SYNTHESIS OF SOME NOVEL (E)-METHYL 2,4-DIMETHYL-5-(3-OXO-3-PHENYLPROP-1-EN-1-YL)-1H-PYRROLE-3-CARBOXYLATE DERIVATIVES AS ANTIMICROBIAL AGENT

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ABSTRACT

Objective: The objective of the present study was to synthesize a series of some novel (E)-methyl 2,4-dimethyl-5-(3-oxo-3-phenylprop-1-en-1-yl)-1H-pyrrole-3-carboxylate derivatives and to evaluate it’s in vitro antimicrobial activities.

Methods: A novel series of (E)-methyl 2,4-dimethyl-5-(3-oxo-3-phenylprop-1-en-1-yl)-1H-pyrrole-3-carboxylate derivative (8a-l) has been synthesized by cyclization (Knorr reaction) hydrolysis, decarbonylation, and Vilsmeier-Haack formulation reaction. 5-formyl-2,4-dimethyl-1H-pyrrole-3-carboxylate 6 undergo condensation with acetophenone derivatives 7a-l in methanol and potassium hydroxide. The synthesized compounds were screened for in vitro antimicrobial screening.

Results: The structures of the synthesized compounds were characterized by infrared, 1H nuclear magnetic resonance, and mass spectroscopy. The antimicrobial activity data revealed that the synthesized derivatives possess good antibacterial and antifungal activity which is attributed due to the presence of the heterocyclic ring; further, the activity increased with the introduction of a methoxy group in the structure.

Conclusions: New pyrrole chalcone derivatives act as significant antimicrobial agents, easy work-up procedure and reaction take place with minimum side product. Antimicrobial activity report provides an interesting template for the syntheses of new antimicrobial agents and may be helpful for the design of new therapeutic tools.

Keywords: Antimicrobial, Gram-positive, Gram-negative bacteria, Pyrrole chalcones.

INTRODUCTION

Heterocyclic compounds have a wide range of applications in pharmaceutical chemistry. Heterocyclic compounds have natural as well as synthetic source. Pyrrole has gain remarkable importance among the new heterocycles due to biological potential. Pyrrole has antimalarial and enzyme inhibition properties. Pyrrole is five-membered nitrogen-containing heterocyclic compound. Colorless liquid compound and on exposing to air it becomes darker. It has importance in more complex macromolecules such as porphyrins of heme and chlorophyll. It was isolated from pyrolysate of bone. Its name came from Greek [1]. In recent years, there has been growing interest in chalcones and their important role in the prevention of various diseases such as cancer and chronic inflammation.

The chalcones are structurally simple and largest class of plant secondary metabolites, which serves as a defense mechanism in plants to counteract reactive oxygen species to prevent damage by micro-organisms [2]. Antimicrobial activity of bis-heterocyclic chalcones was tested against selective pathogens Escherichia coli, Candida albicans, and Aspergillus niger [3]. Several studies have pointed out the antimicrobial potential of chalcones against a wide range of fungi and bacteria, including resistant ones, clearly indicating that they are attractive target compounds for the discovery and development of new anti-infective agents [4-10]. These flavonoid family compounds display an impressive area of biological activities, among which antibacterial [11], antifungal [12], antioxidant [13,14], antileishmanial [15], antimalarial [16], angiogenesis inhibitor [17], anti-inflammatory [18,19], antiviral [20], antimitotic [21], anticancer [22], anti-invasive [23,24], and antiproliferative [25]. Activities have been cited in the literature. Chalcones strongly inhibit the polymerization of tubulin by binding to the colchicine-binding site [16]. The α, β-unsaturated compounds may possess diverse pharmacological activities, and it has been found that the synthesized chalcones interact with the bovine serum albumin, a protein mainly responsible for the transportation of a number of compounds [26]. Substantial preliminary antibacterial activity was reported for the new chalcones at C8 of the coumarin nucleus, against Gram-positive and a Gram-negative bacteria [27]. In view of the pharmacological activities of phenothiazine, pyrazole, and pyrrole a number of the 2-substituted, pyrrole derivatives have been synthesized which contain pyrrole heterocycles [28].

Therefore, in view of these important biological activities of pyrrole scaffold. We reported the synthesis of some novel (E)-methyl 2,4-dimethyl-5-(3-oxo-3-phenylprop-1-en-1-yl)-1H-pyrrole-3-carboxylate derivatives and tested the antibacterial and antifungal activities of the new resulting compounds which have been found to possess an interesting profile of pharmacological activity, against Gram-negative, Gram-positive bacteria and fungi 8f, 8e, 8c. Chemically chalcones are open chain flavonoids in which the two aromatic rings are joined together by three carbons, α, and β-unsaturated carbonyl system. Fundamentally, they are considered as derivatives of phenyl styryl ketone. In view of the varied biological and pharmacological applications, we have synthesized some new (E)-methyl 2,4-dimethyl-5-(3-oxo-3-phenylprop-1-en-1-yl)-1H-pyrrole-3-carboxylate derivatives.

In general, chalcones contain C6–C3–C6 unit but the compounds reported in the present study contain C6–C3–C6 unit.
EXPERIMENTAL METHODS

The melting points of all the synthesized compounds were determined in open capillary tubes and are uncorrected. The purity of all compounds was checked by thin-layer chromatography (TLC). Silica gel 60 F254 Infrared (IR) spectra were recorded on Fourier-transform IR (Model: Nicolet iS10 Mid). 1H NMR spectra were recorded on 400 MHz-NMR Spectrometer (CDCl3/dimethyl sulfoxide (DMSO)-d6) (Bruker Biospin International AG); chemical shifts (δ) are in parts per million (ppm) relative to TMS and coupling constant (J) is expressed in hertz (Hz). Mass spectra were recorded on a Thermo Scientific Q-Exactive, Accela 1250 pump, Macro mass spectrometer (Water, Milford, MA) by electrospray method (ES). TLC was performed on the and visualized in UV light. All chemicals and solvents were obtained from Spectrum, Aldrich, or Merck and are of analytical grade, and were used freshly after opening.

Synthesis of diethyl 3, 5-dimethyl-1H-pyrrole-2,4-dicarboxylate (2)

To prepare diethyl 3, 5-dimethyl-1H-pyrrole-2,4-dicarboxylate, a mixture of acetic acid (40 ml) and ethyl acetoacetic ester (20 ml, 15.6 mmol) was added to an aqueous solution of sodium nitrite (5.6 g, 81 mmol) in water (6 ml) cooled in an ice bath to keep the temperature of reaction mixture 5–10°C. The yellowish clear reaction mixture was kept for another 2.5 h at the same temperature to further complete reaction. Then, zinc powder (11 g, 52.34 mmol, 300 mesh) was added to this mixture portion wise while the reaction mixture was kept <25°C. It was then heated to 45–50°C for 10 min and then continued slow heating to 95°C for another 1 h exotherm was observed. Cooled reaction mass to room temperature, the yellowish precipitate was collected and washed with ice water to remove the acetic acid. Recrystallized the yellow crude solid from ethanol to afford a pale yellow crystalline solid powder of pure 2 (5.6 g, yield 81%).

Synthesis of 4-(ethoxy carbonyl)-3, 5-dimethyl-1H-pyrrole-2-carboxylate (3)

To prepare 4-(ethoxy carbonyl)-3,5-dimethyl-1H-pyrrole-2-carboxylic acid, compound 2 (20 g, 83.6 mmol) was added into a flask equipped with a stirrer, a thermometer, and a condenser; absolute ethanol (120 ml) was added into the flask. An aqueous solution (240 ml) of potassium hydroxide (1.18 g, 21.03 mmol) was added and heated at 50–60°C for 10 h with monitoring by TLC until reaction completion. The reaction was then stopped by cooling down to room temperature. The reaction mixture was then poured into ice-water (600 ml) to form an aqueous solution. A small amount of insoluble substance was filtered and kept aside. Aqueous layers were adjusted with hydrochloric acid (2 mol/L) to pH 4 to afford some white precipitates, which were collected through filtering and washing with ice-water to provide 3 (15 g, yield 85%).

Synthesis of methyl 4-formyl-2,4-dimethyl-1H-pyrrole-3-carboxylate (4)

To prepare methyl 4-formyl-2,4-dimethyl-1H-pyrrole-3-carboxylate, compound 2 (10 g, 47.35 mmol) was added into flask equipped with a stirrer, a thermometer, and a condenser; HCl (5 ml) and methanol (150 ml) were added into the flask and heated up to 75–80°C for 1.5 h with monitoring by TLC until completion of decarboxylation. The reaction was cooled down to room temperature, further cooled up to 0–5°C. Slowly added solution of KOH (8.8 g) dissolving in water (100 ml) and the reaction mass was heated at 60–65°C temperature for 5–6 h with monitoring by TLC until completion of hydrolysis. Stopped heating and cooled to room temperature. The reaction mass was extracted with ethyl acetate to remove impurities and separated aqueous layer was collected and acidified with 1:1 HCl to pH 3–4 to afford light brown solid precipitates, which was collected through filtering and washing with ice-water to provide compound 4 (5.5 g, yield 83%), m. p. 75–76°C [lit. 74–76°C [30] (eluent: chloroform/methanol, v/v = 10:1)].

Synthesis of methyl 2,4-dimethyl-1H-pyrrole-3-carboxylate (5)

To prepare methyl 2,4-dimethyl-1H-pyrrole-3-carboxylate, compound 4 (5.0 g, 35 mmol) was added into flask equipped with a stirrer, a thermometer, and condenser; H2SO4 (0.2 ml) and methanol (75 ml) into the flask and heated up to 60–65°C for 1.5 h monitored by TLC until reaction completion. The reaction was then stopped by cooling down to room temperature, further cooled up to 0–5°C. The reaction mixture was poured into ice-water (100 ml) to afford light brown solid precipitates, which was collected through filtering and washing with ice-water to provide 5 (4.126g, 68%), m. p. 70–72°C [eluent: chloroform/methanol, v/v = 10:1].

Synthesis of methyl 5-formyl-2,4-dimethyl-1H-pyrrole-3-carboxylate (6)

Anhydrous DMF (2.5 ml, 200 mmol) in a dry flask was precooled to 0°C with a salt-ice bath, into which a solution of POCl3 (3.60 ml, 320 mmol) in anhydrous dichloromethane (10 ml) was added dropwise. The temperature should be kept <3°C during the whole addition process. The reaction mixture was stirred for 0.5 h using an ice-bath before dropwise addition of a solution of methyl 2,4-dimethyl-1H-pyrrole-3-carboxylate 5 (4.0, 26.11 mmol) in anhydrous dichloromethane (20.0 ml). The mixture was heated under reflux for 3 h and monitored by TLC until reaction completion. The reaction was then stopped by cooling down to room temperature, further cooled up to 0–5°C. Neutralize the reaction mixture using 30%aq. (Aqueous) potassium carbonate solution till pH 8–9. Stirred the reaction mass for 1 h at room temperature, separated the organic layer using separating funnel, extracted the aqueous layer with DCM (100 ml × 3 times) separated organic layer was washed with water (50 ml × 3 times) and separated organic layer was dried over sodium sulfate, distilled out DCM under vacuum completely to afford pale-gray solid, dried under vacuum to afford compound 6 (4.5 g, yield 95%), m. p. 167–169°C [eluent: chloroform/methanol, v/v = 10:1]. IR (KBr): cm−1 3244 (-NH), 1698 (C=O), 1559 (C=O), ‘H NMR (400 MHz, CDCl3) δ 2.569–2.592 (6H,-CH2), 3.863 (3H, -OCH3), 9.637 (1H, -CHO), 9.932 (1H, NH).

General procedure for the synthesis of (E)-methyl 2,4-dimethyl-5-(3-oxo-3-(4-fluorophenyl)prop-1-yl)-1H-pyrrole-3-carboxylate (8a–i)

An aqueous solution of KOH (0.093 g, 1.65 mmol) was added to cooled suspension of 1-(4-fluorophenyl)ethanone 7a (0.190 g, 1.38 mmol) and methanol-water mixture 5 ml (9:5:0.5) mixture was stirred 5–10°C for 30 min. After 30 min added methyl 5-formyl-2,4-dimethyl-1H-pyrrole-3-carboxylate 6 (0.250 g, 1.38 mmol). Raised temperature up to room temperature and the mixture was stirred at room temperature for overnight. The reaction mixture was poured into water. The solid product (8a) separated out was filtered and washed with plenty of water finally crystallized from ethanol. The compounds (8a–i) were prepared by following the aforementioned procedure. Their structures have been confirmed by mass, IR, and ‘H NMR spectra.

(E)-methyl 5-(3-(4-fluorophenyl)-3-oxoprop-1-en-1-yl)-1H-pyrrole-3-carboxylate (8a)

Yield: 75%; molecular formula (MF): C16H13FNO3; 301.31; yellow solid; MP: 207–210°C; IR (KBr) cm−1: 3235 (-NH), 1696 (C=O), 1530 (C=O); ‘H NMR (400 MHz, DMSO-d6) δ 2.396 (3H, -CH3) 2.574 (3H, -CH3), 3.838 (3H, -OCH3), 6.978–7.016 (d, J = 15.2 Hz, 1H, ethynyl), 7.100–7.143 (2H, -Ar), 7.789–7.828 (d, J = 15.6 Hz, 1H, ethynyl), 7.978–8.014 (2H, -Ar), 9.015 9 (s, 1H, -NH), HRMS = 301.1817 [M+H].

(E)-methyl 2,4-dimethyl-5-(3-oxo-3-(3,4,5-trimethoxyphenyl)prop-1-en-1-yl)-1H-pyrrole-3-carboxylate (8b)

Yield: 79%; MF: C21H15NO2; 373.4; yellow solid; MP: 138–141°C; IR (KBr) cm−1: 3379 (-NH), 1675 (C=O), 1545 (C=C); ‘H NMR (400 MHz, DMSO-d6) δ 2.412 (3H, -CH3), 2.508 (3H, -CH3), 3.842 (3H, -OCH3), 3.919–3.932 (9H, -OCH3), 6.990–6.958 (d, J = 15.2 Hz, 1H, ethynyl), 7.199–7.216 (2H, -Ar), 7.794–7.853 (d, J = 15.6 Hz, 1H, ethynyl), 8.939 (s, 1H, -NH), HRMS = 374.1589 [M+H].
(E)-methyl 5-(3-(4-cyanophenyl)-3-oxo-2-propyl-1-enyl)-2,4-dimethyl-1H-pyrrrole-3-carboxylate (8c)

Yield: 69%; MP: C$_7$H$_7$NO$_2$/308.33; yellow solid; MP: 218–220°C; IR(KBr) cm$^{-1}$: 3338 (NH), 1625 (C=O), 1555 (C=C); 'H NMR (400 MHz, DMSO-$d_6$) $\delta$ 3.791 (s, 3H, -OCH$_3$) 7.243–7.31 (d, $J = 15.2$ Hz, 1H, ethylenic), 7.732–7.753 (d, 2H, -Ar), 7.765–7.804 (d, $J = 15.6$ Hz, 1H, ethylenic), 8.083–8.104 (d, 2H, -Ar) 10.862 s (1H, -NH); HRMS: 309.1234 [M+H].

(E)-methyl 2,4-dimethyl-5-(3-(4-nitrophenyl)-3-oxo-2-propyl-1-enyl)-1H-pyrrrole-3-carboxylate (8b)

Yield: 71%; MP: C$_7$H$_7$NO$_2$/328.32; orange solid; MP: 99–101°C; IR(KBr) cm$^{-1}$: 3218 (NH), 1698 (C=O), 1530 (C=C); 'H NMR (400 MHz, DMSO-$d_6$) $\delta$ 2.396 (s, 3H, -CH$_3$) 2.418 (s, 3H, -CH$_3$), 3.849 (s, 3H, -OCH$_3$), 7.680–7.716 (d, $J = 15.2$ Hz, 1H, ethylenic), 7.822–7.872 (d, $J = 15.2$ Hz, 1H, ethylenic), 7.909–7.936 (d, $J = 8.4$ Hz, 2H, -Ar), 9.047 (s, 1H, -NH); ESI-MS: 318.0903 [M+H].

(E)-methyl 5-(3-(4-chlorophenyl)-3-oxo-2-propyl-1-enyl)-2,4-dimethyl-1H-pyrrrole-3-carboxylate (8e)

Yield: 72%; MP: C$_7$H$_7$ClNO$_2$/317.76; brownish yellow solid; MP: 170–172°C; IR(KBr) cm$^{-1}$: 3258 (NH), 1697 (C=O), 1530 (C=C); 'H NMR (400 MHz, DMSO-$d_6$) $\delta$ 2.4 (s, 3H, -CH$_3$), 2.598 (s, 3H, -CH$_3$), 3.848 (s, 3H, -OCH$_3$), 6.926–6.964 (d, $J = 15.2$ Hz, 1H, ethylenic), 7.821–7.859 (d, $J = 15.2$ Hz, 1H, ethylenic), 8.082–8.104 (d, $J = 8.8$ Hz, 2H, -Ar), 8.952 (s, 1H, -NH); HRMS: 329.1132 [M+H].

(E)-methyl 5-(3-(4-methoxyphenyl)-3-oxo-2-propyl-1-enyl)-2,4-dimethyl-1H-pyrrrole-3-carboxylate (8d)

Yield: 67%; MP: C$_7$H$_7$NO$_2$/313.34; yellow solid; MP: 179–180°C; IR(KBr) cm$^{-1}$: 3191 (NH), 1699 (C=O), 1534 (C=C); 'H NMR (400 MHz, DMSO-$d_6$) $\delta$ 2.396 (s, 3H, -CH$_3$), 2.570 (s, 3H, -CH$_3$), 3.849 (s, 3H, -OCH$_3$), 3.882 (s, 3H, -OCH$_3$), 6.904–6.925 (d, $J = 8.4$ Hz, 2H, -Ar), 7.041–7.080 (d, $J = 15.6$ Hz, 1H, ethylenic), 7.787–7.826 (d, $J = 15.6$ Hz, 1H, ethylenic), 7.969–7.990 (d, $J = 8.4$ Hz, 2H, -Ar) 9.106 (s, 1H, -NH); HRMS: 314.1379 [M+H].

(E)-methyl 5-(2-(4-methoxyphenyl)-3-oxo-2-propyl-1-enyl)-2,4-dimethyl-1H-pyrrrole-3-carboxylate (8g)

Yield: 65%; MP: C$_7$H$_7$NO$_2$/343.37; yellow solid; MP: 137–139°C; IR(KBr) cm$^{-1}$: 3225 (NH), 1693 (C=O), 1536 (C=C); 'H NMR (400 MHz, DMSO-$d_6$) $\delta$ 2.375 (s, 3H, -CH$_3$), 2.558 (s, 3H, -CH$_3$), 3.865 (s, 3H, -OCH$_3$), 6.477–6.483 (d, 1H, -Ar), 6.536–6.558 (d, $J = 8.8$ Hz, 1H, -Ar), 6.942–6.981 (d, $J = 15.6$ Hz, 1H, ethylenic), 7.637–7.637 (d, $J = 15.6$ Hz 1H, ethylenic), 7.680–7.701 (d, $J = 8.4$ Hz 1H, -Ar), 8.812 (s, 1H, -NH); ESI-MS: 344.20 [M+H].

Scheme 1: Synthetic scheme, reagent, and conditions: (a) (i) NaNO$_2$, (ii) AcOH, Zn, 90–95°C, (b) Aq. KOH, EtOH, 50–60°C, (c) (i) HCl, MeOH, 75–80°C, (ii) Aq.KOH, MeOH, 60–65°C, (d) Cat.H$_2$SO$_4$, MeOH, 60–65°C, (e) DMF/POCl$_3$, DCM, reflux, (f) Aq.KOH, MeOH, rt.
(E)-methyl 2,4-dimethyl-5-[3-(3-nitrophenyl)-3-oxoprop-1-en-1-yl]-1H-pyrrole-3-carboxylate (8k)
Yield: 57%; MF: C₉H₅N₂O₂; 328.31; yellow solid; MP: 102-105°C; IR (KBr) cm⁻¹: 3,218 (N-H), 1,687 (C=O), 1,558 (C=C); H NMR (400 MHz, DMSO-d₆) δ: 2.446 (s, 3H, -CH₃), 2.632 (s, 3H, -CH₃), 3.855 (s, 3H, -OCH₃), 6.959-6.997 (d, J = 15.2 Hz, 1H, ethylenic), 7.688-7.728 (m, 1H, -Ar), 7.898-7.936 (d, J = 15.2 Hz, 1H, ethylenic), 8.357-8.438 (m, 3H, -Ar), 8.817 (s, 1H, -NH).

(E)-methyl 2,4-dimethyl-5-(3-oxo-3,4,5-trimethylphenyl)prop-1-en-1-yl)-1H-pyrrole-3-carboxylate (8l)
Yield: 55%; MF: C₁₅H₁₈N₂O₂; 325.40; yellow solid; MP: 147-150°C; IR (KBr) cm⁻¹: 3,195 (N-H), 2,985 (C=O), 1,693 (C=O), 1,539 (C=C); H NMR (400 MHz, DMSO-d₆) δ: 2.404 (s, 6H, -CH₃), 2.563 (s, 9H, -CH₃), 3.831 (s, 3H, -OCH₃), 7.056-7.095 (d, J = 15.6 Hz, 1H, ethylenic), 7.212-7.232 (s, 1H, -Ar), 7.784-7.823 (d, J = 15.6 Hz, 1H, ethylenic), 7.861-7.881 (s, 1H, -Ar); 9.269 (s, 1H, -NH).

Biological evaluation
Antibacterial and antifungal activity
Some of the synthesized compounds were screened for in vitro antibacterial and antifungal activities against Gram-positive and Gram-negative bacteria. In Gram-positive bacteria, Micrococcus luteus, Bacillus megaterium, Staphylococcus aureus, and Bacillus cereus were used and in Gram-negative bacteria, Salmonella typhi, E. coli, Pseudomonas aeruginosa, and Salmonella abony were used against standard tetracycline. In antifungal activity, C. albicans, Saccharomyces cerevisiae and Aspergillus niger were used against standard nystatin. Antimicrobial activities were carried out on a nutrient agar with standard composition and by standard procedure of paper disk method [31].

Petri dishes and necessary glassware’s were sterilized in hot air oven (190°C, 45 min). The nutrient agar and saline (0.82% NaCl) were sterilized in an autoclave (121°C, 15 psi, and 20 min). The inoculum was prepared in sterile saline, and optical density of all pathogens was adjusted to 0.10 at 625 nm on Chemito Spectra scan UV 2600 spectrometer (Mumbai, India), which is equivalent to 0.5 McFarland standards. The nutrient agar plates were prepared by pour plate method [31]. The sensitivity of compounds was tested by the disk diffusion method (paper disk method). All the bacterial cells were cultured in nutrient plates and the compounds to be tested were dissolved in DMSO solvent and were soaked in paper disks. The disks were placed into the plates and incubated at 37°C for 24 h. The diameter in millimeter of the zone of inhibition around each disk was measured by the scale.

RESULTS AND DISCUSSION
Chemistry
The synthesis of (E)-methyl 2,4-dimethyl-5-[3-(3-phenylprop-1-en-1-yl)-1H-pyrrole-3-carboxylate derivatives (8a-8l) Scheme 1. Diethyl 3,5-dimethyl-1H-pyrrole-2,4-dicarboxylate (2) was first obtained through Knorr reaction from acetyl acetate ethyl ester (1) and zinc and acetic acid. The diethyl 3,5-dimethyl-1H-pyrrole-2,4-dicarboxylate (2) has selectively hydrolyzed by KOH in ethyl alcohol to yield 4-(ethoxycarbonyl)-3,5-dimethyl-1H-pyrrole-2-carboxylic acid (3). The 4-(ethoxycarbonyl)-3,5-dimethyl-1H-pyrrole-2-carboxylic acid was further subjected to decarboxylation reaction in the presence of potassium hydroxide yield 2,4-dimethyl-1H-pyrrole-3-carboxylic acid (4). Compound 4 on esterification reaction using H₂O and methanol to form methyl 2,4-dimethyl-1H-pyrrole-3-carboxylate (5). Compound 5 further subjected to Wilemeier–Haack formulation reaction to afford methyl 5-formyl-2,4-dimethyl-1H-pyrrole-3-carboxylate (6). The methyl 5-formyl-2,4-dimethyl-1H-pyrrole-3-carboxylate was subjected to base-catalyzed Claisen-Schmidt condensation reaction [32], with appropriate acetoephones (7a-l) in Aq. KOH in MeOH to yield (E)-methyl 2,4-dimethyl-5-[3-(3-phenylprop-1-en-1-yl)-1H-pyrrole-
3-carboxylate derivatives (8a-l). The purity of compounds was checked by TLC. Analytical and spectral data (HNMR, IR, mass) of newly synthesized compounds were in full agreement with the proposed structure. The structure of 6 is interpreted from spectroscopic data its NMR spectrum exhibit characteristics of the aldehydic proton at δ = 9.6 ppm and -NH pyrrole peaks at δ = 9.9 ppm. The structure of 8a is interpreted from spectroscopic data. The IR spectrum of 8a shows characteristic bands at 1696 due to C=O stretching. Its 1H NMR spectrum exhibits the presence of olefinic protons as a doublet at δ = 7.0 and 7.8 regions with a mutual coupling constant value (J = 15.2 and 15.6 Hz). These observed coupling constant values indicate the presence of E-core configuration from the structure and the remaining protons as well aliphatic protons appear at their respective positions. The -NH is highly deshielded and appears at δ = 9.01 ppm. Mass spectrum of (E)-methyl 5-(3-(4-fluorophenyl)-3-oxopropan-1-yl)-2,4-dimethyl-1H-pyrrole-3-carboxylate showed (M+H) peak at 302.11.

Antimicrobial activity
Antimicrobial activity result of synthesized (E)-methyl 2,4-dimethyl-5-(3-oxo-3-phenylprop-1-en-1-yl)-1H-pyrrole-3-carboxylate (8a-I) reveals that the compounds 8f, 8e, and 8c were found to possess significant activity while 8a, 8d, 8g, and 8k showed moderate to good activity compared to the standard reference nystatin and tetracycline Table 1.

The diameter in millimeter of the zone of inhibition around each disk was measured by a scale, and the observed data of antimicrobial and antifungal activities of compounds and the standard drugs are given in Table 1. Among all the compound screened 8f, 8e, and 8c showed significant antibacterial activity against Gram-positive bacteria.

Solvent used-DMSO-The effect of DMSO on pathogen was also checked, but it was found that DMSO does not affect the pathogen.

The concentration of compounds used for the test - 1 mg/ml.

All results (zone of inhibition) are recorded in millimeter (mm).

Compounds 8a and 8f showed significant activity against Gram-negative bacteria as comparable with that of standard drug tested. Compounds 8c, 8e, 8f, 8g, and 8k showed good activity against fungal pathogens as compared with that of standard drug tested. Hence, the result of all preliminary studies indicated that the substituted (E)-methyl 5-[(3-(4-fluorophenyl)-3-oxopropane-1-yl)-2,4-dimethyl-1H-pyrrole-3-carboxylate (8f), (E)-methyl 5-[(3-(4-cyanophenyl)-3-oxopropane-1-yl)-2,4-dimethyl-1H-pyrrole-3-carboxylate (8c) and (E)-methyl 5-[(3-(4-chlorophenyl)-3-oxopropane-1-yl)-2,4-dimethyl-1H-pyrrole-3-carboxylate (8e) moiety represent a new class of pharmacophore for broad spectrum of antimicrobial activity.

CONCLUSION
In summary, we have synthesized a series of pyrrole incorporated new (E)-methyl 2,4-dimethyl-5-(3-oxo-3-phenylprop-1-yl)-1H-pyrrole-3-carboxylate derivatives (8a-l), and their antimicrobial activities have been evaluated. The compounds 8f, 8e, and 8c demonstrated significant inhibition and 8a, 8d, 8g, and 8k showed moderate to good activity against the tested strains. The importance of such work lies in the possibilities that the new compounds might be more efficacious drugs against bacteria, which could be helpful in designing more potent antibacterial agent for therapeutic use.

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AUTHORS’ CONTRIBUTIONS
Mahesh Hublikar carried out the experiment, synthesis, and wrote this article. Kshenuthth Bhalke provided guidance, critical review and revision. Prashant Dixit supported for biological evaluation of synthesized compounds, Vikas Kadu contributed to the final manuscript. Sachin Shirame contributed for interpretation of data. Dattatraya Raut encouraged Mahesh Hublikar to explore the finding of work.

CONFLICTS OF INTEREST
The authors declare that they have no conflicts of interest.

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