

# Synthesis, Characterization and Antimicrobial Study of Some New Cyclohexenone Derivatives

Anil N. Mayekar

Department of Studies in Chemistry, University of Mysore  
Manasagangotri 570 006, India

&

SeQuent Scientific Limited, 120 A and B, Industrial Area  
Baikampady, New Mangalore, 575 011, India  
Tel: 91-824-240-2306 E-mail: anilmayekar@gmail.com

Hongqi Li

Key Laboratory of Science & Technology of Eco-Textile  
Ministry of Education, College of Chemistry  
Chemical Engineering & Biotechnology, Donghua University  
Shanghai 201620, China  
Tel: 86-21-6779-2594 E-mail: hongqili@dhu.edu.cn

H. S. Yathirajan (Corresponding author)

Department of Studies in Chemistry, University of Mysore  
Manasagangotri 570 006, India  
Tel: 91-821-241-9656 E-mail: yathirajan@hotmail.com

B. Narayana

Department of Chemistry, Mangalore University  
Mangalagangotri 574 199, India  
Tel: 91-824-228-7262 E-mail: nbadiadka@yahoo.co.uk

N. Suchetha Kumari

Department of Biochemistry, Justice K.S. Hegde Medical Academy, Deralakatte  
Mangalore-574 162, India  
Tel: 91-824-220-2471 E-mail: suchetha.shetty@rediffmail.com

*This research work is supported by University of Mysore, Mysore, SeQuent Scientific Ltd., Mangalore and Donghua University, People's Republic of China.*

## Abstract

A series of chalcones and its cyclohexenone derivatives derived from 6-methoxy-2-naphthaldehyde are described. The chalcones synthesized through Claisen-Schmidt condensation reaction were treated with ethylacetoacetate in presence of NaOH to get the cyclocondensed product ethyl-4-(aryl)-6-(6-methoxy-2-naphthyl)-2-oxo-cyclohex-3-ene-1-carboxylate. The synthesized compounds were characterized from elemental analysis and spectral data. Ethyl 4-(4-chlorophenyl)-6-(6-methoxy-2-naphthyl)-2-oxo-cyclohex-3-ene-1-carboxylate (**4f**) was studied by single crystal X-ray diffraction. The newly synthesized compounds were screened for their antimicrobial activity.

**Keywords:** Cyclohexenones, 6-Methoxy-2-naphthyl chalcones, Single Crystal XRD, Antimicrobial activity

## 1. Introduction

Chalcones belong to a class of  $\alpha,\beta$ -unsaturated aromatic ketones which occur abundantly in nature, and have drawn much attention because of their benefits to human application (Dhar, 1981; Torigoo *et al.*, 1983; Sukumaran & Kuttan, 1991). These compounds are precursors of flavonoids and isoflavonoids which are abundant in plants. Various biological activities are associated with chalcones, such as anticancer (Modzelewska *et al.*, 2006), antitumor (Kumar *et al.*, 2003), antioxidant (Suksamrarn *et al.*, 2003) and antimalarial (Ferrer *et al.*, 2009). In addition of being used in pharmaceutical industries, chalcones also find wide applications in dyes (Asiri, 2003) and cosmetic compositions (Forestier *et al.*, 1989). Apart from being biologically important compounds, chalcone derivatives show non-linear optical (NLO) properties with excellent blue light transmittance and good crystallizability (Shettigar *et al.*, 2006). The presence of naphthyl ring in chalcones has shown promising antimicrobial and antihyperglycemic activity (Damazio *et al.*, 2010; Rajendraprasad *et al.*, 2006). Michael addition of ethylacetoacetate to chalcone yields 4,6-diaryl-2-oxo-cyclohex-3-ene-1-carboxylate derivatives, which are efficient synthons for fused isoxazoles pyrazoles and quinazolins (Padmavathi *et al.*, 2000; Senguttuvan *et al.*, 2010). Cyclohexenone derivatives are well known lead molecules for the treatment of inflammation and autoimmune diseases (Tanaka *et al.*, 1997). Several reports have pointed out the importance of cyclohexenones for antimicrobial and antitubercular activity (Vyas *et al.*, 2009). Nebumetone, 4-(6-methoxy-2-naphthyl)-2-butanone and Naproxen, 2-(6-methoxy-2-naphthyl) propanoic acid have been shown to possess good nonsteroidal anti-inflammatory (NSAI) activity. Keeping in view of the useful findings of 6-methoxy-2-naphthyl group and in continuation of our work on 4,6-diaryl-2-oxo-cyclohex-3-ene-1-carboxylates (Sreevidya *et al.*, 2009), we have synthesized some new cyclohexenone derivatives. We have already reported the crystal and molecular structure studies of some of the related chalcones and cyclohexenone derivatives (Li *et al.*, 2009, 2009a; Jasinski *et al.*, 2009; Yathirajan, Mayekar, Narayana *et al.*, 2007; Yathirajan, Mayekar, Sarojini *et al.*, 2007; Yathirajan, Sarojini, Bindya *et al.*, 2006; Yathirajan, Narayana, Ashalatha *et al.*, 2006). The present paper reports the synthesis, characterization and antimicrobial activity of some new cyclohexenone derivatives derived from 6-methoxy-2-naphthylaldehyde. One of the newly synthesized compound, viz., ethyl 4-(4-chlorophenyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (**4f**) is characterized by single crystal studies.

## 2. Experimental

TLC was run on a Merck silica gel 60 F254 coated aluminum plates and melting points were taken in open capillary tubes and are uncorrected. Elemental analysis was carried out using Flash EA 1112 Series, CHNSO Analyzer (Thermo). IR spectra in KBr pellets were recorded on Jasco FT/IR-4100 FTIR spectrophotometer.  $^1\text{H-NMR}$  spectra were recorded in  $\text{CDCl}_3$  and in  $\text{DMSO-d}_6$  on a Bruker (400 MHz) spectrometer using TMS as internal standard and Mass spectra were recorded on LC/MSD Trap XCT spectrometer. Bruker SMART CCD diffractometer was used for crystal structure studies.

### 2.1 General procedure for preparation of 1-(aryl)-3-(6-methoxynaphthalen-2-yl)prop-2-en-1-one (**3a-3l**)

To a thoroughly stirred solution of 6-methoxy-2-naphthaldehyde (1.86 g, 10 mmol) and substituted acetophenones (10 mmol) in 15 ml methanol, 5 ml of 40% KOH solution was added. The reaction mixture was stirred overnight and the solid separated was collected by filtration. The product obtained was recrystallized from methanol.

#### (2E)-1-(2-Hydroxyphenyl)-3-(6-methoxy-2-naphthyl)prop-2-en-1-one (**3b**)

Yellow crystals, Yield 70%, M.p.: 216-218 °C, Anal. Calculated %  $\text{C}_{20}\text{H}_{16}\text{O}_2$ : C-78.93, H-5.29. Found C-78.90, H-5.26. IR (KBr  $\nu_{\text{max}}$   $\text{cm}^{-1}$ ): 1650 (C=O), 1630 (C=C) and 3400 (OH);  $^1\text{H NMR}$  (DMSO,  $\delta$  ppm): 3.88 (s, 3H, -OCH<sub>3</sub>), 6.89 (d, 2H,  $J = 8.5$  Hz, ArH), 7.20 (d, 1H,  $J = 6.7$  Hz, ArH), 7.36 (d, 1H,  $J = 2.1$  Hz, ArH), 7.79 (d, 1H,  $J = 15.5$  Hz, CH=C), 7.86 (m, 2H, ArH), 7.97 (d, 1H,  $J = 15.5$  Hz, CH=C), 8.02-8.10 (m, 3H, ArH), 8.22 (s, 1H, ArH), 10.41 (s, 1H, OH). LCMS: 305 (M+1).

#### (2E)-1-(4-chlorophenyl)-3-(6-methoxy-2-naphthyl)prop-2-en-1-one (**3f**)

Yellow crystals, Yield 78%, M.p.: 200-202 °C, Anal. Calculated %  $\text{C}_{20}\text{H}_{15}\text{ClO}_2$ : C-74.42, H-4.68. Found C-74.38, H-4.63. IR (KBr  $\nu_{\text{max}}$   $\text{cm}^{-1}$ ): 1695 (C=O) and 1660 (C=C);  $^1\text{H NMR}$  (DMSO,  $\delta$  ppm): 3.89 (s, 3H, -OCH<sub>3</sub>), 7.19 (d, 1H,  $J = 8.5$  Hz, ArH), 7.38 (d, 1H,  $J = 2.2$  Hz, ArH), 7.64 (d, 2H,  $J = 8.6$  Hz, ArH), 7.84-7.90 (m, 4H, ArH), 8.02 (d, 1H,  $J = 15.1$  Hz, CH=C), 8.20 (d, 2H,  $J = 8.5$  Hz, ArH), 8.26 (s, 1H, ArH). LCMS: 323 (M+1).

#### (2E)-3-(6-Methoxy-2-naphthyl)-1-(4-nitrophenyl)prop-2-en-1-one (**3g**)

Yellow crystals, Yield 78%, M.p.: 178-180 °C, Anal. Calculated % C<sub>20</sub>H<sub>15</sub>NO<sub>4</sub>: C-70.06, H-4.53, N-4.20. Found C-70.01, H-4.49, N-4.18. IR (KBr  $\nu_{\max}$  cm<sup>-1</sup>): 1686 (C=O) and 1643 (C=C); <sup>1</sup>H NMR (DMSO,  $\delta$  ppm): 3.89 (s, 3H, -OCH<sub>3</sub>), 7.23 (d, 1H, *J* = 8.9 Hz, ArH), 7.38 (d, 1H, *J* = 2.2 Hz, ArH), 7.87 (d, 1H, *J* = 15.1 Hz, CH=C), 7.97 (d, 1H, *J* = 15.4 Hz, CH=C), 8.06 (d, 2H, *J* = 6.9 Hz, ArH), 8.10 (s, 1H, ArH), 8.29 (s, 1H, ArH), 8.38 (m, 4H, ArH). LCMS: 334 (M+1).

*(2E)-3-(6-Methoxy-2-naphthyl)-1-(1-naphthyl)prop-2-en-1-one (3j)*

Yellow crystals, Yield 70%, M.p.: 192-194 °C, Anal. Calculated % C<sub>24</sub>H<sub>18</sub>O<sub>2</sub>: C-85.18, H-5.02. Found C-85.15, H-5.06. IR (KBr  $\nu_{\max}$  cm<sup>-1</sup>): 1682 (C=O) and 1641 (C=C); <sup>1</sup>H NMR (DMSO,  $\delta$  ppm): 3.90 (s, 3H, -OCH<sub>3</sub>), 7.22 (dd, 1H, *J* = 6.7 Hz, ArH), 7.40 (s, 1H, ArH), 7.64-7.69 (m, 2H, ArH), 7.73 (d, 1H, *J* = 15.9 Hz, CH=C), 7.89-7.92 (m, 2H, ArH), 7.99 (d, 1H, *J* = 15.5 Hz, CH=C), 8.04-8.21 (m, 6H, ArH), 8.29 (s, 1H, ArH), 8.9 (s, 1H, ArH). LCMS: 339 (M+1).

*(2E)-1-(3-Bromophenyl)-3-(6-methoxy-2-naphthyl)prop-2-en-1-one (3k)*

Yellow crystals, Yield 78 %, M.p.: 154-156 °C, Anal. Calculated % C<sub>20</sub>H<sub>15</sub>BrO<sub>2</sub>: C-65.41, H-4.11. Found C-65.38, H-4.07. IR (KBr  $\nu_{\max}$  cm<sup>-1</sup>): 1670 (C=O) and 1640 (C=C); <sup>1</sup>H NMR (DMSO,  $\delta$  ppm): 3.89 (s, 3H, -OCH<sub>3</sub>), 7.22 (dd, 1H, *J* = 6.7 Hz, ArH), 7.31-7.38 (m, 3H, ArH), 7.82 (s, 1H, ArH), 7.86-7.89 (m, 2H, ArH), 7.94 (d, 1H, *J* = 14.6 Hz, CH=C), 8.05-8.08 (m, 2H, ArH and CH=C), 8.25 (s, 1H, ArH), 8.35 (s, 1H, ArH). LCMS: 368 (M+1).

2.1.1 General procedure for preparation of Cyclohexenones (**4a-4l**)

1-(Aryl)-3-(6-methoxynaphthalen-2-yl)prop-2-en-1-one (5 mmol) and ethyl acetoacetate (5 mmol) were refluxed for 4-6 hrs in 15 ml ethanol in presence of 0.8 ml of 10% NaOH. The reaction mixture was cooled to room temperature and the reaction mass was filtered. The compound was recrystallized from methanol.

*Ethyl 4-(2-hydroxyphenyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (4b)*

Off white crystals, IR (KBr  $\nu_{\max}$  cm<sup>-1</sup>): 1632 cm<sup>-1</sup> (C=O ketone), 1742 cm<sup>-1</sup> (C=O ester) 3411 cm<sup>-1</sup> (-OH); <sup>1</sup>H NMR (CDCl<sub>3</sub>, 400MHz,  $\delta$  ppm): 1.00 (t, 3H, *J* = 6.8 Hz, CH<sub>2</sub>-CH<sub>3</sub>), 2.97-3.18 (m, 2H, CH<sub>2</sub>, CH<sub>2</sub>-CHAr), 3.86 (s, 1H, CH), 3.90 (s, 1H, CH), 3.93 (s, 3H, OCH<sub>3</sub>), 4.0 (q, 2H, *J* = 6.0 Hz, CH<sub>2</sub>-CH<sub>3</sub>), 5.4 (bs, 1H, OH), 6.55 (s, 1H, =CH-CO-), 6.86 (d, 2H, *J*=8.7 Hz, ArH), 7.13-7.17 (m, 2H, ArH), 7.42 (d, 1H, *J*=8.4 Hz, ArH), 7.49 (d, 2H, *J*=8.7 Hz, ArH), 7.69-7.75 (m, 3H, ArH). LCMS: 417 (M+1).

*Ethyl 4-(4-hydroxyphenyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (4c)*

Off white crystals, IR (KBr  $\nu_{\max}$  cm<sup>-1</sup>): 1609 cm<sup>-1</sup> (C=O ketone), 1730 cm<sup>-1</sup> (C=O ester) 3376 cm<sup>-1</sup> (-OH); <sup>1</sup>H NMR (CDCl<sub>3</sub>, 400MHz,  $\delta$  ppm): 0.99 (t, 3H, *J* = 6.8 Hz, CH<sub>2</sub>-CH<sub>3</sub>), 3.11-3.14 (m, 2H, CH<sub>2</sub>, CH<sub>2</sub>-CHAr), 3.91 (s, 3H, OCH<sub>3</sub>), 3.93(s, 1H, CH), 3.94 (s, 1H, CH), 4.0 (q, 2H, *J* = 6.0 Hz, CH<sub>2</sub>-CH<sub>3</sub>), 6.76 (s, 1H, =CH-CO-), 6.81 (bs, 1H, OH), 6.90 (d, 2H, *J*=6.4 Hz, ArH), 7.10-7.14 (m, 2H, ArH), 7.20-7.27 (m, 2H, ArH), 7.40 (dd, 1H, *J*=6.8 Hz, ArH), 7.67 (d, 2H, *J*=8.8 Hz, ArH), 7.70 (s, 1H, ArH). LCMS: 417(M+1).

*Ethyl 4-(4-methoxyphenyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (4d)*

White crystals, IR (KBr  $\nu_{\max}$  cm<sup>-1</sup>): 1666 cm<sup>-1</sup> (C=O ketone), 1743 cm<sup>-1</sup> (C=O ester); <sup>1</sup>H NMR (CDCl<sub>3</sub>,  $\delta$  ppm): 0.98 (t, 3H, *J* = 7.0 Hz, CH<sub>2</sub>-CH<sub>3</sub>), 3.02-3.19 (m, 2H, CH<sub>2</sub>, CH<sub>2</sub>-CHAr), 3.84 (s, 4H, OCH<sub>3</sub> and CH), 3.88 (s, 1H, CH), 3.97 (s, 3H, OCH<sub>3</sub>), 3.99 (q, 2H, *J* = 6.9 Hz, CH<sub>2</sub>-CH<sub>3</sub>), 6.55 (s, 1H, =CH-CO-), 6.92 (d, 2H, *J*=8.8 Hz, ArH), 7.12-7.17 (m, 2H, ArH), 7.41 (d, 1H, *J* = 7.2 Hz, ArH), 7.53 (d, 2H, *J* = 8.7 Hz, ArH), 7.71 (d, 2H, *J* = 9.7 Hz, ArH), 7.75 (s, 1H, ArH). LCMS: 431(M+1).

*Ethyl 4-(4-methylphenyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (4e)*

White crystals, IR (KBr  $\nu_{\max}$  cm<sup>-1</sup>): 1634 cm<sup>-1</sup> (C=O ketone), 1735 cm<sup>-1</sup> (C=O ester); <sup>1</sup>H NMR (CDCl<sub>3</sub>, 400MHz,  $\delta$  ppm): 1.00 (t, 3H, *J* = 7.0 Hz, CH<sub>2</sub>-CH<sub>3</sub>), 2.39 (s, 3H, CH<sub>3</sub>), 3.06-3.19 (m, 2H, CH<sub>2</sub>, CH<sub>2</sub>-CHAr), 3.87 (s, 1H, CH), 3.91(s, 1H, CH), 3.94 (s, 3H, OCH<sub>3</sub>), 4.0 (q, 2H, *J* = 6.9 Hz, CH<sub>2</sub>-CH<sub>3</sub>), 6.59 (s, 1H, =CH-CO-), 7.13-7.26 (m, 4H, ArH), 7.41-7.48 (m, 3H, ArH), 7.72-7.75 (m, 3H, ArH). LCMS: 415(M+1).

*Ethyl 4-(4-chlorophenyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (4f)*

White crystals, IR (KBr  $\nu_{\max}$  cm<sup>-1</sup>): 1651 cm<sup>-1</sup> (C=O ketone), 1758 cm<sup>-1</sup> (C=O ester); <sup>1</sup>H NMR (CDCl<sub>3</sub>,  $\delta$  ppm): 0.98 (t, 3H, *J* = 6.8 Hz, CH<sub>2</sub>-CH<sub>3</sub>), 3.05 (m, 2H, CH<sub>2</sub>, CH<sub>2</sub>-CHAr), 3.84 (s, 1H, CH), 3.88 (s, 1H, CH), 3.90 (s, 3H, OCH<sub>3</sub>), 3.99 (q, 2H, *J* = 6.9 Hz, CH<sub>2</sub>-CH<sub>3</sub>), 6.54 (s, 1H, =CH-CO-), 7.12-7.16 (m, 3H, ArH), 7.38 (d, 3H, *J*=8.0 Hz, ArH), 7.47 (d, 2H, *J* = 8.1 Hz, ArH), 7.67-7.74 (m, 2H, ArH). LCMS: 435(M+1), 436(M+2).

*Ethyl 4-(4-nitrophenyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (4g)*

Yellow crystals, IR (KBr  $\gamma_{\max}$   $\text{cm}^{-1}$ ): 1658  $\text{cm}^{-1}$  (C=O ketone), 1761  $\text{cm}^{-1}$  (C=O ester);  $^1\text{H}$  NMR ( $\text{CDCl}_3$ ,  $\delta$  ppm): 0.98 (t, 3H, J= 6.8 Hz,  $\text{CH}_2\text{-CH}_3$ ), 3.05 (m, 2H,  $\text{CH}_2$ ,  $\text{CH}_2\text{-CHAr}$ ), 3.84-3.99 (m, 5H,  $\text{OCH}_3$  and 2CH), 4.01 (q, 2H, J= 6.9 Hz,  $\text{CH}_2\text{-CH}_3$ ), 6.54 (s, 1H, =CH-CO-), 7.12-7.16(m, 3H, ArH), 7.38 (d, 3H, J=8.0 Hz, ArH), 7.47 (d, 2H, J= 8.1 Hz, ArH), 7.67-7.74 (m, 2H, ArH). LCMS: 446 (M+1).

*Ethyl 4-(pyridin-4-yl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (4h)*

Yellow crystals, IR (KBr  $\gamma_{\max}$   $\text{cm}^{-1}$ ): 1638  $\text{cm}^{-1}$  (C=O ketone), 1742  $\text{cm}^{-1}$  (C=O ester);  $^1\text{H}$  NMR ( $\text{CDCl}_3$ ,  $\delta$  ppm): 0.98 (t, 3H, J= 6.8 Hz,  $\text{CH}_2\text{-CH}_3$ ), 3.11-3.18 (m, 2H,  $\text{CH}_2$ ,  $\text{CH}_2\text{-CHAr}$ ), 3.88 (s, 1H, CH), 3.90 (s, 4H,  $\text{OCH}_3$  and CH), 3.99 (q, 2H, J= 6.9 Hz,  $\text{CH}_2\text{-CH}_3$ ), 6.40 (s, 1H, =CH-CO-), 7.11-7.17 (m, 2H, ArH), 7.37-7.55 (m, 5H, ArH), 7.70-7.73 (m, 3H, ArH). LCMS: 402(M+1).

*Ethyl 4-(1-naphthyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (4j)*

White powder, IR (KBr  $\gamma_{\max}$   $\text{cm}^{-1}$ ): 1651  $\text{cm}^{-1}$  (C=O ketone), 1758  $\text{cm}^{-1}$  (C=O ester);  $^1\text{H}$  NMR ( $\text{CDCl}_3$ ,  $\delta$  ppm): 1.01 (t, 3H, J= 7.2 Hz,  $\text{CH}_2\text{-CH}_3$ ), 3.29 (m, 2H,  $\text{CH}_2$ ,  $\text{CH}_2\text{-CHAr}$ ), 3.92 (s, 3H,  $\text{OCH}_3$ ), 3.93 (s, 1H, CH), 3.95 (s, 1H, CH), 4.03 (q, 2H, J= 6.9 Hz,  $\text{CH}_2\text{-CH}_3$ ), 6.74 (s, 1H, =CH-CO-), 7.13-7.17(m, 2H, ArH), 7.68 (dd, 1H, J=8.8 Hz, ArH), 7.70 (s, 1H, ArH), 7.72 (d, 3H, J=8.1, ArH) 7.76 (d, 2H, J= 8.4 Hz, ArH), 7.82-7.88 (m, 3H, ArH), 8.02 (s, 1H, ArH). LCMS: 451(M+1).

*Ethyl 4-(3-bromophenyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (4k)*

Yellow crystals, IR (KBr  $\gamma_{\max}$   $\text{cm}^{-1}$ ): 1661  $\text{cm}^{-1}$  (C=O ketone), 1754  $\text{cm}^{-1}$  (C=O ester);  $^1\text{H}$  NMR ( $\text{CDCl}_3$ , 400MHz,  $\delta$  ppm): 0.99 (t, 3H, J= 7.0 Hz,  $\text{CH}_2\text{-CH}_3$ ), 3.04-3.08 (m, 2H,  $\text{CH}_2$ ,  $\text{CH}_2\text{-CHAr}$ ), 3.89 (s, 1H, CH), 3.90 (s, 1H, CH), 3.91 (s, 3H,  $\text{OCH}_3$ ), 4.01 (q, 2H, J= 6.0 Hz,  $\text{CH}_2\text{-CH}_3$ ), 6.55 (s, 1H, =CH-CO-), 7.12 (d, 1H, J= 2.8 Hz, ArH), 7.15 (d, 1H, J=8.6 Hz, ArH), 7.29 (d, 1H, J=8.0 Hz, ArH), 7.40 (dd, 1H, J=8.4 Hz, ArH), 7.48 (d, 1H, J=8.0Hz, ArH), 7.55 (d, 1H, J=7.6Hz, ArH), 7.66-7.68 (m, 2H, ArH), 7.71 (s, 1H, ArH), 7.72 (s, 1H, ArH), 7.74 (s, 1H, ArH). LCMS: 480(M+1), 481(M+2)

*Ethyl 4-(2,4-dichlorophenyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (4l)*

Off white crystals, IR (KBr  $\gamma_{\max}$   $\text{cm}^{-1}$ ): 1665  $\text{cm}^{-1}$  (C=O ketone), 1743  $\text{cm}^{-1}$  (C=O ester);  $^1\text{H}$  NMR ( $\text{CDCl}_3$ , 400MHz,  $\delta$  ppm): 1.00 (t, 3H, J= 7.0 Hz,  $\text{CH}_2\text{-CH}_3$ ), 2.95-3.05 (m, 2H,  $\text{CH}_2$ ,  $\text{CH}_2\text{-CHAr}$ ), 3.89 (s, 1H, CH), 3.91 (s, 3H,  $\text{OCH}_3$ ), 3.92 (s, 1H, CH), 4.02 (q, 2H, J= 6.0 Hz,  $\text{CH}_2\text{-CH}_3$ ), 6.21 (s, 1H, =CH-CO-), 7.0 (d, 1H, J= 2.8 Hz, ArH), 7.13-7.17 (m, 2H, ArH), 7.19 (s, 1H, ArH), 7.28-7.30 (m, 2H, ArH), 7.44 (d, 1H, J= 2.0 Hz, ArH), 7.66 (s, 1H, ArH), 7.70 (s, 1H, ArH). LCMS: 470(M+1), 471(M+2).

## 2.2 Pharmacology

### 2.2.1 Antibacterial studies

The newly synthesized compounds were screened for their antibacterial activity against *Escherichia coli* (ATTC-25922), *Staphylococcus aureus* (ATTC-25923), *Pseudomonas aeruginosa* (ATCC-27853) and *Klebsiella pneumoniae* (recultured) bacterial stains by serial plate dilution method (Barry, 1991; James, 1970). Serial dilutions of the drug in Mueller Hinton broth were taken in tubes and their pH was adjusted to 5.0 using phosphate buffer. A standardized suspension of the test bacterium was inoculated and incubated for 16-18 h at 37°C. The minimum inhibitory concentration (MIC) was noted by seeing the lowest concentration of the drug at which there was no visible growth.

A number of antimicrobial discs were placed on the agar for the sole purpose of producing zones of inhibition in the bacterial lawn. Twenty milliliters of agar media was poured into each Petri dish. Excess of suspension was decanted and plates were dried by placing in an incubator at 37°C for an hour. Using a punch, wells were made on these seeded agar plates and minimum inhibitory concentrations of the test compounds in dimethylsulfoxide (DMSO) were added into each labeled well. A control was also prepared for the plates in the same way using solvent DMSO. The Petri dishes were prepared in triplicate and maintained at 37°C for 3-4 days. Antibacterial activity was determined by measuring the diameter of inhibition zone. Activity of each compound was compared with ampicillin as standard. Zone of inhibition was determined for newly synthesized compounds and the results are summarized in **Table 2**.

### 2.2.2 Antifungal studies

Newly prepared compounds were screened for their antifungal activity against *Aspergillus flavus* (NCIM No.524), *Aspergillus fumigates* (NCIM No. 902), *Penicillium marneffeii* (recultured) and *Trichophyton mentagrophytes* (recultured) in DMSO by serial plate dilution method (Arthington-skaggs 2000; Verma, 1998). Sabourands agar media was prepared by dissolving peptone (1 g), D-glucose (4 g) and agar (2 g) in distilled water (100 ml) and adjusting the pH to 5.7. Normal saline was used to make a suspension of spore of fungal strains for lawning. A

loopful of particular fungal strain was transferred to 3 ml saline to get a suspension of corresponding species. Twenty milliliters of agar media was poured into each Petri dish. Excess of suspension was decanted and plates were dried by placing in incubator at 37°C for 1 h. Using a punch, wells were made on these seeded agar plates minimum inhibitory concentrations of the test compounds in DMSO were added into each labeled well. A control was also prepared for the plates in the same way using solvent DMSO. The Petri dishes were prepared in triplicate and maintained at 37°C for 3-4 days. Antifungal activity was determined by measuring the diameter of inhibition zone. Activity of each compound was compared with itraconazole as standard. Zones of inhibition were determined and the results are summarized in **Table 3**.

### 3. Results and discussion

Chalcones were synthesized by a base catalyzed Claisen–Schmidt condensation reaction of 6-methoxy-2-naphthaldehyde and substituted acetophenones. The reaction of chalcones with ethyl acetoacetate is known to lead three structurally diverse types of compounds, depending on the experimental conditions employed. Thus in the presence of a base, chalcones (**3a-3i**) and ethyl acetoacetate produce cyclohexenones (**4a-4i**) by means of an intermediate Michael adduct, as given in **Scheme 1**. The cyclocondensation of ethyl acetoacetate with chalcones leads to the generation of two chiral centers in the structure of cyclohexenones and both configuration of chiral carbon atoms are expected in the synthesized cyclohexenones, which would result in a mixture of diastereomers. No attempt has been undertaken to separate the diastereomeric cyclohexenones and have been characterized as such. The newly synthesized compounds were characterized by elemental analysis, IR, <sup>1</sup>H NMR, LCMS and single crystal X-ray analysis. The elemental analysis data of cyclohexenone derivatives are reported in **Table 1**.

The IR spectra of (2*E*)-1-(2-hydroxyphenyl)-3-(6-methoxy-2-naphthyl)prop-2-en-1-one (**3b**) revealed a strong band at 3400 cm<sup>-1</sup> confirming the presence of –OH group and a sharp band at 1650 cm<sup>-1</sup> indicating the presence of C=O. The proton NMR showed a singlet at δ 3.88 ppm representing three protons of methoxy group. The peaks of CH<sub>α</sub>=CH<sub>β</sub> were observed at δ 7.79 ppm as doublet with *J* = 15.48 Hz and at δ 7.97 ppm as doublet with *J* = 15.5 Hz, respectively. The LCMS showed the peak at *m/z* 305 (M+H), confirming the structure of the compound.

The IR spectra of ethyl 4-(2-hydroxyphenyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (**4b**) revealed a strong sharp band at 1632 cm<sup>-1</sup> representing C=O group of the cyclohexenone ring and the band for C=O of ester was observed at 1742 cm<sup>-1</sup>.

In the <sup>1</sup>H NMR spectra, the CH<sub>3</sub> protons of ester group came into resonance at δ 1.0 as triplet with *J* = 6.8 Hz. The CH<sub>2</sub> protons of cyclohexenone ring were observed as multiplets at δ 2.97-3.18 ppm and two CH proton of cyclohexenone ring resonated at δ 3.86 ppm and at δ 3.90 ppm as singlet. The OCH<sub>3</sub> protons resonated as singlet at δ 3.93 ppm accounting for three protons. The CH<sub>2</sub> protons of ester group resonated as quartet at δ 4.0 ppm with *J* = 6.0 Hz. A broad singlet was observed at δ 5.4 ppm representing OH proton. The signal for =CH-CO- of cyclohexenone ring appeared at δ 6.55 ppm as singlet. The aromatic protons appeared at δ 6.86 ppm a doublet with *J* = 8.7 Hz accounting for two protons. Two protons came into resonance as multiplet at δ 7.13-7.17 ppm. Doublets were observed at δ 7.42 ppm and 7.49 ppm with *J* = 8.4 Hz and *J* = 8.7 Hz respectively representing two aromatic protons. Three aromatic protons resonated as multiplets at δ 7.69-7.75 ppm. The LCMS of this compound showed the M+1 Peak at *m/z* 417.

Compound **4f** was obtained as colourless blocks by recrystallisation from acetonitrile solution and its structure was elucidated by single crystal X-ray diffraction. Molecular structure of ethyl 4-(4-chlorophenyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (**4f**) is shown in **Fig. 1**. C<sub>26</sub>H<sub>23</sub>ClO<sub>4</sub>, *M<sub>r</sub>* = 434.89, monoclinic, *P*2(1)/*c*, *a* = 18.5546 (8) Å, *b* = 11.4501 (5) Å, *c* = 10.2810 (5) Å, β = 93.454 (1)°, *V* = 2180.25 (17) Å<sup>3</sup>, *Z* = 4, ρ<sub>calc</sub> = 1.325 g cm<sup>-3</sup>, μ = 0.21 mm<sup>-1</sup>, *F*(000) = 912, Bruker SMART CCD diffractometer, Mo Kα radiation, λ = 0.71073 Å, *T* = 296 K, *R*(*F*) = 0.054, *wR*(*F*<sup>2</sup>) = 0.172, *S* = 1.03. Atoms C9 and C10 and their attached H atoms are disordered over two sets of sites on a 0.745 (8):0.255 (8) ratio. The dihedral angle between the naphthalene and benzene rings is 70.75 (7)°. Atom C23 deviates from the naphthalene ring mean plane by a negligible amount [0.011 (6) Å]. Atoms C9 and C10 are disordered over two sites in a 0.745 (8):0.255 (8) ratio. The major orientation corresponds to the *S,S*-enantiomer and the minor orientation to the *R,R*-enantiomer: crystal symmetry generates a racemic mixture. In the crystal, a weak intermolecular C—H⋯O interaction occurs. Similar disorder of the equivalent atoms have been seen in related structures (Li *et al.*, 2009, 2009a). All the H atoms were placed in idealized locations (C—H = 0.93-0.98 Å) and refined as riding with *U*<sub>iso</sub>(H) = 1.2*U*<sub>eq</sub>(C) or 1.5*U*<sub>eq</sub>(methyl C). The methyl groups were allowed to rotate, but not to tip, to best fit the electron density. [X-ray crystallographic files, in CIF format, for the structure

determinations of (I) (CCDC 769575) has been deposited with the Cambridge Crystallographic Data Centre, CCDC: 26091. Copies of this information may be obtained free of charge from the Director, CCDC, 12 Union Road, Cambridge, CB2 1EZ (fax: +44-1223-336033; email: deposit@ccdc.cam.ac.uk or <http://www.ccdc.cam.ac.uk>.]

All the newly synthesized compounds were screened for their antimicrobial activity. The investigation of antibacterial and antifungal screening data revealed that some of the compounds (**4a-l**) showed moderate to good inhibition at  $\mu\text{g ml}^{-1}$  in DMSO. The compounds **4k** and **4l** showed comparatively good activity against all the bacterial and fungal strains. The good activity is attributed to the presence of pharmacologically active group attached to the cyclohexenone ring. Introduction of aryl moiety carrying bromo and dichloro group enhanced activity compared to the standard against *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Penicillium marneffeii*, *T. mentagrophytes*, *A. flavus* and *A. fumigatus*. Results of antibacterial and antifungal screening are discussed in **Table-2** and **Table-3** respectively.

#### 4. Conclusion

The research study reports the successful synthesis of chalcones derived from 6-methoxy-2-naphthaldehyde. These chalcones were converted to cyclohexenone derivatives by treating with ethyl acetoacetate in presence of a base, which are useful intermediates in the synthesis of heterocyclic compounds. The newly synthesized cyclohexenone derivatives were tested for their antibacterial and antifungal activity. Some of these derivatives have shown promising results. Ethyl 4-(4-chlorophenyl)-6-(6-methoxy-2-naphthyl)-2-oxo-cyclohex-3-ene-1-carboxylate (**4f**) was studied by single crystal X-ray diffraction.

#### Acknowledgements

ANM is thankful to University of Mysore for research facilities and SeQuent Scientific Ltd for allowing to undertake research. HSY thanks University of Mysore for sabbatical leave.

#### References

- Arthington-Skaggs, B. A., Motley, M., Warnock, D.W., Morrison, C.J. (2000). Comparative Evaluation of PASCO and National Committee for Clinical laboratory standards M27-A broth micro dilution methods for antifungal drug susceptibility testing of yeasts. *Journal of Clinical Microbiology*, 38:2254-2260.
- Asiri, A.M. (2003). Synthesis and Absorption Spectral Properties of Bis-methine Dyes Exemplified by 2,5-Bis-arylidene-1-dicyanomethylene-cyclopentanes. *Bulletin of the Korean Chemical Society*, 24:426-430.
- Barry, A. L. (1991). Procedure for testing antimicrobial agents in agar media, in: VL Corian (Ed), *Antibiotics in Laboratory Medicine*, Williams and Wilkins, Baltimore, MD, 1-16.
- Damazio, R.G., Zanatta, A.P., Cazarolli, L.H., Chiaradia, L.D., Mascarello, A., Nunes, R.J., Yunes, R.A., Barreto Silva, F.R. (2010). Antihyperglycemic activity of naphthylchalcones. *European Journal of Medicinal Chemistry*, 45:1332-1337.
- Dhar, D.N. (1981). *The Chemistry of Chalcones and Related Compounds*, Wiley, New York, p 213.
- Ferrer, R., Lobo, G., Gamboa, N., Rodrigues, J., Abramjuk, C., Jung, K., Lein, M., Charris, J.E. (2009). Synthesis of [(7-chloroquinolin-4-yl)amino]chalcones: Potential antimalarial and anticancer agents. *Scientia Pharmaceutica*, 77:725-741.
- Forestier, S, Moire, C., Lang, G. (1989). Cosmetic composition containing hydroxylated chalcone derivatives and its use for protecting the skin and the hair against luminous radiations, new hydroxylated chalcone derivatives employed and process for their preparation. US Patent 4,867,964.
- James, D., Lowry, M., Jaqua, M. J., Selepak, S. T. (1970). Detailed Methodology and Implementation of a Semi automated Serial Dilution Micro technique for Antimicrobial Susceptibility Testing. *Applied Microbiology*, 20: 46-53.
- Jasinski, J.P., Butcher, R.J., Mayekar, A.N., Yathirajan, H.S., Narayana, B. (2009). Structures of three chalcones derived from 6-methoxy-2-naphthaldehyde. *Journal of Chemical Crystallography*, 39:157-162.
- Kumar, S.K, Hager, E., Catherine, P., Gurulingappa, H., Davidson, N.E., Khan, S.R. (2003). Synthesis and evaluation of novel boronic-chalcone derivatives as antitumor agents. *Journal of Medicinal Chemistry*, 46:2813-2815.
- Li, H., Mayekar, A.N., Narayana, B., Yathirajan, H.S., Harrison, W.T.A. (2009a). Ethyl 6-(6-methoxy-2-naphthyl)-2-oxo-4-(2-thienyl)cyclohex-3-ene-1-carboxylate. *Acta Crystallographica*, E65:o1533.

- Li, H., Mayekar, A.N., Narayana, B., Yathirajan, H.S., Harrison, W.T.A. (2009). ( $\pm$ )-Ethyl 6-(6-methoxy-2-naphthyl)-4-(4-methylphenyl)-2-oxocyclohex-3-ene-1-carboxylate. *Acta Crystallographica* E65:o1186.
- Modzelewska, A., Pettit, C., Achanta, G., Davidson, N.E., Huang, P., Khan, S.R. (2006). Anticancer activities of novel chalcone and bis-chalcone derivatives. *Bioorganic and Medicinal Chemistry*, 14:3491-3495.
- Padmavathi, V., Reddy, B.J.M., Balaiah, A., Reddy, K.V., Reddy, D.B. (2000). Synthesis of some fused pyrazoles and isoxazoles. *Molecules*, 5:1281-1286.
- Rajendra Prasad, Y., Ravi Kumar, P., Asha Deepti, C.H., Venkata Ramana, M. (2006). Synthesis and antimicrobial activity of some novel chalcones of 2-hydroxy-1-acetonaphthone and 3-acetyl coumarin *E-Journal of Chemistry*, 3:236-241.
- Senguttuvan, S., Nagarajan, S. (2010). Synthesis of 2-Amino-5-aryl-5,6-dihydro-7-(naphthalene-2-yl) Quinazolin-4-ols. *International Journal of Chemistry*, 2: 108-112.
- Shettigar, S., Chandrasekharan, K., Umesh, Sarojini, B.K., Narayana, B. (2006). Studies on nonlinear optical parameters of bis-chalcone derivatives doped polymer. *Polymer*, 47:3565-3567.
- Sreevidya, T.V., Narayana, B., Yathirajan, H.S. (2009). Synthesis and characterization of some chalcones and their cyclohexenone derivatives. *Central European Journal of Chemistry*, 8:174-181.
- Sukumaran, K., Kuttan, R. (1991). Screening of 11 ferns of cytotoxic and antitumor potential with special reference to *Pityrogramma calomelanos*. *Journal of Ethnopharmacology*, 36:93-96.
- Suksamrarn, A., Poomsing, P., Aroonrerk, N., Punjanon, T., Suksamrarn, S., Kongkun, S. (2003). Antimycobacterial and antioxidant flavones from *Limnophila geoffrayi*. *Archives of Pharmacal Research*, 26:816-820.
- Tanaka, M., Nara, F., Suzuki, K., Hosoya, T., Ogita, T. (1997). Structural elucidation of sciphostatin, an inhibitor of membrane-bound neutral sphingomyelinase. *Journal of the American Chemical Society*, 119: 7871-7872.
- Torigoo, T., Arisawa, M., Iloch, S., Fujiu, M., Mayuyama, H.B. (1983). Anti-mutagenic chalcones: Antagonizing the mutagenicity of benzo(a)pyrene in *Salmonella typhimurium*. *Biophysical Research Communications*, 112:833-842.
- Verma, R. S., Khan, Z. K., Singh, A. P. (1998). (Ed), Antifungal Agents: Past, Present and Future Prospects, National Academy of Chemistry and Biology, Lucknow, India, 55-128.
- Vyas, D.H., Tala, S.D., Akbari, J.D., Dhaduk, M.F., Joshi, H.S. (2009). Synthesis, antimicrobial and antitubercular activity of some cyclohexenone and indazole derivatives. *Indian Journal of Chemistry*, 48B: 1405-1410.
- Yathirajan, H.S., Mayekar, A.N., Narayana, B., Sarojini, B.K., Bolte, M. (2007). (2E)-1-(2,4-Dichlorophenyl)-3-(6-methoxy-2-naphthyl)prop-2-en-1-one. *Acta Crystallographica*, E63:o540.
- Yathirajan, H.S., Mayekar, A.N., Sarojini, B.K., Narayana, B., Bolte, M. (2007). (2E)-3-(6-Methoxy-2-naphthyl)-1-phenylprop-2-en-1-one. *Acta Crystallographica*, E63:o1012.
- Yathirajan, H S, Sarojini, B.K., Bindya, S., Narayana, B., Bolte, M. (2006). 3-(6-Methoxy-2-naphthyl)-1-(2-naphthyl)prop-2-en-1-one. *Acta Crystallographica*, E62:o4046.
- Yathirajan, H.S., Narayana, B., Ashalatha, B.V., Sarojini, B.K., Bolte, M. (2006). 3-(6-Methoxy-2-naphthyl)-1-(2-thienyl)prop-2-en-1-one. *Acta Crystallographica*, E62:o4440.

Table 1. Characterization data of compounds (4a-4l)


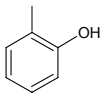

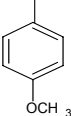
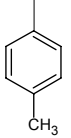
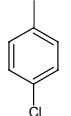
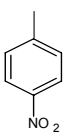
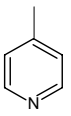
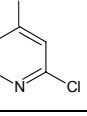
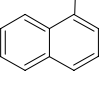
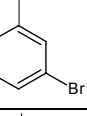
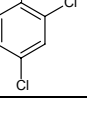
Compd	R	m.p (°C)	Yield %	Elemental analysis		
				% found (calculated)		
				C	H	N
4a		152-154	70	77.94 (77.98)	6.02 (6.04)	-
4b		172-174	56	74.91 (74.98)	5.01 (5.08)	-
4c		182-184	62	74.92 (74.98)	5.04 (5.080)	-
4d		148-150	60	75.30 (75.33)	6.02 (6.08)	-
4e		162-164	68	78.21 (78.24)	6.30 (6.32)	-
4f		150-152	72	71.78 (71.80)	5.30 (5.33)	-
4g		120-122	50	70.11 (70.10)	5.18 (5.20)	3.10 (3.14)
4h		202-204	58	74.76 (74.79)	5.71 (5.72)	3.45 (3.48)
4i		178-180	51	68.84 (68.880)	5.06 (5.08)	3.18 (3.21)
4j		124-126	60	79.95 (79.98)	5.79 (5.81)	
4k		146-148	68	65.12 (65.14)	4.81 (4.83)	-
4l		120-122	51	66.50 (66.53)	4.68 (4.72)	-



Table 2. Antibacterial activity of the compounds. MIC in  $\mu\text{gml}^{-1}$  (Zone of inhibition in mm)

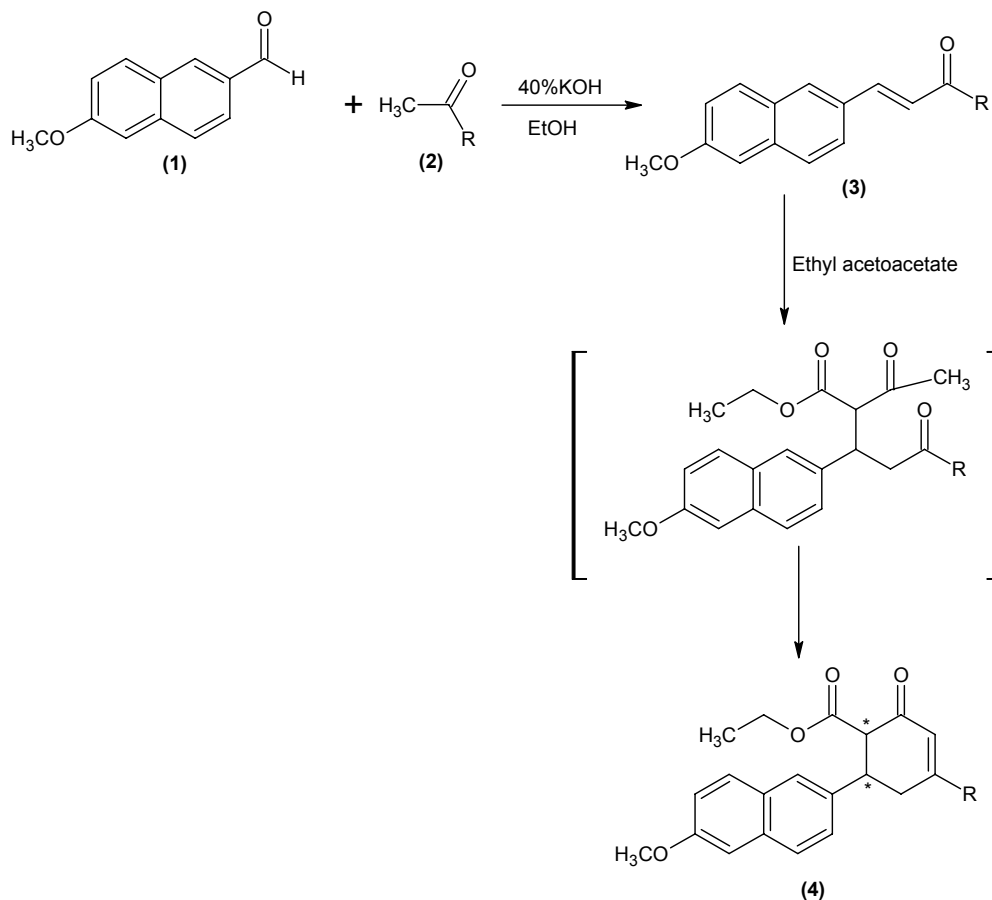
Compounds	<i>E.Coli</i> (ATTC-25922)	<i>Staphylococcus</i> (ATTC-25923)	<i>Klebsiella</i> (Recultured)	<i>Pseudomonas</i> (ATTC-27853)
4a	-	-	-	-
4b	-	-	-	-
4c	-	-	-	-
4d	12.5(15)	12.5(16)	12.5(16)	12.5(16)
4e	12.5(10)	12.5(12)	12.5(12)	12.5(12)
4f	6.25(11)	6.25(11)	6.25(11)	6.25(11)
4g	-	-	-	-
4h	6.25(10)	6.25(11)	6.25(12)	6.25(11)
4i	12.5(16)	12.5(16)	12.5(16)	12.5(16)
4j	-	-	-	-
4k	6.25(18)	6.25(18)	6.25(18)	6.25(18)
4l	6.25(18)	6.25(20)	6.25(18)	6.25(20)
Ampicillin	6.25(30)	6.25(30)	6.25(27)	6.25(28)

Note: The MIC values were evaluated at concentration range, 1.5-12.5  $\mu\text{g/ml}$ . The figures in the table show the MIC values in  $\mu\text{g/ml}$  and the corresponding zone of inhibition in mm.

Table 3. Antifungal activity of the compounds. MIC in  $\mu\text{gml}^{-1}$  (Zone of inhibition in mm)

Compounds	<i>Penicillium</i> <i>Marneffi</i> (Recultured)	<i>Trichophyton</i> <i>Mentagrophytes</i> (Recultured)	<i>AS Flavus</i> (NCIM No.524)	<i>AS fumigatus</i> (NCIM No.902)
4a	-	-	-	-
4b	-	-	-	-
4c	-	-	-	-
4d	12.5(15)	12.5(16)	12.5(14)	12.5(16)
4e	12.5(10)	12.5(12)	12.5(12)	12.5(12)
4f	12.5(11)	12.5 (12)	12.5 (12)	12.5 (11)
4g	-	-	-	-
4h	6.25(10)	6.25(11)	6.25(12)	6.25(11)
4i	12.5(14)	12.5(16)	12.5(14)	12.5(14)
4j	-	-	-	-
4k	6.25(20)	6.25(18)	6.25(18)	6.25(18)
4l	6.25(20)	6.25(18)	6.25(18)	6.25(18)
Itraconazole	6.25(28)	6.25(30)	6.25(28)	6.25(28)

Note: The MIC values were evaluated at concentration range, 1.5-12.5  $\mu\text{g/ml}$ . The figures in the table show the MIC values in  $\mu\text{g/ml}$  and the corresponding zone of inhibition in mm.



Scheme 1. Synthesis of ethyl-6-(6-methoxy-2-naphthyl)-2-oxo-4-(aryl)cyclohex-3-ene-1-carboxylate (4a-4l)

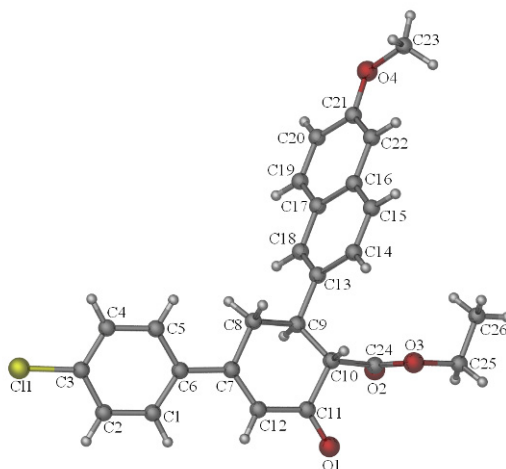


Figure 1. Molecular structure of ethyl 4-(4-chlorophenyl)-6-(6-methoxy-2-naphthyl)-2-oxocyclohex-3-ene-1-carboxylate (4f)