International Journal of Scientific Research in Science and Technology
Available online at : www.ijsrst.com
OPEN ACCESS
Print ISSN: 2395-6011 | Online ISSN: 2395-602X
doi : https://doi.org/10.32628/IJSRST25122221

Synthesis of Novel Vanillic Acid Hybrid Derivative

Vijay D. Gangan[1*], Pradip S. Shelar[2], Sharad G. Shilkande[3], Pradnya Lokhande[4], Ragini Sinha[1], Manjiri Zimbar[1], Pranit P. Motling[1], Karuna Raddi[5]

[1]Department of Chemistry, Reena Mehta College of Arts, Commerce, Science and Management Studies, Bhayandar (W), Thane – 401101, Maharashtra, India

[2]Department of Chemistry, K. J. Somaiya college of Arts, Science and Commerce, Mohinirajnagar, Kopargaon - 423601, Dist. Ahemadnagar, Maharashtra, India

[3]Department of Chemistry, Sheth J. N. PaliwalaCollege of Arts, Science and Commerece, Pali-Sudhagad, Dist. Raigad – 410205, Maharashtra, India

[4]Department of Chemistry, Bhavan's College of Arts, Commerce and Science, Munshi Nagar, Andheri (West), Mumbai – 400058, Maharashtra, India

[5]Department of Chemistry, Patkar College of Arts, Commerce and Science, Goregaon (W), Mumbai 400062, Maharashtra, India

ARTICLEINFO

Article History:

Accepted : 13 March 2025
Published: 15 March 2025

Publication Issue :
Volume 12, Issue 2
March-April-2025

Page Number :
273-277

ABSTRACT

Vanillic acid (4-hydroxy-3-methoxybenzoic acid) is a dihydroxybenzoic acid derivative used as a flavoring agent. It is used in the synthesis of various active pharmaceutical ingredients such as Etamivan, Modecainide, Brovanexine, Vanitiolide, Vanyldisulfamide etc. In this paper, novel ester / hybrid derivative of vanillic acid was synthesized. This combinatorial synthesis of novel vanallic ester / hybrid derivative can be a useful approach to generate potent chemotherapeutic agents in developing new drug candidates.

Keywords: Vanillic acid, IR, 1HNMR, TOF MS, DCC, DMAP, antibacterial, Gram +ve and Gram -ve etc,

I. INTRODUCTION

Phenolic phytochemicals are known to exhibit anti-inflammatory, antioxidant, anticarcinogenic, antidiabetic, antiatherosclerosis and immunomodulatory activities in animals[1,2]. These are mostly polyphenols known as secondary plant metabolites[3] , present in plants and trees. Polyphenols are commonly divided into flavonoids and the hydroxyl cinnamic acids. Vanillic acid is a naturally occurring active compound having antimicrobial, anti-inflammatory and antioxidant / anticancer properties[4-9]. In continuation to our earlier work[10,11], we thought of synthesizing compound with novel ether, ester and hybrid derivative of Vanillic acid wherein Vanillic acid would be first esterified,

Copyright © 2025 The Author(s): This is an open-access article distributed under the terms of the Creative Commons Attribution **4.0 International License (CC BY-NC 4.0)**

etherified, hydrolysed and finally hybridized with various other compounds and to check whether these compounds possess above biological activities. The objective of this study is to condense two molecules of the same disease domain to produce more potent candidate in the same disease domain or to condense two molecules of different disease domain to produce mixed variety of those disease domain or to have drug candidate with entirely different biological activity.

II. MATERIALS AND METHODS

A. Materials

Chemicals used were of a laboratory grade. The reactions were monitored by TLC on aluminium-backed silica plate visualized by UV-light.

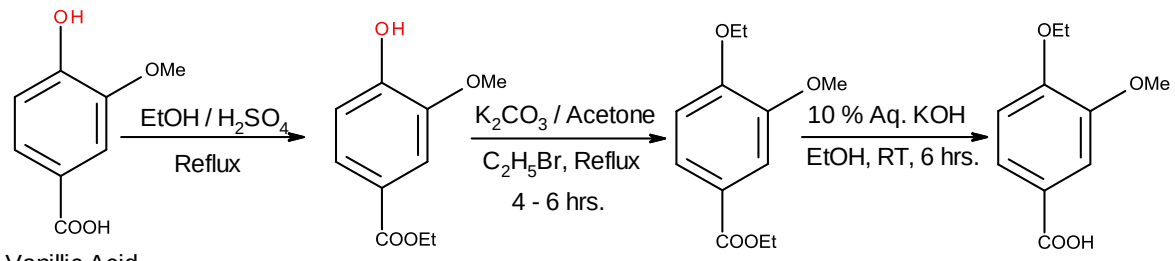
B. Experimental

Melting points were determined on a Thomas Hoover capillary melting point apparatus using digital thermometer. IR spectra were recorded on a Shimadzu FTIR Prestige model as KBr pellet. ^{1}H NMR spectra were recorded on a Varian 400 MHz spectrometer in $CDCl_3$. Chemical shifts were recorded in parts per million down field from tetramethyl silane. Mass spectra were recorded on a TOF MS ES mass spectrometer. Elemental analysis were carried out as a percentage on a Thermo finnigan, Flash EA 1112 series, Italy.

III.RESULTS AND DISCUSSION

Preparation of 3,4-dialkoxy benzoic acids :- Vanillic acid was subjected to esterification (EtOH / Conc. H_2SO_4) followed by etherification (K_2CO_3 / Acetone / C_2H_5Br) to yield crude 4-Ethoxy-3-methoxyethylbenzoate which was purified by column chromatography. This purified ether derivative was subjected to hydrolysis (Aq. KOH / EtOH and then Conc. HCl) to yield 4-Ethoxy-3-methoxybenzoic acid respectively.

Reaction Scheme 1 :



Vanillic Acid (1)

Ethyl Vanillate (2)

4-Ethoxy-3-methoxy Ethylbenzoate (3)

4-Ethoxy-3-methoxy benzoic acid (4)

Above 4-Ethoxy-3-methoxybenzoic acid was then condensed with ethyl vanillate under DCC/DMAP/Pyridine condition to yield hybrid derivative whose structure was unambiguously confirmed by IR, ^{1}H NMR, Mass spectroscopy and elemental analysis.

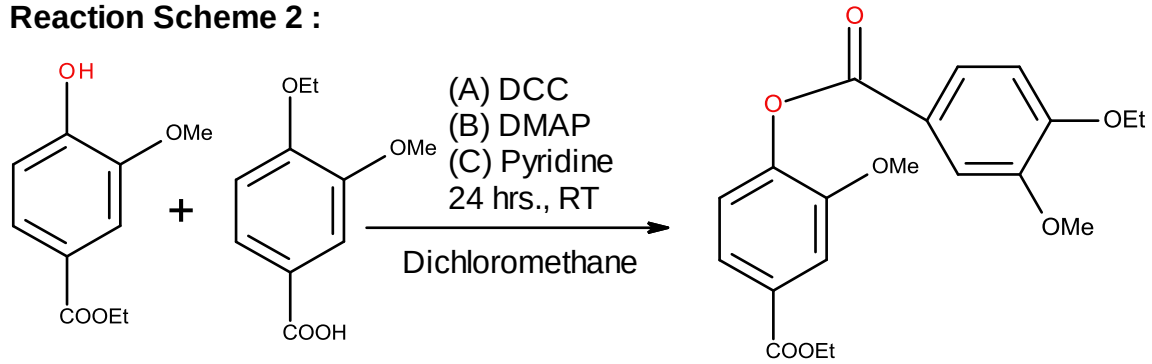
Synthesis of Hybrid Molecule using compound (2) and (4) :- It was prepared by following general method as depicted below.

To a stirred solution of ethyl vanillate [A] (1 eq.) in 30 ml dichloromethane was added DCC [A] (1.3 eq.), DMAP [B] (0.05 eq.), pyridine [C] (0.5 eq.) and the reaction mixture stirred at room temperature for 5 min. Clear solution of reaction mixture was obtained. To this, compound [B] aromatic / substituted aromatic acid (1.3 eq.) was added and stirring continued at room temperature for next 24 hrs. As the reaction proceeds, urea derivative precipitates out as by product. The progress of the reaction was monitored by TLC for completion of reaction.

Work up :-The reaction mixture filtered through celite bed which get rids of by product urea derivative. The filtrate was concentrated to minimum, preadsorbed on silica gel (100 – 200 mesh) and

purified by column chromatography with increase in concentration of ethyl acetate in petroleum ether. The general yields of these reactions ranges between 70 – 80 %. This is another method of preparing ester and follows green chemistry parameters.

Reaction Scheme 2 :



Ethyl Vanillate (2)

4-Ethoxy-3-methoxy benzoic acid (4)

(4-ethoxycarbonyl-2-methoxyphenyl) -4-ethoxy-3-methoxybenzoate

Sr. No.	2	4	Synthetic Derivative (5)
1	Ethyl vanillate	4-ethoxy-3-methoxy benzoic acid	(4-ethoxycarbonyl-2-methoxyphenyl)-4-ethoxy-3-methoxybenzoate

Compound 1 : (4-ethoxycarbonyl-2-methoxyphenyl)-4-ethoxy-3-methoxybenzoate

^{1}H NMR ($CDCl_3$, 400 MHz) δ ppm : 1.21 (t, *J* = 7.2 Hz, 3H, terminal methyl from ethyl bromide moiety), 1.41 (t, *J* = 7.0 Hz, 3H, - CH_3 from –$COOCH_2$**CH_3** group), 3.93 (s, 6H, 2 x Ar–**OCH_3** group), 4.08 (q, *J* = 6.8 Hz, 13.6 Hz, 2H, 1 x -OCH_2 from ethyl bromide moiety), 4.38 (q, *J* = 7.0 Hz, 14 Hz, 2H, -CH_2 from -COO**CH_2**CH_3 group), 6.8 – 7.9 (m, 6H, ArH); TOF MS ES: 397 (M + Na); IR (KBr) cm-1:- 2929, 2854, 2873 (methyl, methylenes, methines), 1732 – 1726 (2 x ester carbonyl), 1597 (aromatic); Molecular Formula $C_{20}H_{22}O_7$; Pale pinkish solid; Melting range 136 – 140^{0}C; Elemental Analysis, Calcd.: C 64.22 %, H 5.92 %, O 29.86 %. Found C 64.25 %, H 5.95 %, O 29.83 %;

4-ethoxy-3-methoxy benzoic acid (4)

Off white solid; Molecular Formula $C_{10}H_{12}O_4$; ^{1}H NMR ($CDCl_3$, 400 MHz) δ ppm : 1.21 (t, *J* = 7.2 Hz, 3H, terminal methyl from ethyl bromide moiety), 3.91 (s, 3H, 1 x Ar–**OCH_3** group), 4.08 (q, *J* = 6.8 Hz, 13.6 Hz, 2H, 1 x -**OCH_2** from ethyl bromide moiety), 6.8 – 7.9 (m, 3H, ArH), 10.8 (brs, 1H, -OH, D_2O exchangeable); TOF MS ES: 219 (M + Na); IR (KBr) cm-1:- 2929, 2854, 2873 (methyl, methylene, methines), 1710 (acid carbonyl), 1597 (aromatic);

CHROMATOGRAPHIC SYSTEM:

Column chromatography : For column chromatography 100 – 200 mesh Acme grade silica gel is used. The crude reaction mixture is concentrated under reduced pressure to yield crude mass which is preadsorbed on silica gel and purified by column chromatography with increase in concentration of Ethyl acetate in Petroleum ether. The fractions having similar 'rf" values were pooled together, concentrated and subjected for characterization using various spectroscopic techniques.

Thin layer chromatography : TLC plates were prepared using silica gel G (ACME, BOMBAY). Pet. ether: EtOAc (95 : 5) was used as the solvent system.

BIOLOGICAL ACTIVITY :

Antibacterial Activity using ditch plate method[12] :-

The synthesized molecules were screened for their antibacterial activity at 100 µg/ml concentration using ditch plate method against Gram positive (*Staphylococcus aureus*) and Gram negative (*Escherichia coli*) bacterial species qualitatively. The

results of the antibacterial activities are summarized in **Table 1**.

Theory : Ditch plate method is the method of chosen to test the anti-bacterial activity of compounds. It is a preliminary method to screen the anti-microbial potential of compounds / drugs, which are insoluble or partially soluble in aqueous phase. In this method , the test compound is seeded in an agar plate and the test organisms are streaked across to test the inhibition of the growth as a marker of anti-microbial activity.

PROCEDURE : A ditch (10 mm x 70 mm) is cut into sterile MH agar plate. The test drug / compound is added to 5 ml molten MH agar butt at 40^0C and this mixture is poured into the ditch and allowed to solidify. The ditch should be made in level with the rest of the agar by pouring the mixture . The different bacterial cultures are streaked perpendicular to the ditch using nichrome wire loop. The plate is then incubated at 37^0 C for 24 hours. The results are observed as inhibition of bacterial growth on the ditch as well as adjacent to the ditch .

Table 1 : Antibacterial Activity Results

Sr. No	Compound No.	Antibacterial Activity	
		Against Gram - ve bacteria species (*Escherichia coli*)	Against Gram + ve bacterial species (*Staphylococcus aureus*)
1. Base molecule	Ethyl Vanillate	+	+
2. Std Drug	Ampicillin	+	+
3.	Synthetic Derivative	+	-

The above results shows that the base molecule, ethyl vanillate has antibacterial activity against both the bacterial cultures. Its derivative *viz.* **1** is active against only *Escherichia coli* (Gram negative bacteria). Thus, hybrid derivative of ethyl vanillate was potential antibacterial candidate. In depth analysis of this compound through structure activity relationship studies would provide further insight and can be an interesting topic of future studies.

IV. ACKNOWLEDGEMENT

One of the authors Mrs. Karuna Bhagwat thanks Dr. Vijay D. Gangan, Research Guide and HOD, Reena Mehta College of Arts, Commerce, Science and Management Studies for suggesting a problem and constant valuable guidance through out the course and providing laboratory facilities.

REFERENCES

[1]. Wattenberg L. W., Coccia J. B. and Lam L. K. Inhibitory effects of phenolic compounds on benzo[a]pyrene induced neoplasia. Cancer Res. 1980, 40 : 2820 – 2823.

[2]. Talalay P, De Long M. J. and Prochaska H. J. Identification of a common chemical signal regulating the induction of enzymes that protect against chemical carcinogens. Proc. Natl. Acad. Sci. 1988, 85 : 8261 – 8265.

[3]. Macheix J. J., Fleuriet A. and Billot J. Fruit phenolics. CRC press Inc., Boca Raton 1990, FL.

[4]. Natarajan K, Singh S, Burke T. R. Jr., Grunberger D. and Aggarwal B. B. Caffeic acid phenethyl ester is a potent and specific inhibitor of activation of nuclear transcription factor NF-kappa B. Proc. Natl. Acad. Sci. 1996, 93 : 9090 – 9095.

[5]. Chiao C., Carothers A. M., Grunberger D., Solomon G., Preston G. A. and Barrett J. C. Apoptosis and altered redox state induced by caffeic acid phenethyl ester (CAPE) in transformed rat fibroblast cells. Cancer Res. 1995, 55 : 3576 – 3583.

[6]. Burke T. R. Jr, Fesen M. R., Mazumder A., Wang J., Carothers A. M., Grunberger D., Driscoll J., Kohn K. and Pommier Y. Hydroxylated aromatic inhibitors of HIV-1 integrase. J. Med. Chem. 1995, 38 (21) : 4171 – 4178.

[7]. Bose J. S., Gangan V. D., Jain S. K. and Manna S. K. Novel caffeic acid ester derivative induces apoptosis by expressing FasL and downregulating NFKappaB : Potentiation of cell death mediated by chemotherapeutic agents. J. Cell Physiol. 2009, 218 (3) : 653 – 662.

[8]. Bose J. S., Gangan V. D., Jain S. K. and Manna S. K. Downregulation of inflammatory responses by novel caffeine acid ester derivative by inhibiting NF kappaB. Journal of Clinical Immunology 2009, 29, pp. 90 - 98.

[9]. Bose J. S., Gangan V. D. (2007). Applied for US patent on “NOVEL CHEMOTHERAPEUTIC AGENTS AGAINST INFLAMMATION AND CANCER” from Reliance Life Sciences [INDIA 1696/MUM/2006, Publication No. WO/2008/062466, [International Application No. PCT/IN2007/000488].

[10]. Satpute M. S., Shastri, I. and Gangan V. D. (2018); International Journal for Research in Applied Science & Engineering Technology, 6 (XII), pp. 330 - 334.

[11]. Satpute M. S., Shastri, I. and Gangan V. D. International Journal of Scientific Research in Science and Technology 2018, 4 (11), pp. 219 - 223.

[12]. a) Finn R. K. Theory of Agar Diffussion Methods for Bioassay. Anal. Chem., 1959, 31 (6), pp 975 – 977. b) Al lafi T et. al.. The effect of miswak used in Jordan and Middle East on oral bacteria. International Dental Journal, 1995, 45 (3), pp. 218 – 222.