with Phenylboronic Acids and Cupric Acetate. *Tetrahedron Letters*, 39(19), 2933-2936. doi: 10.1016/S0040-4039(98)00503-6

Farias, M.D., Oliveira, P.S., Dutra, F.S.P., Fernandes, T.J., Pereira, C.M.P., Oliveira, S.Q., Stefanello, F.M., Lencina, C.L. & Barschaka, A.G. (2014). Eugenol derivatives as potential anti-oxidants: is phenolic hydroxyl necessary to obtain an effect?. Journal *Pharmacy and Pharmacology*, 66(5), 733-46. doi: 10.1111/jphp.12197

Ferroni, C., Pepe, A., Kim, Y. S., Lee, S., Guerriri, A., Parenti, M. D. & Varchi, G. (2017). 1,4-Substituted Triazoles as Non-Steroidal Antiandrogens for Prostate Cancer Treatment. *Journal of Medicinal Chemistry*, 60(7), 3082-3093. doi: 10.1021/acs.jmedchem.7b00105

Gulçin, İ. (2011). Antioxidant Activity of Eugenol: A Structure–Activity Relationship Study. *Journal of Medicinal Food*, 14(9), 975–985. doi: 10.1089/jmf.2010.0197

Gupta, A., Kumar, R., Ganguly, R., Singh, A.K., Rana, H.K. & Pandey, A.K. (2020). Antioxidant, anti-inflammatory and hepatoprotective

activities of *Terminalia bellirica* and its bioactive component ellagic acid against diclofenac induced oxidative stress and hepatotoxicity. *Toxicology Reports*, 24(8), 44-52. doi: 10.1016/j.toxrep.2020.12.010

Kumar, N.V., Kumar, S.C.S., Srinivas, P. & Bettadaiah, B.K. (2015). An Improved Route to the Preparation of 6-, 8-, 10-Gingerols. *Organic Preparations and Procedures International*, 47(6), 443-448. doi: 10.1080/00304948.2015.1088754

Kumar, N.V., Srinivas, P. & Bettadaiah, B.K. (2012). New scalable and eco-friendly synthesis of gingerols. *Tetrahedron Letters*, 53(24), 2993-2995. doi: 10.1016/j.tetlet.2012.03.092

Lalwani, K.G. & Sudalai, A. (2015). First Enantioselective Synthesis of Surinamensinol B and a Non-Natural Polysphorin Analogue by a Two-Stereocentered Hydrolytic Kinetic Resolution. *European Journal Organic Chemistry*, 2015(33), 7344–7351. https://doi.org/10.1002/ejoc.201501009

Rahim, N.H.C.A., Asari, A., Ismail, N. & Osman, H. (2016). Synthesis and Antibacterial Study of Eugenol Derivatives. *Asian Journal of Chemistry*, 29(1), 22-26. Doi: 10.14233/ajchem.2017.20100

Silva, F.F.M., Monte, F.J.Q., Lemos, T.L.G., Nascimento, P.G.G., Costa, A.K.M. & Paiva, L.M.M. (2018). Eugenol derivatives: synthesis, characterization, and evaluation of antibacterial and antioxidant activities. *Chemistry Central Journal*, 12, 34. doi: 10.1186/s13065-018-0407-4

Spurg, A. & Waldvogel, S.R. (2008). High-Yielding Cleavage of (Aryloxy)acetates. Eur *Journal of Organic Chemistry*, 2, 337–342. https://doi.org/10.1002/ejoc.200700769

Tan, B.L., Norhaizan, M.E., Liew, W.P.P. & Rahman, H.S. (2018). Antioxidant and Oxidative Stress: A Mutual Interplay in Age-Related Diseases. *Front Pharmacol*, 9, 1162. doi: 10.3389/fphar.2018.01162.

Teixeira, R.R., Gazolla, P.A.R., Da Silva, A.M., Borsodi, M.P.G., Bergmann, B.R, Ferreira, R.S., Vaz, B.G., Vasconcelos, G.A. & Lima, W.P. (2018). Synthesis and leishmanicidal activity of eugenol derivatives bearing 1,2,3-triazole functionalities. *European Journal Medicinal Chemistry*, 146, 274-286. doi: 10.1016/j.ejmech.2018.01.046

Van, D.E.N., Dool, H. & Kratz, P.D. (1963). A Generalization of the Retention Index System Including Linear Temperature Programmed Gas-Liquid Partition Chromatography. *Journal of Chromatography A*, 11, 463-471. https://doi.org/10.1016/S0021-9673(01)80947-x