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Synthesis, characterization and antimicrobial activity of some novel 3-(2-bromoacetyl)phenyl benzoate dithiocarbamate derivatives

Kotresh O[1*], Sachin P. Ambekar[1], Mahesh Kumar K[1], Arunkumar M. Shirahatti[1]

1. Department of Chemistry, Karnatak Science College, Dharwad-580001, Karanataka-INDIA.

ABSTRACT

3-(2-bromacetyl) phenyl benzoate is key starting material used in synthesis of phenylephrine, (R)-3-[-1-hydroxy-2-(methylamino)ethyl]phenol which is selective α1-adrenergic receptor agonist used primarily as decongestant, as an agent to dilate the pupil, and to increase blood pressure. Hence the current research work was aim to synthesize a series of 3-(2-bromacetyl) phenyl benzoate derivatives by treatment of secondary amines with carbon disulphide and 3-(2-bromacetyl) phenyl benzoate in presence of strong base in ethanol afforded the corresponding Dithiocarbamates. Their chemical structures are characterized by ^{1}H &^{13}C NMR, MS, Elemental analysis, and chromatography methods (TLC). The antimicrobial activity was evaluated by their MIC and zone of inhibition by taking Penicillin, Streptomycin and Amphotericin as standard reference drugs. The microbial assay revealed that compounds **D4** and **D5** showed the most potent antimicrobial activity against variety of bacteria as well as fungal isolates, which may be a promising antimicrobial leading compound for the further research.

Keywords: 3-(2-bromacetyl) phenyl benzoate, Dithiocarbamates, Carbon disulphide, Antimicrobial activity.

*Corresponding Author Email:okotreshkscd@yahoo.com

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INTRODUCTION

Although several classes of antibacterial agents are presently available, resistance in most of pathogenic bacteria to these drugs constantly emerges, In order to present this critical medical problem, the discovery of new types of antibacterial agent is very important task[1], Since 3-(2-bromacetyl) phenyl benzoate is used in synthesis of phenylephrine a pharmaceutical drug which has effectiveness as decongestant stem from its vasco constriction of nasal blood vessels, thereby decreasing mucosal edema[2], Therefore, phenylephrine is less likely to cause side effects such as central nervous system stimulation, insomnia, anxiety, irritability and restlessness[3]. Due to their unique biological properties of 3-(2-bromacetyl) phenyl benzoates have played an important role in chemotherapeutic approaches to a variety of disorders. As a result the development of novel dithiocarbamte derivatives has stimulated great interest and has been the focus of many research groups over the years[4].

Dithiocarbamate acid esters are a common class of organic molecules. They exhibit valuable biological effects, including anti-bacterial, anti-fungal, ant-oxidantactivity[5], inhibition of cardiachypertrophy[6]. Dithiocarbamates are known to be active as anti-cancer[7-11]. Recently, it was found by Hirschelman's group that 5- oxohexyldithiocarbamic acid methyl ester are potent phase II enzyme inducers which could be used as cancer chemo preventive agent[12-14].

Some DTC's were also found to be pharmacologically active and are being used for the treatment of alcoholism[15] and have been tested in clinical trials for various indications including HIV[16-19], cancer[20-22].Furthermore,diarylthioureas have emerged as one the promising non-vanilloidTRPVI antagonists, possessing excellent therapeutic potentials in pain regulation[23], and human CB1 and CB2 cannabinoid receptor affinity[24].Dithiocarbamate (DTC) derivatives are well known as organic intermediates, rubber additive, additive of polluted water, vulcanizing agents and fungicides[25].Numerous studies regarding dithiocarbamates, have demonstrated that these compounds have potential anticholinergic[26-28], antimicrobial[30-32], antiviral activities[8-9].In view of these above literature, the current research is to synthesize series of novel Dithiocarbamates using 3-(2-bromacetyl) phenyl benzoate as key material for synthesis of dithiocarbamates derivatives was carried out. All the synthesized agents were evaluated for biological activity.

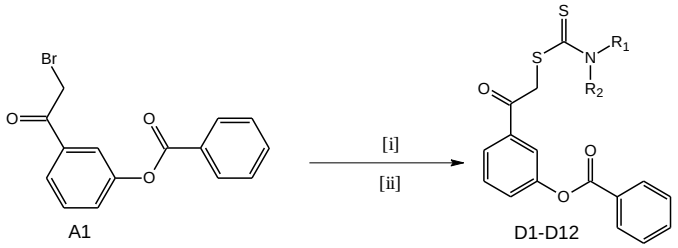
MATERIALS AND METHODS

General Procedure.

Melting points were determined with an electrical melting point apparatus Buchi, Switzerland, ^{1}H NMR spectra were recorded with tetramethylsilane as internal standard on a Bruker DPX 300 in

$CDCl_3$-d_6, Chemical shifts are given in δ ppm, coupling constant (*J)* in Hz. The ^{13}C NMR spectra were recorded at 75MHz in $CDCl_3$. Elemental analysis analyses were determined on Carlo-Rrba 1016 instrument. Mass spectra recorded on Thermofischer Scientific Model: LCQ Deca XP MAX, TLC analysis were performed on commercial Kieselgel F silica gel plates. IR Spectra were recorded on Thermo scientific model: FT/IR 460 plus.

The synthetic procedure adopted to obtain the target compounds is shown in scheme-1. The starting material, 3-(2-bromacetyl) phenyl benzoate **A1** was prepared by benzoylation of 3-hydroxy acetophenone and silicate in presence of strong base in toluene, gave intermediate, which on brominated with bromine to give 3-(2-bromacetyl) phenyl benzoate as per the described procedure[33]. The starting material (**A1**) was confirmed by crystal structure analysed on XRD data[34], main synthetic route to dithiocarbamates is based on the reaction between the desired secondary amines with CS_2 in aqueous sodium hydroxide solution and ethanol, further this dithiocarbamate salts was converted into various dithiocarbamates esters. The progress of the reaction was monitored by TLC, after the completion of the reaction, the solvent were evaporated with reduced pressure and recrystallized from ethanol at low temperature.



Scheme 1: Method-I, **Method-II**

General procedure for synthesis of 3-(2-bromacetyl) phenyl benzoate (A1):

The starting material was synthesized according to already reported procedure[4] involving 3-hydroxyacetophenone (0.01mole) and silicate (0.02mole) in toluene and was continuously stirred at room temperature, then added 1.008 g of caustic lye and continued the stirring until the reaction is complete. The completion of reaction was monitored by on TLC. On completion of the reaction, 50mL of water was added and allowed it stir for 30min, the organic layer was separated , the organic layer was cooled, to this Bromine slowly added by maintaining the 0-5°C temperature for 6hr until solid was formed The resulting precipitate was filtered washed with water and recrystallized to get pure product, Colorless white crystals.

General procedure for synthesis of 3-{[(morpholin-4-ylcarbonothioyl)sulfanyl]acetyl} phenyl benzoate (D1-12):

Method 1:

To a solution of NaOH (1 mmol) in 3mL water was added a mixture of secondary amines (1 mmol) in ethanol (25 mL). After stirring at room temperature for about 20 min, carbon disulfide (1.2 mmol) was added drop wise and resulted mixture was further stirred at room temperature for 90 min. Then 3-(2-bromacetyl) phenyl benzoate (1 mmol) were added and stirring was continued. After completion of the reaction (monitored by TLC), the solvent was removed under vacuo and the residue was extracted with dichloromethane (2 x 25 mL) and dried over anhyd. $MgSO_4$. The solvent was evaporated and the compound recrystallized from ethanol-chloroform mixture (3:1) to get the title compound **(D1-12)**.

Method 2:

Sodium salt /Potassium salt of dithiocarbamates (1 mmol) and 3-(2-bromacetyl) phenyl benzoate (1 mmol) were stirred in ethanol (25mL) at room temperature. After completion of the reaction (monitored by TLC), the solvent was removed by vacuo and the residue was extracted twice with dichloromethane and dried over anhyd. $MgSO_4$. The solvent was evaporated and the compound recrystallized from ethanol-chloroform mixture (3:1) to get the title compound **(D1-12)**.

Antimicrobial activities

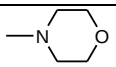
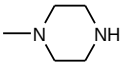
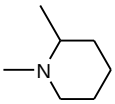
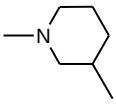
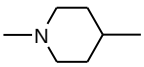
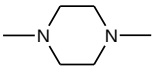
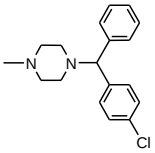
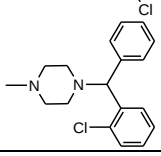
The final compounds **D1-D12** have been tested in vitro for their antimicrobial activity against gram positive bacteria:*Escherichia coli*, *Bacillus megaterium*, *Staphylococcus aureus*, *Enterococcus faecalis*, using Penicillin and Streptomycin drug as a standard drug. MICs of compounds **D1** to **D12** against these bacterial strains are tested by the standard method (Agar Diffusion Method, cultivation at 37°C for 18 hr)[36-37] was used. The tested compounds were dissolved in DMSO and then diluted with water. The agar plates of the media were prepared and wells were made in plate. Each plate inoculated with 18h old culture (100µL, 10-4 cfu). They were also evaluated for their *in vitro* antifungal activity against two mycoticstarins (*A. flavous and C. Albicans*), using Amphotericin drug as a standard drug, The agar plates of the media Czapek-Dox Agar were prepared and wells were made in plate, Each plate inoculated with 48hrs old culture (100µL,10^{-4}cfu) [36-37]. And the plates were incubated at 27°C for 48hrs [36-37],

RESULTS AND DISCUSSION

A series of 3-(2-bromacetyl) phenyl benzoate dithiocarbamates (**D1-D12**) as depicted in (**Scheme 1**) were synthesized by two methods[35] and were evaluated for chemical structures and characterized by ^{1}H &^{13}C NMR, MS, Elemental analysis, and chromatography methods (TLC). The yields and melting points of the compounds (**D1-12**) are given in **Table 1**. The structures of

the dithiocarbamate products **D1-D12** were confirmed by NMR techniques, IR spectroscopy, mass spectral and micro-analytical data.

Table 1. The yields and melting points of the compounds (D1-12)

Compd.	R1	R2	Yield(%)		m.p (°C)	
			Method I	Method II	Method I	Method II
D1		H	52	82	141-143	140-14
D2		H	50	92	138-140	140-142
D3		H	54	91	118-120	117-119
D4		H	51	82	108-110	111-113
D5		H	59	90	136-138	135- 137
D6		H	50	92	145-147	142-144
D7		H	50	79	122-125	125-127
D8		H	60	93	119-121	122-124
D9		H	58	90	120-122	118-120
D10		H	55	91	142-144	138-140
D11		H	51	91	111-113	108-110
D12		H	55	95	128-130	125-127

[a]Yields of pure, isolated products**.**

The spectral details of all these are given in the Experimental section. For example, there is one triplet signals at δ = 3.78 ppm and one broad peak signal at δ = 4.23 ppm can be attributed to morpholine ring, one singlet peak at 4.91ppm corresponds to CH2 protons in the ^{1}H NMR spectrum of compound **D1,** and there are multiplet at δ = 7.26-7.68 ppm, triplet signal at δ = 7.90 ppm, triplet of doublet signal at δ = 7.99 ppm and doublet at δ = 8.02 ppm corresponding to the aromatic protons in the expanded region (**Figure-1**), Also there are 15 different carbon in ^{13}C NMR spectrum of which morpholine ring having 4 carbon atoms, the spectrum contributes only one signals, for spectrum of ^{13}C NMR spectrum of compound **D1.** Moreover the FT-IR spectrum

of **D1** in KBr showed an absorption band at 1228 (C=S), 1061 (C-N) cm^{-1} corresponding to the CS & CN functional group. All this evidence plus molecular ion peak at *m/z* 402.33 (M+) and micro-analytical data strongly support the dithiocarbamate structure of compound **D1**.

3-{[(Morpholin-4-ylcarbonothioyl)sulfanyl]acetyl}phenyl benzoate (D1):

Light brown solid, IR (KBr, ν_{max} cm^{-1}): 2857, 3073 (C-H aliphatic and aromatic). 1732 (C=O), 1421, 1680 (C=C), 1264 (C-O), 1228 (C=S), 1061 (C-N); ^{1}H NMR (300MHz, $CDCl_3$): δH 3.77-3.80 (t, 4H), 4.23-4.43 (broad peak, 4H), 4.91 (s, 2H), 7.26-768 (m, 5H), 7.90-7.91 (t, 1H), 7.99-8.02 (d, 1H), 8.20-8.22 (d, 2H). ^{13}CNMR (75 MHz, $CDCl_3$): δC 44.52 (C9), 44.64 (C3-C5), 66.21 (C2, C6), 121.84 (C18), 126.03 (C14), 127.07 (C16), 128.67 (C17), 129.84 (C24, C26), 130.25 (C23, C27), 133.85(C22), 137.62 (C25), 151.28 (C13), 164.93 (C15), 192.16 (C10), 195.76 (C7). MS, m/z: 402.10 $[M+H]^+$; Analysis calcd. (%) for $C_{20}H_{19}NO_4S_2$: C, 59.83: H, 4.77: N, 3.49: S, 15.97. Found: C, 59.72: H, 4.85: N, 3.61: S, 15.94.

3-{[(Piperazin-1-ylcarbonothioyl)sulfanyl]acetyl}phenyl benzoate (D2):

Pale yellow powder, IR (KBr, ν_{max} cm^{-1}): 2857, 3070 (C-H aliphatic and aromatic), 1738 (C=O), 1431, 1693 (C=C), 1264 (C-O), 1219 (C=S), 1062 (C-N), 3458 (N-H). ^{1}H NMR (300MHz, $CDCl_3$): δH 4.25 (s, 8H), 5.01(s, 2H), 7.47-7.77 (m, 5H), 7.90-8.04 (m, 2H), 8.16-8.18 (d, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δC 24.2 (C2, C6, C3, C5), 44.8 (C10), 121.8 (C18), 126.08 (C16), 126.92 (C24, C26), 128.64 (C14), 129.19 (C15), 130.24 (C22, C23, C27), 133.80 (C25), 137.3 (C13), 151.24 (C17), 164.92 (C20), 192.60 (C7), 193.79 (C11). MS, m/z: 402.10 $[M+H]^+$; Analysis calcd. (%) for $C_{20}H_{20}N_2O_3S_2$: C, 59.98: H, 5.03: N, 6.99: S, 16.01. Found: C, 59.83: H, 5.14: N, 6.84: S, 16.28.

3-{[(Piperidin-1-ylcarbonothioyl)sulfanyl]acetyl}phenyl benzoate (D3):

Brown colour powder, IR (KBr, ν_{max} cm^{-1}): 2851, 3073 (C-H aliphatic and aromatic), 1731 (C=O), 1429, 1688 (C=C), 1259 (C-O), 1237 (C=S), 1061 (C-N). ^{1}H NMR (300MHz, $CDCl_3$): δH 1.99 (m, 6H), 3.74-3.96 (m, 4H), 4.91 (s, 2H), 7.26-7.66 (m, 5H), 7.91-8.00 (t, 1H), 8.02-8.03 (m, 1H), 8.20-8.23 (d, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δC 24.9 (C3, C5), 25.6 (C4), 44.2 (C10), 48.7 (C2, C6), 120.3 (C18), 125.6 (C14), 126.5 (C16), 128.7 (C24, C26), 129.1 (C15), 130.3 (C22, C23), 131.0 (C25), 137.2 (C13), 151.30 (C17), 165.2 (C20), 194.2 (C11), 201.9 (C7). MS, m/z: 401.09 $[M+H]^+$; Analysis calcd. (%) for $C_{21}H_{21}NO_3S_2$: C, 63.13: H, 5.03: N, 3.51: S, 16.05. Found: C, 63.24: H, 5.15: N, 3.40: S, 16.09.

3-({[(2-Methylpiperidin-1-yl)carbothioyl]sulfanyl}acetyl)phenyl benzoate (D4):

Light Brown colour powder, IR (KBr, ν_{max} cm^{-1}): 2851, 3073 (C-H aliphatic and aromatic), 1734 (C=O), 1421, 1688 (C=C), 1259 (C-O), 1215 (C=S), 1056 (C-N). ^{1}H NMR (300MHz, $CDCl_3$): δH 2.35 (s, 3H), 2.50 (m, 6H), 4.05-4.45 (m, 4H), 7.48-7.77 (m, 5H), 7.96-8.04 (m, 2H), 8.17-8.19 (d, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δC 18.1 (C9), 23.2 (C4), 25.2 (C3), 33.1 (C5), 44.2 (C11), 46.5 (C2), 54.7 (C6), 120.3 (C19), 125.6 (C15), 126.0 (C17), 129.3 (C16), 128.7 (C25, C27),130.6 (C28, C23, C24), 136.2 (C14), 151.3 (C18), 165.2 (C21), 194.0 (C12), 201.9 (C7). MS, m/z: 415.11 $[M+H]^+$; Analysis calcd. (%) for $C_{22}H_{23}NO_3S_2$: C, 63.89: H, 5.61: N, 3.39: S, 15.31. Found: C, 63.96: H, 5.53: N, 3.48: S, 15.31.

3-({[(3-Methylpiperidin-1-yl)carbothioyl]sulfanyl}acetyl)phenyl benzoate (D5):

Light yellow colour powder, IR (KBr, ν_{max} cm^{-1}): 2851, 3073 (C-H aliphatic and aromatic), 1731 (C=O), 1431, 1688 (C=C), 1237 (C=S), 1061 (C-N). ^{1}H NMR (300MHz, $CDCl_3$): δH 1.06-1.07 (d, 3H), 1.34-1.59 (m, 2H), 1.45-1.55 (m, 2H), 2.61-2.87 (m, 2H), 2.70-2.83 (m, 2H), 4.13 (s, 2H), 7.28-7.66 (m, 5H), 7.90-8.01 (m, 2H), 8.20-8.22 (d, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δC 18.1 (C12), 22.4 (C5), 28.8 (C3), 33.8 (C4), 44.9 (C9), 49.1 (C6), 54.7 (C2), 120.3 (C15), 125.6 (C19), 126.5 (C17), 128.7 (C25, C27), 129.3 (C18), 130.3 (C23, C24, C28), 134.4 (C26), 137.9 (C14), 151.40 (C16), 165.3 (C21), 194.3 (C10), 201.8 (C7). MS, m/z: 414.2 $[M+H]^+$; Analysis calcd. (%) for $C_{22}H_{23}NO_3S_2$: C, 63.89: H, 5.61: N, 3.39: S, 15.51. Found: C, 63.72: H, 5.82: N, 3.54: S, 15.43.

3-({[(4-Methylpiperidin-1-yl)carbothioyl]sulfanyl}acetyl)phenyl benzoate(D6):

Light yellow colour powder, IR (KBr, ν_{max} cm^{-1}): 2852, 3074 (C-H aliphatic and aromatic), 1734 (C=O), 1428, 1692 (C=C), 1264 (C=S), 1054 (C-N). ^{1}H NMR (300MHz, $CDCl_3$): δH 1.21-1.43(m, 3H), 1.34-1.59 (m, 4H), 2.5-2.8 (m, 4H), 4.91 (m, 2H), 7.27-7.66 (m, 5H), 7.91-8.00 (m, 2H), 8.16-8.19 (d, 2H); ^{13}C NMR (75 MHz, $CDCl_3$): δC 20.4 (C12), 31.7 (C3, C4, C5), 46.2 (C2, C6), 120.3 (C15), 125.6 (C19), 126.1 (C17), 129.2 (C18), 128.7 (C27, C25), 130.3 (C23, C24, C28), 134.0 (C26), 137.2 (C14), 151.3 (C16), 165.2 (C21), 194.2 (C10), 201.9 (C8). MS, m/z: 414.12 $[M+H]^+$; Analysis calcd. (%) for $C_{22}H_{23}NO_3S_2$: C, 63.89: H, 5.61: N, 3.39: S, 15.51. Found: C, 63.58: H, 5.83: N, 3.52: S, 15.63.

3-({[(4-Methylpiperazin-1-yl)carbonothioyl]sulfanyl}acetyl)phenyl benzoate(D7):

Yellow colour powder, IR (KBr, ν_{max} cm^{-1}): 2852, 3074 (C-H aliphatic and aromatic), 1734 (C=O), 1412, 1686 (C=C), 1264 (C=S), 1058 (C-N). ^{1}H NMR (300MHz, $CDCl_3$): δH 2.35 (s, 3H), 2.51-2.54 (m, 4H), 4.05-4.32 (m, 4H), 4.90 (s, 2H), 7.46-7.69 (m, 5H), 7.90-7.91 (m, 1H), 7.99-8.02 (m, 1H), 8.20-8.23 (m, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δC 43.1 (C7), 44.2 (C11), 51.1 (C2, C6), 54.0 (C3, C5), 120.3 (C19), 125.6 (C15), 126.0 (C17), 129.1 (C18), 128.7 (C25, C27), 130.3 (C24,

C23, C28), 134.0 (C26), 137.2 (C14), 151.3 (C16), 165.2 (C21), 194.2 (C12), 196.5 (C8). MS, m/z: 415.10 $[M+H]^+$; Analysis calcd. (%) for $C_{21}H_{22}N_2O_3S_2$: C, 60.84: H, 5.35: N, 6.76: S, 15.47. Found: C, 60.76: H, 5.59: N, 6.46: S, 15.63.

3-{[(Dimethylcarbamothioyl)sulfanyl]acetyl}phenyl benzoate(D8):

Brown colour powder, IR (KBr, ν_{max} cm^{-1}): 2853, 3084 (C-H aliphatic and aromatic), 1734 (C=O), 1503, 1692 (C=C), 1229 (C=S), 1058 (C-N). ^1H NMR (300MHz, $CDCl_3$): δH 1.25 (t, 6H), 2.58 (q, 4H), 4.13 (s, 2H), 7.34-7.65 (m, 5H), 7.81-8.01 (m, 2H), 8.12-8.28 (d, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δC 12.3 (C24, C26), 44.2 (C18), 44.9 (C23, C25), 120.3 (C6), 125.6 (C4), 126.2 (C2), 129.1 (C3), 128.7 (C14, C16), 130.3 (C12, C13, C17), 134.0 (C15), 137.2 (C5), 151.3 (C1), 165.5 (C8), 194.2 (C10), 197.5 (C20). MS, m/z: 388.15 $[M+H]^+$; Analysis calcd. (%) for $C_{20}H_{21}NO_3S_2$: C, 61.99: H, 5.46: N, 3.61: S, 16.55. Found: C, 61.83: H, 5.69: N, 3.52: S, 16.68.

3-{[(Diethylcarbamothioyl)sulfanyl]acetyl}phenyl benzoate (D9):

Brown colour powder, IR (KBr, ν_{max} cm^{-1}): 2856, 3067 (C-H aliphatic and aromatic), 1736 (C=O), 1503, 1686 (C=C), 1228 (C=S), 1058 (C-N). ^1H NMR (300MHz, $CDCl_3$): δH 2.47 (s, 6H), 4.13 (s, 2H), 7.34-7.65 (m, 5H), 7.81-8.01 (m, 2H), 8.12-8.28 (d, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δC 41.1 (C23, C24), 44.2 (C18), 120.3 (C6), 125.6 (C4), 126.3 (C2), 128.7 (C14, C16), 129.1 (C3), 130.3 (C12, C13, C17), 134.0 (C15), 137.2 (C5), 151.3 (C1), 165.5 (C8), 194.2 (C10), 197.5 (C20). MS, m/z: 361.08$[M+H]^+$; Analysis calcd. (%) for $C_{18}H_{17}NO_3S_2$: C, 60.14: H, 4.77: N, 3.90: S, 17.84. Found: C, 60.28: H, 4.86: N, 3.72: S, 17.90.

3-{[(Pyrrolidin-1-ylcarbonothioyl)sulfanyl]acetyl}phenyl benzoate (D10):

Light brown colour, IR (KBr, ν_{max} cm^{-1}): 2852, 3074 (C-H aliphatic and aromatic), 1733 (C=O), 1403, 1686 (C=C), 1249 (C=S), 1057 (C-N). ^1H NMR (300MHz, $CDCl_3$): δH 1.89-2.09 (m, 4H), 3.67-3.77 (q, 4H), 4.91 (s, 2H), 7.26-766 (m, 5H), 7.91-8.01 (m, 1H), 8.16-8.19 (d, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δC 24.7 (C3, C4), 44.2 (C9), 52.7 (C2, C5), 120.3 (C17), 125.6 (C13), 126.3 (C15), 128.7 (C23, C25), 129.1 (C14), 130.3 (C21, C22, C26), 134.0 (C24), 137.3 (C12), 151.3 (C16), 165.2 (C19), 194.3 (C10), 201.0 (C6). MS, m/z: 386.04 $[M+H]^+$; Analysis calcd. (%) for $C_{20}H_{19}NO_3S_2$: C, 62.31: H, 4.97: N, 3.63: S, 16.64. Found: C, 62.47: H, 4.81: N, 3.49: S, 16.73.

4-({[(4-Chlorophenyl)(phenyl)methyl piperazine-1yl)carbonthiony] sulphanyl}acetyl) phenyl benzoate (D11):

Pale yellow colour powder, IR (KBr, ν_{max} cm^{-1}): 2859, 3091 (C-H aliphatic and aromatic), 1706 (C=O), 1421, 1628 (C=C), 1233 (C=S), 1047(C-N). ^1H NMR (300MHz, $CDCl_3$): δH 2.48-2.55 (m, 4H), 2.65-2.78 (m, 4H), 4.13 (s, 2H), 5.19 (s, 1H), 7.01-7.18 (m, 9H), 7.31-7.54 (m, 5H), 7.75 -

7.79 (d, 2H), 8.14-8.19 (d, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δC 44.2 (C17), 48.8 (C3, C5), 51.71 (C2, C6), 73.6 (C9), 120.3 (C32), 125.6 (C28), 126.0 (C30), 126.3 (C23), 128.3 (C25, C21), 128.7 (C38, C40), 129.1 (C29), 129.3 (C22, C24), 129.4 (C14, C12), 129.7 (C11, C15), 130.3 (C36, C37, C41), 131.8 (C13), 137.2 (C27), 140.9 (C10), 142.7 (C20), 151.5 (C31), 165.4 (C34), 194.2 (C18), 196.4 (C7). MS, m/z: 601.14 $[M+H]^+$; Analysis calcd. (%) for $C_{33}H_{29}ClN_2O_3S_2$: C, 65.93: H, 4.86: N, 4.66: S, 10.67. Found: C, 65.80: H, 4.97: N, 4.84: S, 10.79.

4-({[(2-Chlorophenyl)(phenyl)methylpiperazine-1yl)carbonthiony]sulphanyl}acetyl) phenyl benzoate (D12):

Brown colour powder, IR (KBr, ν_{max} cm^{-1}): 2810, 3093 (C-H aliphatic and aromatic), 1708 (C=O), 1423, 1619 (C=C), 1241 (C=S), 1039 (C-N). ^{1}H NMR (300MHz, $CDCl_3$): δH 2.49-2.56 (m, 4H), 2.66-2.76 (m, 4H), 4.18 (s, 2H), 5.20 (s, 1H), 7.01-7.20 (m, 9H), 7.32-7.56 (m, 5H), 7.76-7.80 (d, 2H), 8.16-8.18 (d, 2H). ^{13}C NMR (75 MHz, $CDCl_3$): δC 44.2 (C17), 48.8 (C3, C5), 51.7 (C2, C6), 64.5 (C9), 120.5 (C32), 125.8 (C28), 126.1 (C30), 126.4 (C23), 127.5 (C14), 127.7 (C13), 128.3 (C21, C25), 128.7 (C38, C40), 129.4 (C22, C24), 129.4 (C12, C15), 129.3 (C29), 130.8 (C36, C37, C41), 133.9 (C11), 134.2 (C39), 137.2 (C27), 142.9 (C20), 143.7 (C10), 151.5 (C31), 165.4 (C34), 194.2 (C18), 196.5 (C7). MS, m/z: 602.14 $[M+H]^+$; Analysis calcd. (%) for $C_{33}H_{29}ClN_2O_3S_2$: C, 65.93: H, 4.86: N, 4.66: S, 10.67. Found: C, 65.81: H, 4.92: N, 4.74: S, 10.82.

Antimicrobial activities

The results of biological screening results of minimum inhibitory concentrations (MIC in µg/mL) are presented in **Table 2 & Table 3**.

Table 2. Antibacterial screening result of compounds (D1-D12) (MIC in µg/mL)

Compound	*E-coli*	*B.megaterium*	*S. aureus*	*E. Facecalis*
D1	5	-	-	10
D2	8	-	-	22
D3	-	9	-	13
D4	16	18	11	-
D5	25	20	18	12
D6	-	-	11	-
D7	18	22	9	-
D8	10	6	7	2
D9	18	2	8	-
D10	3	-	-	-
D11	19	4	6	5
D12	13	18	8	-
Penicillin	10	12	15	16
Streptomycin	20	14	19	22

Table 3. Antifungal screening result of compounds (D1-D12)(MIC in µg/mL)

Compound	*A. flavus*	*C. Albicans*
D1	-	8
D2	4	-
D3	-	12
D4	10	6
D5	14	-
D6	-	-
D7	12	3
D8	16	-
D9	15	3
D10	5	-
D11	-	6
D12	12	8
Amphotericin	10	15

CONCLUSSION

The screening results revealed that the compounds with methyl substituents have shown significant antimicrobial activity against *E. coli*. In this study compound **D-5** is more active against *E. coli, B. megaterium, S. aureus*strains. The Compound **D-7** is showed good activity against *E. coli, B. megaterium*,**D-2** is showed activity against *E. Facecalis*,**D-9** is showed activity against *E. coli* and**D-12** is showed activity against *B. megaterium.* The compound **D3** is more active against *C. Albicans*and compound **D8** showed potent antifungal activity against *A. flavus*comparing to standard drug.

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REFERENCE

1. Leeb M. Nature. Antibiotics: A shot in the arm. 2004; 431: 892-893.
2. Cooper BE, Review and Update on Inotropes and Vasopressors. AACN Advanced Critical Care, 2008; 19: 5-15.
3. Junichi I, Takuro I, Chikara K and Mitsutaka K. Ann Rept. G. Tanabe Co. Ltd. 1956; 1: 17-20.
4. Naoto O, Mariko O, Takashi S, Satoshi K, Atsuko M, Noriaki U, Yoshisuke N, Keishi K, Masamori S and Yushi K. PCT Int. Appl. WO 2008029825/A1 20080313, 2008.
5. Schreck R, Meier B, Mannel DN, Droge W and Baeuerle PA. Dithiocarbamates as potent inhibitors of nuclear factor kappa B activation in intact cells.J Exp Med 1992; 175: 1181-1194.

6. Ha T, Li Y, Gao X, McMullen JR, Shioi T, Izumo S, Kelley JL, Zhao A, Haddad GE, Williams DL, Browder IW, Hao RL and Li C.Attenuated cardiovascular hypertrophy and oxidant generation in response to angiotensin II infusion in glutaredoxin-1 knockout mice. Free RadicBiol Med 2005; 39: 1570-1580.
7. Scho¨nenberger, Helmut and Lippert P.Cytostatics. 16. Antimicrobial and tumor-inhibiting properties of dithiourethane and studies on the mechanism of action.Pharmazie 1972; 27: 139-145.
8. Scozzafava A, Mastrolorenzo A and Supuran CT. Arylsulfonyl-N,N-diethyl-dithiocarbamates: a novel class of antitumor agents.Bioorg Med ChemLett 2000; 10: 1887-1891.
9. Cao S-L, Feng Y-P, Jiang Y-Y, Liu S-Y, Ding G-Y and Li R-T. Synthesis and in vitro antitumor activity of 4(3H)-quinazolinone derivatives with dithiocarbamate side chains.Bioorg Med ChemLett 2005; 15: 1915-1917.
10. David W, Robert RW, Grayson WS, Linda BW and Thomas K. Method of treating cancer usingdithiocarbamate derivatives.US2005096304 (A1) 2005.
11. Yusuf OI, Emin O, Abdulmuttalip S, Mustafa C, Alper O and Adnan S. Chemoprotective Effect of a Nuclear Factor-kB Inhibitor, PyrrolidineDithiocarbamate, Against Cisplatin-Induced Testicular Damage in Rats. Journal of Andrology, 2009; 30(5): 505-514.
12. Hirschelman WH, Song LS, Park EJ, Tan Y, Yu R, Hawthorne M, Mehta RG, Grubbs CJ, Lubet RA, Moriarty RM and Pezzuto JM. 224th ACS National Meeting: Division of Medicinal Chemistry 2002; 98.
13. Hirschelman WH, Kosmeder II JW, Song LS, Park EJ, Moriarty RM and Pezzuto JM. 224th ACS National Meeting: Division of Medicinal Chemistry 2002; 178.
14. Hirschelman WH. 225th ACS National Meeting: Division of Medicinal Chemistry2003; 86.
15. Jacobsen E. Biochemical Methods in the Treatment of Alcoholism, with Special Reference to Antabuse.Proc Roy Soc Med 1950; 43: 519-526.
16. Programmed cell death of T lymphocytes in AIDS related HIV and SIV infections. AIDS Res Hum Retrov 1993; 9: 83-89.
17. Eersh EM, Brewton G, Abrams D, Bartlett J, Galpin J, Gill P, Gorter R, Gottlieb M, Jonikas JJ and Landesman S. Ditiocarb Sodium (Diethyldithiocarbamate) Therapy in Patients With Symptomatic HIV Infection and AIDSA Randomized, Double-blind, Placebo-Controlled, Multicenter Study. JAMA. 1991, 265, 1538-1544.

18. Kaplan CS, Petersen EA, Yocum D and Hersh EM.A randomized, controlled dose response study of intravenous sodium diethyldithiocarbamate in patients with advanced human immunodeficiency virus infection. Life Sci 1989; 45: iii-ix.

19. Lang JM, Touraine JL, Trepo C, Choutet P, Kirstetter M, Falkenrodt A, Herviou AL, Livrozet JM, Retornaz G and Touraine F.Randomised, double-blind, placebo-controlled trial of ditiocarb sodium ('Imuthiol') in human immunodeficiency virus infection. Lancet 1988; 2: 702-706.
20. Dufour P, Lang JM, Giron C, Duclos B, Haehnel P, Jaeck D, Jung J and Oberling MF. Sodium dithiocarb as adjuvant immunotherapy for high risk breast cancer: a randomized study. Biotherapy 1993; 6: 9-12.
21. Francis P, Markman M, Hakes T, Reichman B, Rubin S, Jones W, Lewis JL, Curtin J, Barakat R and Phillips M.Diethyldithiocarbamatechemoprotection of carboplatin-induced hematological toxicity.J Cancer Res ClinOncol 1993; 119: 360-362.
22. Verma S, Stewart DJ, Maroun JA and Nair RC.A Randomized Phase II Study of Cisplatin Alone Versus CisplatinPlusDisulfiram. Am J ClinOncol 1990; 13: 119.
23. Suh YG, Lee YS, Min KH, Park OH, Kim JK, Seung HS, Seo SY, Lee BY, Nam YH, Lee KO, Kim HD, Park HG, Lee J, Oh U, Lim JO, Kang SU, Kil MJ, Koo JY, Shin SS, Joo YH, Kim JK, Jeong YS, Kim SY and Park YH. Novel Potent Antagonists of Transient Receptor Potential Channel, Vanilloid Subfamily Member 1: Structure-Activity Relationship of 1,3-Diarylalkyl Thioureas Possessing New Vanilloid Equivalents. J Med Chem2005; 48:5823-5836.
24. Muccioli GG, Wouters J, Scriba GKE, Poppitz W, Poupaert JH and Lambert DM. 1-Benzhydryl-3-phenylurea and 1-Benzhydryl-3-phenylthiourea Derivatives: New Templates among the CB_1 Cannabinoid Receptor Inverse Agonists. J Med Chem2005;48: 7486-7490.
25. Cvek B and Dvorak ZT. Targeting of Nuclear Factor-κB and Proteasome by Dithiocarbamate Complexes with Metals. Curr Pharm Des 2007; 30: 3155-3167.
26. Ozkani F, Dalkara S, Calis U, Erol K and Ozdemir M. Synthesis and anticholinergic activity of some new N,N-disubstitutedcarbamodithioic acid 2-oxo-2- (diphenylamino)ethyl esters. II-Farmaco 1993; 48: 1153-1158.
27. Calis U, Ozkani F, Dalkara S, Erol K and Ozdemir M. Synthesis and anticholinergic properties of some N,N-disubstitutedcarbamodithioic acid 2-oxo-2-(phenylamino)ethyl esters. Pharmazie 1993; 48: 945-946.

28. Karali N, Apak I, Özkirimli S, Gürsoy A, Dogan SU, Eraslan A and Özdemir O.Synthesis and pharmacology of new dithiocarbamic acid esters derived from phenothiazine and diphenylamine. Arch Pharm Pharm Med Chem 1999; 332: 422-426.
29. Weuffen W and Kewitsch A. Orientating studies of the tuberculostatic properties of some dithiocarbamic acid esters (dithiourethanes) and dithiocarbamic salts. Arch ExpVeterinarmed 1967; 21: 1049-1059.
30. Chabric P, Maillard G and Quevauviller A. Relation between chemical structure and antibacterial and antifungal activity of esters of N-disubstituteddithiocarbamic acid. Ann Pharm Franc 1956; 14: 720-728.
31. Gupta SP and Garg DML. Potential fungicidal compounds. II. Some bis(halonitrophenyl) and (-naphtyl) esters of piperazinobis(dithiocarbamic acid). J Indian ChemSoc 1965; 42: 412-414.
32. Erol DD, Calis U and Yulug N. Synthesis and antimicrobial activities of some dithiocarbamate derivatives of kojic acid. Boll ChimFarmaceutico 1995; 134: 620-623.
33. Jaakko H and Erkki H. Synthesis of Phenolic Alkamine Ethers of Adrenaline and Some Related Compounds. ActaChemScand 1959; 13: 329-333.
34. Sachin PA, Deverajegowda HC, Shylajakumari J, MaheshKumar K, and Kotresh O. 3-(2-Bromoacetyl)phenyl benzoate. ActaCryst 2013; E69: 322.
35. Joke Vandenbergh, JeroenDergent, Bert Conings TVV, Gopala Krishna, WouterMaes, Thomas JC, Laurence Lutsen, Jean Manca and Dirk JMV. Synthesis and characterization of water-soluble poly(p-phenylenevinylene) derivatives via the dithiocarbamate precursor route. European Polymer Journal 2011; 47: 1827-1835.
36. Threlfall EJ, Fisher IST, Ward L, Tschape H and Gernersmidt P. Harmonization of antibiotic susceptibility testing for Salmonella: results of a study by 18 national reference laboratories within the European Union-funded Enter-net group. Microb Drug Resist 1999; 5: 195-200.
37. Walker RD, (2000a). In Antimicrobial Therapy in Veterinary Medicine, 3rd edn. Eds Prescott JF, Baggot JD, Walker RD, Iowa State University press, Ames, IA, 12-26.