Synthesis of novel antibacterial and antifungal dithiocarbamate-containing piperazine derivatives via re-engineering multicomponent approach

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GRAPHICAL ABSTRACT

ABSTRACT

A metal-free multicomponent synthetic route for the diverse preparation of dithiocarbamate-containing piperazine derivatives was developed through the C-N bond cleavage of DABCO ring. This multicomponent re-engineering approach proceeds via the reaction of amines, CS2, and DABCO salts in one pot. Various DABCO salts and secondary amines are tolerated well in this protocol to afford a broad spectrum of dithiocarbamate-containing piperazines in good to high yields. Then, the selected compounds have been deployed against some critical types of bacteria and fungi. A certain number of synthesized compounds revealed not only appropriate antibacterial activity as investigated by disc fusion and minimum inhibitory concentration methods against bacteria (Gram-positive and Gram-negative), but also depicted good to excellent antifungal activity.

1. Introduction

Nowadays, the world is encountering the coronavirus disease 2019 (COVID-19) pandemic. It has been amply clear that this is a viral disease. Nevertheless, it could still weaken patients' immune systems, hence leading to secondary bacterial or fungal infections taking hold [1, 2]. For instance, the COVID-19 delta variant in some patients led to apparent of mucormycosis, previously called zygomycosis and also known as black fungus [3, 4]. Considering the extensive repercussion of this pandemic, scientists must come up with novel and effective antiviral drugs [5, 6], and need to find new antibacterial and antifungal compounds [7, 8]. Indisputably, discovering highly efficient remedies for these infectious diseases is of paramount importance.
diseases has exponentially emerged as an area of focus [9, 10, 11]. In this context, the synthesis of various dithiocarbamates (DTCs), which revealed remarkable properties and diverse applications in bioorganic and medicinal chemistry such as fungicides and pesticides, crop protection agents and anticancer agents, is thoroughly crucial [12, 13, 14, 15, 16, 17, 18]. For example, take Zineb, Maneb and disulfiram as imperative dithiocarbamate-based biologically active molecules (Figure 1) [19,20]. Further, piperazine is a six-membered heterocyclic compound encompassing two nitrogen atoms in the positions 1 and 4 [21]. On account of the importance of the substituted piperazines in many different biomedical applications, a swift approach to the direct synthesis of novel piperazines has been tremendously beneficial. Among various methods reported for the synthesis of piperazines, DABCO bond cleavage found widespread applications for the direct preparation of valuable piperazines scaffolds. A broad spectrum of nucleophiles such as carboxylic acids, amines, thiols, phenolates, indoles, azide, and alcohols were successfully deployed for the cleavage of C–N bond in DABCO [22]. Piperazine-based drugs such as Indinavir, an antiretroviral drug used to treat HIV, Ciprofloxacin, Buspirone, and Prazosin are available in the market (Figure 1). In addition, Vestipitant, a NK-1 receptor antagonist is in clinical trials to treat anxiety and tinnitus [23, 24]. Until now, MDL@Drug Data Report (MDDR) database includes approximately 11800 scaffolds bearing the piperazine heterocycles. Since piperazine compounds are normally applied as a linker between two portions of a bioactive molecules, combining piperazine along with dithiocarbamate moieties in a single structure may have synergistic effect for designing of novel and promising pharmacologically active compounds.

Since the conventional synthesis encompassing the reaction of two reagents have limitations for preparing diverse and complex products, multicomponent reactions (MCRs) or one-pot three or more reactants procedures, making it viable to generate ensued molecules with more complexity and efficiency [25]. Besides, the re-engineering approach makes it possible to adopt specific nonelementary two-component reactions into higher-order MCRs. Implementing the re-engineering approach as a rational design culminated in increasing the dimensionality of MCRs and achieving diverse skeletal molecules [26]. In addition, in terms of the green chemistry criteria, MCRs enjoy excellent green properties such as pot, atom, and step economy and appropriate environmental impact factor (E-factor), as the lower amount of waste products were produced [27]. Consequently, in this project, dithiocarbamate-containing piperazine derivatives which were recently prepared with us via a two-component reaction [28], effectively were

Figure 1. Representative piperazine- and dithiocarbamate-centered drugs and biologically active compounds.

Scheme 1. Single-pot three-component approach for the synthesis of dithiocarbamate-containing piperazines (4) using secondary amines (1), CS₂ (2) and DABCO salts (3).

Scheme 2. Model reaction for optimization of the reaction conditions for the synthesis of dithiocarbamate-containing piperazine (4f) using amine (1a), CS₂ (2) and DABCO salt (3b).
synthesized through a novel re-engineering multicomponent approach and subsequently, their antifungal and antibacterial properties were investigated (Scheme 1).

2. Experimental

2.1. General

Starting materials and solvents were purchased from Merck and Fluka and were applied as received. All reactions were carried out in sealed vessel at high temperature for overnight. ¹H and ¹³C NMR spectra were recorded on a Bruker AMX 300 MHz device with tetramethylsilane (TMS) as internal standard for NMR solvents. Purity of products and progress of the reactions were checked by thin-layer chromatography using TLC plates and visualization was carried out using iodine or KMnO₄ solution. Melting points were measured by an electrothermal digital apparatus. HRMS (High Resolution Mass Spectra) was measured on a Bruker FTMS Exactive instrument equipped with an APCI source in the positive-ion mode.

2.2. General procedure for preparation of quaternary ammonium salts from DABCO

DABCO (10 mmol, 1.12 g) was dissolved in THF (20 mL) and then an alkyl halide such as isopropyl, isobutyl, allyl, benzyl, isopentyl halides (10 mmol) was added and the reaction mixture was heated at 70 °C for 24 h to afford a precipitate. Filtration of the precipitate, washing with diethyl ether (3 × 15 mL), and drying under vacuum afforded the corresponding product. The isolated compounds were well characterized by NMR spectroscopy and HRMS analysis.

2.2.1. 2-(4-isopropylpiperazin-1-yl)ethyl diethylcarbamodithioate (4a)

Yellow solid (192 mg, 70%); mp 53.5–55 °C. ¹H NMR (300 MHz, CDCl₃) δ 3.53 (s, 3H), 3.48–3.41 (t, J = 6.9 Hz, 2H), 3.36 (s, 3H), 2.72–2.61 (m, 3H), 2.56 (brs, 8H), 1.04 (d, J = 6.5 Hz, 6H) ppm; ¹³C NMR (75 MHz, CDCl₃) δ 197.1, 192.2, 191.5, 189.5, 56.8, 54.3, 53.2, 48.4, 45.2, 41.3, 34.5, 18.5 ppm; HRMS (ESI) calculated for C₁₂H₂₅N₃S₂ [M + H]⁺: 316.1881; Found: 316.1877.

2.2.2. 2-(4-isopropylpiperazin-1-yl)ethyl diethyldiaminomodiocarbamothioate (4b)

Pale yellow solid (208 mg, 89%); mp 53–56 °C. ¹H NMR (300 MHz, CDCl₃) δ 3.81 (t, J = 6.9 Hz, 2H), 3.55 (s, 3H), 2.74 (brs, 8H), 1.27 (t, J = 6.8 Hz, 6H), 1.07 (d, J = 6.5 Hz, 6H) ppm; ¹³C NMR (75 MHz, CDCl₃) δ 195.4, 192.2, 191.5, 189.5, 56.9, 54.6, 54.0, 52.7, 51.3, 48.4, 48.2, 45.4, 33.9, 18.4, 12.4, 11.5 ppm; HRMS (ESI) calculated for C₁₄H₂₇N₃S₂ [M + H]⁺: 304.1881; Found: 304.1873.

2.2.3. 2-(4-isopropylpiperazin-1-yl)ethyl pyrrolidine-1-carbodithioate (4c)

Colorless viscous oil (270 mg, 89%); ¹H NMR (300 MHz, CDCl₃) δ 2.76 (brs, 8H), 2.56 (brs, 2H), 3.89 (brs, 2H), 3.47 (t, J = 6.9 Hz, 6H) ppm; ¹³C NMR (75 MHz, CDCl₃) δ 195.5, 192.2, 191.5, 189.5, 56.7, 54.6, 54.0, 52.7, 51.3, 48.4, 43.4, 34.0, 59.2, 50.2, 48.1, 33.0, 25.6, 18.3 ppm; HRMS (ESI) calculated for C₁₄H₂₇N₃S₂ [M + H]⁺: 304.1881; Found: 304.1873.
2.3.5. 2-(4-isopropylpiperazin-1-yl)ethyl morpholine-4-carbodithioate (4e)
Pale yellow solid (168 mg, 53%); mp 70–75°C; 1H NMR (300 MHz, CDCl3) δ 4.17 (brs, 2H), 3.91 (brs, 2H), 3.75–3.66 (m, 4H), 3.51–3.37 (m, 2H), 2.68–2.62 (m, 3H), 2.55 (brs, 8H), 1.01 (d, J = 6.4 Hz, 6H) ppm; 13C NMR (75 MHz, CDCl3) δ 197.3, 66.1, 66.0, 56.5, 54.2, 52.7, 50.8, 50.5, 48.1, 33.7, 18.2 ppm; HRMS (ESI) calculated for C14H27N3OS2 [M+H]+: 318.1674; Found: 318.1673.

2.3.6. 2-(4-isobutylpiperazin-1-yl)ethyl diethylcarbamodithioate (4f)
Colorless viscous oil (301 mg, 95%); 1H NMR (300 MHz, CDCl3) δ 4.01 (q, J = 7.1 Hz, 2H), 3.74 (q, J = 7.1 Hz, 2H), 3.44 (t, J = 7.6 Hz, 2H), 2.66 (t, J = 6.3 Hz 2H), 2.55 (brs, 4H), 2.41 (brs, 4H), 2.06 (d, J = 6.5 Hz, 6H) ppm; 13C NMR (75 MHz, CDCl3) δ 195.5, 66.8, 57.0, 53.3, 52.9, 49.3, 46.5, 34.0, 25.2, 20.8, 12.3, 11.5 ppm; HRMS (ESI) calculated for C15H31N3S2 [M+H]+: 318.2038; Found: 318.2033.

2.3.7. 2-(4-isobutylpiperazin-1-yl)ethyl dimethylcarbamodithioate (4g)
Colorless viscous oil (266 mg, 92%); 1H NMR (300 MHz, CDCl3) δ 3.54 (brs, 3H), 3.44 (t, J = 6.5 Hz, 2H), 3.36 (brs, 3H), 2.67 (t, J = 7.4 Hz, 2H), 2.52 (brs, 4H), 2.42 (brs, 4H), 2.07 (d, J = 7.3 Hz, 2H), 1.76 (m, 1H), 0.88 (d, J = 6.4 Hz, 6H) ppm; 13C NMR (75 MHz, CDCl3) δ 197.1, 66.7, 56.9, 53.3, 52.9, 45.3, 41.3, 34.4, 25.2, 20.8 ppm; HRMS (ESI) calculated for C13H27N3S2 [M+H]+: 290.1725; Found: 290.1718.

2.3.8. 2-(4-isobutylpiperazin-1-yl)ethyl piperidine-1-carbodithioate (4h)
Yellow solid (210 mg, 64%); mp 64–70°C; 1H NMR (300 MHz, CDCl3) δ 4.30–4.20 (brs, 2H), 3.90–3.80 (brs, 2H), 3.47 (t, J = 7.3 Hz, 2H), 2.68–2.62 (m, 3H), 2.50–2.35 (brs, 4H), 2.05 (d, J = 7.3 Hz, 2H), 1.85–1.70 (m, 1H), 1.70–1.55 (brs, 6H), 0.86 (d, J = 6.5 Hz, 6H) ppm; 13C NMR (75 MHz, CDCl3) δ 195.4, 66.9, 57.0, 53.3, 53.1, 51.1 (2C), 33.9, 25.8, 25.3, 25.2, 24.2, 20.8 ppm; HRMS (ESI) calculated for C16H31N3S2 [M+H]+: 330.2038; Found: 330.2032.

2.3.9. 2-(4-isobutylpiperazin-1-yl)ethyl pyrrolidine-1-carbodithioate (4i)
Colorless viscous oil (252 mg, 80%); 1H NMR (300 MHz, CDCl3) δ 3.84 (t, J = 6.8 Hz, 2H), 3.58 (t, J = 6.8 Hz, 2H), 3.42 (t, J = 7.2 Hz, 2H), 2.64 (t, J = 7.6 Hz, 2H), 2.49 (brs, 4H), 2.35 (brs, 4H), 2.06–1.86 (m, 6H), 1.68 (m, 1H), 0.81 (d, J = 6.6 Hz, 6H) ppm; 13C NMR (75 MHz, CDCl3) δ 192.4, 66.7, 56.8, 54.6, 53.1, 52.7, 50.2, 31.3, 25.7, 25.0, 23.9, 20.6 ppm; HRMS (ESI) calculated for C15H29N3S2 [M+H]+: 316.1881; Found: 316.1873.

2.3.10. 2-(4-isobutylpiperazin-1-yl)ethyl morpholine-4-carbodithioate (4j)
White solid (261 mg, 79%); mp 68–75°C; 1H NMR (300 MHz, CDCl3) δ 4.27–4.01 (brs, 4H), 3.76 (t, J = 4.9 Hz, 4H), 3.53–3.46 (m, 2H), 2.73–2.67 (m, 2H), 2.56 (brs, 4H), 2.43 (brs, 4H), 2.08 (d, J = 7.4 Hz, 2H), 1.77 (m, 1H), 0.89 (d, J = 6.6 Hz, 6H) ppm; 13C NMR (75 MHz, CDCl3) δ 197.6, 66.7, 66.1, 66.1, 56.7, 53.3, 52.9, 50.5, 33.9, 25.2, 20.8 ppm; HRMS (ESI) calculated for C15H29N3OS2 [M+H]+: 332.1830; Found: 332.1827.

Scheme 3. Diversity in the synthesis of piperazines containing dithiocarbamate motif a,b (4) from secondary amines (1), CS2 (2) and DABCO salts (3). a Isolated yield. b Reaction conditions: DABCO salt (1 mmol), secondary amine (1 mmol), carbon disulfide(1.5 mmol), K2CO3 (1 mmol), solvent (5 mL), 90°C and 16 h.
2.3.11. 2-(4-benzylpiperazin-1-yl) ethyldimethyl-4-carbodithioate (4k)
Pale yellow solid (252 mg, 78%); mp 65–72 °C; 1H NMR (300 MHz, CDCl3) δ 7.35–7.22 (m, 5H), 3.55 (s, 3H), 3.52 (s, 2H), 3.45 (t, J = 7.5 Hz, 2H), 2.66–2.54 (brs, 4H), 2.54–2.38 (brs, 4H) ppm; 13C NMR (75 MHz, CDCl3) δ 197.1, 137.9, 129.2, 128.1, 127.0, 62.9, 56.8, 52.8, 45.3, 41.4, 34.4 ppm; HRMS (ESI) calculated for C16H25N3S2 [M + H]+: 324.1568; Found: 324.1565.

2.3.12. 2-(4-benzylpiperazin-1-yl) ethyldiethyl-4-carbodithioate (4l)
Yellow viscous oil (287 mg, 82%); 1H NMR (300 MHz, CDCl3) δ 7.48–7.17 (m, 5H), 4.03 (q, J = 7.0 Hz, 2H), 3.75 (q, J = 7 Hz, 2H), 2.65–2.55 (brs, 4H), 2.55–2.44 (brs, 4H), 1.28 (m, 6H) ppm; 13C NMR (75 MHz, CDCl3) δ 195.4, 137.9, 129.2, 128.1, 127.0, 62.9, 57.0, 52.8 (2C), 49.4, 46.6, 34.1, 12.4, 11.2 ppm; HRMS (ESI) calculated for C18H29N3S2 [M + H]+: 352.1881; Found: 352.1875.

2.3.13. 2-(4-benzylpiperazin-1-yl) ethyl pyrrolidine-1-carbodithioate (4m)
Yellow oil (223 mg, 64%); 1H NMR (300 MHz, CDCl3) δ 7.48–7.17 (m, 5H), 3.93 (t, J = 6.8 Hz, 2H), 3.65 (t, J = 6.8 Hz, 2H), 3.52 (s, 2H), 3.47 (t, J = 7.6 Hz, 2H), 2.69 (t, J = 7.6 Hz, 2H), 2.64–2.54 (brs, 4H), 2.54–2.36 (brs, 4H), 2.14–2.02 (m, 2H), 2.02–1.89 (m, 2H) ppm; 13C NMR (75 MHz, CDCl3) δ 192.6, 137.9, 129.2, 128.2, 126.9, 62.9, 57.0, 54.9, 52.8 (2C), 50.5, 33.2, 25.9, 24.2 ppm; HRMS (ESI) calculated for C18H27N3S2 [M + H]+: 350.1725; Found: 350.1719.

2.3.14. 2-(4-benzylpiperazin-1-yl)ethyl morpholine-4-carbodithioate (4n)
Colorless viscous oil (190 mg, 52%); 1H NMR (300 MHz, CDCl3) δ 7.37–7.22 (m, 5H), 4.30–4.01 (brs, 4H), 3.76 (t, J = 4.8 Hz, 4H), 3.53 (s, 2H), 3.49 (t, J = 7.4 Hz, 2H), 2.71 (t, J = 7.3 Hz, 2H), 2.54–2.47 (m, 8H) ppm; 13C NMR (75 MHz, CDCl3) δ 197.5, 137.9, 129.0, 128.0, 126.9, 66.3, 66.1, 62.9, 56.7, 52.8, 50.6, 45.8, 34.1, 29.5 ppm; HRMS (ESI) calculated for C18H27N3OS2 [M + H]+: 366.1674; Found: 366.1671.

2.3.15. 2-(4-allylpiperazin-1-yl)ethyl dimethylcarbamodithioate (4o)
Colorless viscous oil (153 mg, 56%); 1H NMR (300 MHz, CDCl3) δ 5.76 (m, 1H), 5.13–5.00 (m, 2H), 3.44 (s, 3H), 3.35 (t, J = 7.4 Hz, 2H), 3.27 (s, 3H), 2.90 (d, J = 6.5 Hz, 2H), 2.59 (t, J = 2.25 Hz, 2H), 2.49 (brs, 4H), 2.40 (brs, 4H) ppm; 13C NMR (75 MHz, CDCl3) δ 196.6, 134.5, 117.6, 61.3, 56.5, 52.4, 45.0, 41.1, 34.1 ppm; HRMS (ESI) calculated for C12H23N3S2 [M + H]+: 274.1412; Found: 274.1402.

2.3.16. 2-(4-allylpiperazin-1-yl)ethyl diethylcarbamodithioate (4p)
Colorless viscous oil (219 mg, 73%); 1H NMR (300 MHz, CDCl3) δ 5.91–5.81 (m, 1H), 5.13–5.00 (m, 2H), 4.02 (q, J = 7 Hz, 2H), 3.75 (q, J = 7.0 Hz, 2H), 3.54–3.35 (t, J = 7.4 Hz, 2H), 3.01–3.00 (m, 2H), 2.69 (t, J = 7.4 Hz, 2H), 2.64–2.54 (brs, 4H), 2.54–2.38 (brs, 4H), 1.27–1.25 (m, 6H) ppm; 13C NMR (75 MHz, CDCl3) δ 195.4, 134.9, 118.0, 61.7, 56.9, 52.9 (2C), 49.4, 46.6, 33.9, 12.40, 11.5 ppm; HRMS (ESI) calculated for C14H27N3S2 [M + H]+: 302.1725; Found: 302.1722.

Figure 2. Antibacterial effects of synthetic compounds on bacteria: (a) B.subtilis was sensitive to 4f (20 ± 1 mm inhibition zone) and intermediate to 4j (18 mm ± 1 inhibition zone), (b) MRSA was resistant to all of synthetic compounds, (c) K.pneumoniae was resistant to all of synthetic compounds, and (d) E.coli was sensitive to 4f (22 ± 0.6 mm inhibition zone) and intermediate to 4j (15 ± 0.7 mm inhibition zone).
2.3.17. 2-(4-allylpiperazin-1-yl) ethyl pyrrolidine-1-carbamodithioate (4q)

Yellow viscous oil (158 mg, 53%); $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 5.89–5.80 (m, 1H), 5.16 (t, $J = 13.0$ Hz, 2H), 3.92 (t, $J = 6.8$ Hz, 2H), 3.64 (t, $J = 6.8$ Hz, 2H), 3.46 (t, $J = 7.3$ Hz, 2H), 2.99 (d, $J = 6.5$ Hz, 2H), 2.68 (t, $J = 7.3$ Hz, 2H), 2.64–2.54 (brs, 4H), 2.54–2.38 (brs, 4H), 2.12–2.01 (m, 2H), 2.01–1.90 (m, 2H) ppm; $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 192.6, 134.9, 118.0, 61.7, 57.0, 54.9, 52.8, 50.5, 33.3, 25.9, 24.2 ppm; HRMS (ESI) calculated for C$_{14}$H$_{25}$N$_3$S$_2$ [M + H]$^+$: 300.1568; Found: 300.1565.

2.3.18. 2-(4-isopentylpiperazin-1-yl)ethyl dimethyl carbamodithioate (4r)

Pale yellow viscous oil (272 mg, 90%); $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 3.51 (s, 3H), 3.41 (t, $J = 7.2$ Hz, 2H), 3.34 (s, 3H), 2.65 (t, $J = 7.2$ Hz, 2H) 2.56 (brs, 4H), 2.47 (brs, 4H), 2.31 (t, $J = 5.4$ Hz, 2H), 1.54 (m, 1H), 1.41–1.30 (m, 2H), 0.86 (d, $J = 6.6$ Hz, 6H) ppm; $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 197.0, 56.7, 56.7, 52.9, 52.5, 45.2, 41.3, 35.3, 34.4, 26.5, 22.5 ppm; HRMS (ESI) calculated for C$_{14}$H$_{29}$N$_3$S$_2$ [M + H]$^+$: 304.1881; Found: 304.1881.

2.3.19. 2-(4-isopentylpiperazin-1-yl) ethyl diethyl carbamodithioate (4s)

Yellow viscous oil (271 mg, 82%); $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 4.02 (q, $J = 6.9$ Hz, 2H), 3.74 (q, $J = 7.0$ Hz, 2H), 3.45 (t, $J = 7.4$ Hz, 2H), 2.68 (t, $J = 7.4$ Hz, 2H), 2.64–2.39 (brs, 8H), 2.33 (t, $J = 7.8$ Hz, 2H), 1.64–1.48 (m, 1H), 1.43–1.32 (m, 2H), 1.30–1.20 (m, 6H), 0.88 (d, $J = 6.6$ Hz, 6H) ppm; $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 195.4, 56.9, 53.1, 52.8, 49.4, 46.5, 35.7, 33.9, 26.6, 22.7, 12.3, 11.5 ppm; HRMS (ESI) calculated for C$_{16}$H$_{33}$N$_3$S$_2$ [M + H]$^+$: 332.2194; Found: 332.2188.

2.3.20. 2-(4-isopentylpiperazin-1-yl) ethyl pyrrolidine-1-carbamodithioate (4t)

Pale yellow solid (240 mg, 73%); mp 49–52°C; $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 3.91 (t, $J = 6.8$ Hz, 2H), 3.64 (t, $J = 6.8$ Hz, 2H), 3.46 (t, $J = 7.6$ Hz, 2H), 2.67 (t, $J = 7.6$ Hz, 2H), 2.64–2.38 (brs, 8H), 2.36–2.27 (m, 1H), 2.12–2.01 (m, 2H), 2.01–1.90 (m, 2H), 1.62–1.47 (m, 1H), 1.48–1.25 (m, 2H), 0.88 (d, $J = 6.6$ Hz, 6H) ppm; $^{13}$C NMR (75 MHz, CDCl$_3$) $\delta$ 192.6, 57.0, 56.9, 54.9, 53.0, 52.9, 50.5, 35.8, 33.3, 26.6, 25.9, 24.2, 22.7 ppm; HRMS (ESI) calculated for C$_{16}$H$_{31}$N$_3$S$_2$ [M + H]$^+$: 330.2038; Found: 330.2032.

2.3.21. 2-(4-isopentylpiperazin-1-yl) ethyl morpholine-4-carbamodithioate (4u)

White solid (210 mg, 61%); mp 51–54°C; $^1$H NMR (300 MHz, CDCl$_3$) $\delta$ 4.4–4.1 (brs, 2H), 4.10–3.80 (brs, 2H), 3.84–3.61 (m, 4H), 3.53–3.41 (t, $J = 7.5$ Hz, 2H), 2.71–2.63 (t, $J = 7.5$ Hz, 2H), 2.64–2.38 (brs, 8H),

Figure 3. Antifungal activity of synthetic compounds: (a) 4d and 4v were affected on C.albicans with zone inhibition of 23 ± 1 and 22 ± 0.5 mm respectively, (b) 4f and 4v had 19 ± 1 and 22 ± 1 mm inhibition zone on T.asahii. (c) 4d, 4f and 4v with 19 ± 0.7, 19 ± 0.5 and 25 ± 1 mm inhibition zone were effect on A.niger respectively. (d) The selected compounds had no effect on F.oxysporum.
2.4.1. Microorganisms strains

2.4.2. Antimicrobial Tests

2.4.3. Agar well diffusion method for antibacterial activity assay

2.4.4. Agar well diffusion method for antifungal activity assay

2.4.5. Determination of minimum inhibitor concentration of synthetic compounds

Table 2: Numerical results of inhibition diameter zones for synthetic compounds.

| Microorganism | B.subtilis | E. coli | C.albicans | T.asahii | A.niger |
|---------------|------------|---------|------------|----------|---------|
| inhibition diameter zones in millimeters (mm) | (4f) 20 ± 1 | (4f) 22 ± 0.6 | (4f) 19 ± 1 | (4f) 19 ± 0.7 |

Table 3: Numerical results of MIC for synthetic compounds.

| Microorganism | E. coli | B.subtilis | C.albicans | T.asahii | A.niger |
|---------------|---------|------------|------------|----------|---------|
| MIC mg/mL (Average) | 2.5 | 2.5 | 2.5 | 0.625 | 5 |

*Figure 4. The results of MIC test for 4f and 4v. All experiments had three replicates. (a) 4f was tested for bacteria and (b) 4v was tested for fungi.*
with 50 μl of microbial inoculum prepared in MHB or and MHB+ %2 glucose after dilution with culture media, 1:150 v:v of standardized microbial suspension adjusted to 0.5 McFarland scale. After complete mixing, the inoculated 96-well microtitration plates were incubated in 37°C for 24–48 h [29,30,33].

Optical density measuring was used for MIC determination. Thus, by using an ELISA reader, the absorbance of microtiter plates at 570 nm and 530 nm was evaluated for bacteria and yeasts, respectively. The drug free wells were considered as positive control and the lowest concentration with optical density of less than 0.1 was considered as MIC. For molds, MIC was determined visually.

3. Results and discussion

3.1. Chemistry

Here, a straightforward and three-component procedure for the synthesis of dithiocarbamate-containing piperazines 4 is reported via the reaction of secondary amines 1, carbon disulfide 2, and quaternized derivatives of DABCO 3. Normally, design and discovery of a direct step-economy synthetic route to generate substituted piperazine rings is partly complicated; as a result, it has been garnered widespread attention in recent years. In this regard, 1,4-diazabicyclo[2.2.2]octane (DABCO) which has universally known as a base in organic reactions [28, 34], has been deployed as a reagent in C-N bond cleavage synthetic routes for the straightforward preparation of unsymmetrical piperazine compounds [22].

To optimize the reaction conditions, a model reaction was considered (Scheme 2). Initially, we observed that the reaction of DABCO salt 3b (0.5 mmol) with diethylamine (0.5 mmol) and CS2 (0.75 mmol) in THF for 24 h at 120°C without a base provided the product 4f in 40% isolated yield (Table 1, entry 1). Under these conditions, various bases such as NaOH, KOH, K3PO4, Na2CO3 and K2CO3 were used in the model reaction (Table 1, entries 2–6) and the elevated yield (89%) was achieved using K2CO3 (Table 1, entry 6). By screening the model reaction in various protic and aprotic solvents in the presence of K2CO3, we observed that higher yields were obtained in MeOH and EtOH (Table 1, entries 7–8) compare to DMF, n-hexane and CHCl3 (Table 1, entries 9–11). Using aqueous media gave inferior results (Table 1, entry 14). No desired product was obtained in CH2Cl2 and DMSO (Table 1, entry 12–13). By decreasing the reaction temperature to 50, 70 and 90°C, the corresponding product was obtained in 25, 37 and 87 %, respectively (Table 1, entries 15–17). Besides that, we observed that by decreasing the reaction time to 16 h, the yield was improved to 95% (Table 1, entry 19). At the end, deploying THF as a solvent and heating the reaction mixture to 90°C for 16 h in the presence of K2CO3 as a base was used as optimal conditions for the direct synthesis of a wide range of piperazine derivatives.

The generality of the reaction was investigated using various DABCO salts and secondary amines and the results are shown in Scheme 3. Various linear and cyclic secondary amines encompassing diethylamine, dimethylamine, morpholine, pyrrolidine, and piperidine were successfully deployed in this synthetic route to give the desired 1,4-disubstituted piperazines 4a-u in good to excellent yields. In addition, DABCO salts with primary and secondary alkyl groups are compatible with this protocol. By using bis-ammonium salt of DABCO, the corresponding N, N'-bis piperazine 4v containing two dithiocarbamate groups was obtained in moderate yield. In this MCRs, the regioselectivity seems to be related to the alkyl moiety attached to the nitrogen in DABCO salts. In the case of allyl and benzyl salts of DABCO, due to the competition between the carbon of the bridge and the alkyl group for nucleophilic attack, the corresponding alkyl dithiocabamates were obtained as main byproduct in the reaction mixture. The structures of all products were confirmed by 1H NMR, 13C NMR and HRMS analyses (Figures S1–S44 in supplementary material).

3.2. Biological studies

Based on the results, the selected synthetic compounds were effective antifungal compounds because they could inhibit yeasts and molds.

3.2.1. Antimicrobial effects of the synthetic compounds with the disk diffusion test

As shown in Figure 2, 4f, 4j and 4v were affected on different bacteria. But, based on zone inhibition diameter, only 4f was affected on B.subtilis and E.coli with zone inhibition diameter of 20 ± 1 and 22 ± 0.6 mm, respectively and had no effect on MRSA and K.pneumoniae.

Addition antifungal effects of these synthetic compounds were shown in Figure 3. 4d, 4f and 4v had antifungal activity. 4d and 4v were affected on C.albicans with zone inhibition of 23 ± 1 and 22 ± 0.5 mm, respectively, 4f and 4v had 19 ± 1 and 22 ± 1 mm inhibition zone for T.asahii. 4d, 4f and 4v with 19 ± 0.7, 19 ± 0.5 and 25 ± 1 mm inhibition zone were effected on A.niger, respectively. The results of zone inhibition diameter were summarized in Table 2.

3.2.2. Minimum inhibitory concentration (MIC) results of the selected synthetic molecules

For determination of MIC, we selected 4f (effect on both bacteria) and 4v (effect on three fungi). Figure 4 shows the MIC results of the 4f and 4v on sensitive microorganisms. The results of MIC were summarized in Table 3. As shown in Figure 4, compound 4f illustrated antibacterial activity with MIC values of 2.5 mg/mL against E.coli and B.sutilis and also compound 4v showed antifungal activity with MIC values of 2.5, 0.625 and 5 mg/mL against C.albicans, T.asahii and A.niger, respectively.

4. Conclusion

In conclusion, expansion of modern biologically active molecules inspired the organic chemists to render DABCO bond cleavage technique. In this project, a one-pot three-component reaction based on re-engineering approach has been devised for the synthesis of dithiocarbamate-containing piperazine scaffolds by in situ reaction of amines, CS2 and DABCO salts. The reaction proceeds via C-N bond cleavage of DABCO ring. A certain number of synthesized compounds revealed appropriate antibacterial activity against both Gram-positive as well as Gram-negative bacteria and depicted good to excellent antifungal activity. Finally, this synthetic route enjoys various merits namely easy workup procedure, operator friendliness, economical use of reagents, provided satisfactory yields of the small molecules.

Declarations

Author contribution statement

Azim Ziyaei Halimehjani: Conceived and designed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Faeezeh Dehghan, Yazdanbakhsh Lotfi Nosood: Performed the experiments; Analyzed and interpreted the data.

Vida Tafakori, Elaheh Amini: Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Seyyed Emad Hooshmand: Conceived and designed the experiments; Wrote the paper.
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Data included in article/supplementary material/referenced in article.

**Declaration of interests statement**

The authors declare no conflict of interest.

**Additional information**

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