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Synthesis and microbiological activity of some 4-butyl-2*H*-benzo[1,4]thiazin-3-one derivatives

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Abstract

The synthesis and physicochemical properties of 4-butyl-2*H*-benzo[1,4]thiazin-3-one derivatives are described. These new compounds were synthesised by alkylation in 4-N position and acylation and/or alkylation of 6-NH_2 by phase transfer catalysis. Acid hydrolysis of 6-alkylacylamino group yielded 6-alkylamino-4-butyl-2*H*-benzo[1,4]thiazin-3-ones. The antimicrobial in vitro activity was determined on five compounds.

Keywords: Benzothiazines; Antimicrobial activity

1. Introduction

A large number of 2*H*-benzo[1,4]thiazine derivatives exhibit different pharmacological activities: antimicrobial, anticancerous, immunostimulating, aldose reductase inhibitor [1–5]. The alkylation of 4-N position of 2*H*-benzo[1,4]thiazines affords bactericidal and antifungal derivatives [6,7].

The synthesis of 6-alkylacylamino-4-methyl-2*H*-benzo[1,4]thiazin-3-ones has been previously reported [8]. In the present paper, we describe the synthesis and the physicochemical properties of 4-butyl-2*H*-benzo[1,4]thiazin-3-one derivatives. The antimicrobial activity of five of these compounds has been investigated in vitro against cocci and bacilli.

2. Chemistry

Several steps are necessary to synthesise 4-butyl-2*H*-benzo[1,4]thiazin-3-ones. The treatment of 2-chloro-5-nitro-aniline (**1**) with sodium sulfide and sulfur [9] gave 2-amino-4-nitrobenzenethiol (sodium salt **2**), which is cyclised to 6-nitro-2*H*-benzo[1,4]thiazin-3-one (**3**) with chloroacetic acid [1]. N-butylation with butyl bromide in alkaline medium [10] afforded **4**. The reduction of the nitro group by $SnCl_2$ in acidic medium [11] gave the corresponding amine **5**. N-acylation of the amino group followed by phase transfer catalysed alkylation in basic medium [12] led to *N*-alkylacyl (**8**–**11**). Acid hydrolysis of **10** and **11** afforded 6-alkylamino-4-butyl-2*H*-benzo[1,4]thiazin-3-ones **12** and **13** (Scheme 1).

3. Experimental

Melting points were determined with a capillary Büchi apparatus and are uncorrected. The purity of the compounds is controlled by thin layer chromatography on silica pre-coated plates Merck $60F_{254}$. The spots are revealed under UV light or iodine vapour and the R_f values are measured.

IR spectra were recorded in KBr tablets (2%) or film with a Perkin–Elmer 1310 spectrophotometer. ^1H and ^{13}C NMR spectra were recorded with a Brucker AC 200 spectrometer in DMSO-d_6. Chemical shifts (δ) are

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expressed in ppm and coupling constants (J) in Hertz (Hz). Electronic impact mass spectra (70 eV) were measured with an R-1010C Delsi-Nermag spectrometer. No elemental analysis was performed on the new compounds but all mass spectra agree with their proposed structure.

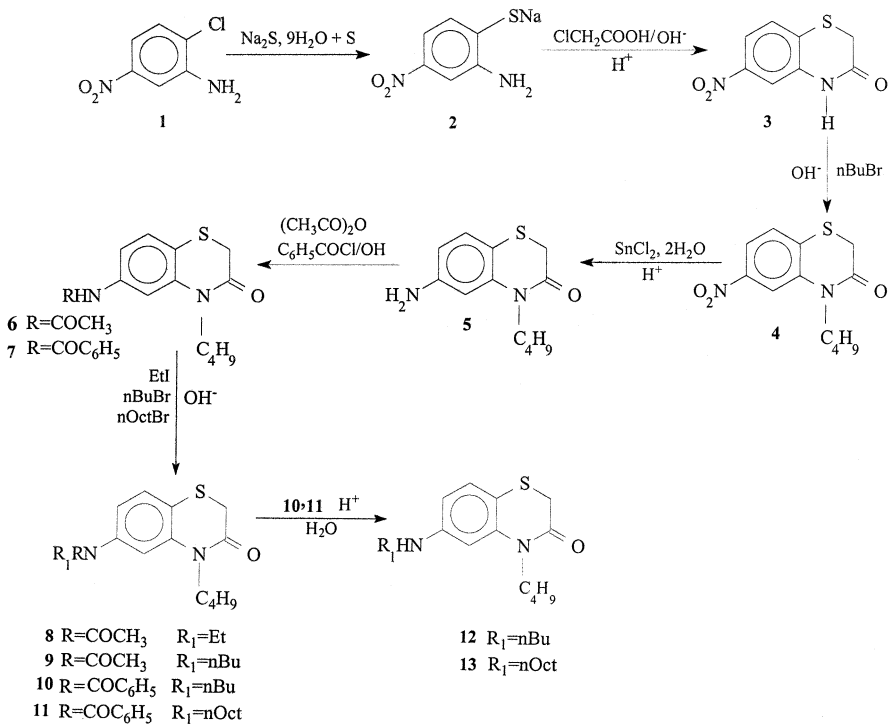
The synthesis, physical and analytical properties of compounds **2** and **3** have been previously described [8].

3.1. 4-Butyl-6-nitro-2H-benzo[1,4]thiazin-3-one (4)

A mixture of compound **3** (1.05 g, 5 mmol) and potassium hydroxide (0.56 g, 10 mmol) in DMSO/ethanol (10:12.5 ml) is stirred at room temperature for 10 min before butyl bromide (10 mmol) was added dropwise. The mixture is heated at 50°C under stirring for 15 h. After cooling, the crude butyl product precipitated when water or crushed ice is added. It is collected and recrystallised from 95% ethanol. Yield 68%. M.p. 86–87°C. TLC: ($C_6H_5CH_3/CH_3COOC_2H_5$ 6:4) R_f 0.79. IR (KBr): ν 2930, 2850, 1680, 1520, 1350, 745 cm^{-1}. ^1H NMR (DMSO-d_6): δ 0.87 (t, 3H, CH_3-4, $J = 7.2$ Hz), 1.12–1.32 (m, 2H, CH_2-4), 1.43–1.57 (m, 2H, CH_2-4), 3.62 (s, 2H, CH_2-2), 4.06 (t, 2H, CH_2-4, $J = 7.2$ Hz), 7.69 (d, 1H, aromatic H, $J = 8.5$ Hz), 7.87 (dd, 1H, aromatic H, $J = 8.5$ and 2.2 Hz), 8.00 (d, 1H, aromatic H, $J = 2.2$ Hz). ^{13}C NMR (DMSO-d_6, BB decoupling and DEPT): δ 13.5 (CH_3), 19.1 (CH_2), 28.6 (CH_2), 29.7 (CH_2), 43.0 (CH_2), 112.4 (CH), 117.5 (CH), 128.9 (CH), 132.8 (C), 139.2 (C), 146.4 (C), 164.2 (CO). MS; m/z (%): 266 (45.2), 219 (35.7), 210 (100), 195 (42), 181 (32.6), 149 (32.2), 135 (18.4).

3.2. 6-Amino-4-butyl-2H-benzo[1,4]thiazin-3-one (5)

To a cold and stirred solution of 3.72 g (16 mmol) of stannous chloride dihydrate in 4 ml of concentrated HCl is added portionwise, over a 15 min period, 3.5 mmol of compound **4**. The mixture is left under stirring for 15 min in an ice-bath. Then the temperature is gradually increased up to reflux, which is kept for 2 h. After cooling, the precipitate is collected, washed with water to eliminate excess of HCl and suspended in water. A 20% hydroxide solution is added (pH 10) to give the corresponding amine, which is collected by filtration, washed with water and 10% NaOH and recrystallised from water. Yield 64%. M.p. 63–65°C (hydrochloride m.p. 218–220°C). TLC: ($CHCl_3/CH_3COCH_3$ 9:1) R_f 0.71. IR (KBr): ν 3460, 3370, 2980, 2865, 1650 cm^{-1}. ^1H NMR (DMSO-d_6): δ 0.88 (t, 3H, CH_3-4, $J = 7.2$ Hz), 1.18–1.37 (m, 2H, CH_2-4), 1.44–1.58 (m, 2H, CH_2-4), 3.32 (s, 2H, CH_2-2), 3.82 (t, 2H, CH_2-4, $J = 7.2$ Hz), 5.22 (s, 2H, NH_2), 6.28 (dd, 1H, aromatic H, $J = 8.2$ and 1.6 Hz), 6.54 (d, 1H, aromatic H, $J = 1.6$ Hz), 6.98 (d, 1H, aromatic H, $J = 8.2$ Hz). ^{13}C NMR (DMSO-d_6, BB decoupling and DEPT): δ 13.6 (CH_3), 19.3 (CH_2), 29.0 (CH_2), 31.5 (CH_2), 43.2 (CH_2), 103.7 (CH), 107.5 (C), 109.4 (CH), 128.5 (CH),



Scheme 1. Synthesis of 4-butyl-2*H*-benzo[1,4]thiazin-3-one derivatives.

139.8 (C), 148.5 (C), 165.0 (CO). MS; *m*/*z* (%): 236 (79.2), 193 (14.1), 180 (85.1), 165 (48.5), 151 (100), 135 (16.7), 77 (16.5).

3.3. 6-Acetylamino-4-butyl-2H-benzo[1,4]thiazin-3-one (6)

A mixture of 1.18 g (5 mmol) of compound **5** and 10 ml of acetic anhydride is refluxed for 10 min. The excess of anhydride is hydrolysed by 10 ml of water and the mixture is again refluxed for 5 min. The precipitate is washed with water and recrystallised from 95% ethanol. Yield 87%. M.p. 103–105°C. TLC: ($CHCl_3$/CH_3COCH_3 9:1) R_f 0.78. IR (KBr): *v* 3310, 2940, 2860, 1680, 1645 (large) cm^{-1}. ^1H NMR (DMSO-d_6): δ 0.91 (t, 3H, CH_3-4, $J = 7.1$ Hz), 1.19–1.37 (m, 2H, CH_2-4), 1.47–1.60 (m, 2H, CH_2-4), 2.04 (s, 3H, $COCH_3$), 3.43 (s, 2H, CH_2-2), 3.86 (t, 2H, CH_2-4, $J = 7.1$ Hz), 7.20–7.30 (m, 2H, aromatic H), 7.67 (s, 1H, aromatic H), 10.07 (s, 1H, NH-6). ^{13}C NMR (DMSO-d_6, BB decoupling and DEPT): δ 13.5 (CH_3), 19.3 (CH_2), 24.0 (CH_3), 28.9 (CH_2), 30.7 (CH_2), 43.4 (CH_2), 108.5 (CH), 113.7 (CH), 116.4 (C), 128.1 (CH), 138.7 (C), 139.2 (C), 164.7 (CO), 168.4 (CO). MS; *m*/*z* (%): 278 (100), 222 (26.5), 207 (39.8), 193 (29.4), 180 (87.3), 165 (31.5), 151 (81.3), 135 (17.1), 43 (69.1).

3.4. 6-Benzoylamino-4-butyl-2H-benzo[1,4]thiazin-3-one (7)

To a suspension of 1.18 g (5 mmol) of compound **5** in 20 ml of 5% NaOH, 2 ml of benzoyl chloride were added dropwise. The mixture is vigorously stirred for 10 min. The precipitate is separated, washed with water and recrystallised from 95% ethanol. Yield 82%. M.p. 139–140°C. TLC: ($C_6H_5CH_3$/$CH_3COOC_2H_5$ 6:4) R_f 0.64. IR (KBr): *v* 3320, 2960, 2865, 1670, 1640 cm^{-1}. ^1H NMR (DMSO-d_6): δ 0.89 (t, 3H, CH_3-4, $J = 7.2$ Hz), 1.21–1.39 (m, 2H, CH_2-4), 1.50–1.65 (m, 2H, CH_2-4), 3.46 (s, 2H, CH_2-2), 3.92 (t, 2H, CH_2-4, $J = 7.2$ Hz), 7.35 (d, 1H, aromatic H, $J = 8.4$ Hz), 7.48–7.62 (m, 4H, aromatic H), 7.82 (d, 1H, aromatic H, $J = 1.7$ Hz), 7.94 (dd, 2H, aromatic H, $J = 7.9$ and 1.7 Hz), 10.31 (s, 1H). ^{13}C NMR (DMSO-d_6, BB decoupling and DEPT): δ 13.6 (CH_3), 19.3 (CH_2), 29.0 (CH_2), 30.7 (CH_2), 43.4 (CH_2), 109.8 (CH), 115.0 (CH), 117.4 (C), 127.6 (2CH), 128.1 (CH), 128.3 (2CH), 131.6 (CH), 134.7 (C), 138.5 (C), 139.1 (C), 164.7 (CO), 165.6 (CO). MS; *m*/*z* (%): 340 (46.3), 284 (12.9), 235 (8.1), 151 (8.4), 105 (100), 77 (75), 51 (10.3).

3.5. Alkylation of the amino group by phase transfer catalysis [12]

A mixture of 5 mmol of acylated compounds dissolved in toluene (100 ml), 1.4 g of potassium carbonate, 7.0 g of sodium hydroxide and 0.16 g of tetrabutylammonium bromide is refluxed under vigorous stirring. The suitable alkyl halide (7.5 mmol) in toluene (10 ml) is added dropwise. Stirring is continued for 4 h at the refluxing temperature. After cooling, the mixture is filtered and the filtrate added with water (50 ml); the organic phase is separated, washed with water, dried over anhydrous magnesium sulfate and evaporated.

We have carried out N-ethylation (ethyl iodide) and N-butylation (butyl bromide) on the acetyl compounds and N-butylation (butyl bromide) and N-octylation (octyl bromide), on the benzoyl ones.

3.6. 6-Acetylethylamino-4-butyl-2H-benzo[1,4]thiazin-3-one (8)

Yield 57%. M.p. 91–93°C. TLC: ($C_6H_5CH_3$/$CH_3COOC_2H_5$ 6:4) R_f 0.38. IR (KBr): *v* 2945, 2855, 1655 (large) cm^{-1}. ^1H NMR (DMSO-d_6): δ 0.83 (t, 3H, CH_3-4, $J = 7.1$ Hz), 0.99 (t, 3H, CH_3-6, $J = 7.1$ Hz), 1.13–1.31 (m, 2H, CH_2-4,), 1.35–1.50 (m, 2H, CH_2-4), 1.76 (s, 3H, $COCH_3$), 3.51 (s, 2H, CH_2-2), 3.63 (q, 2H, CH_2-6, $J = 7.1$ Hz), 4.02 (t, 2H, CH_2-4, $J = 7.1$ Hz), 6.96 (dd, 1H, aromatic H, $J = 7.9$ and 1.8 Hz), 7.27 (d, 1H, aromatic H, $J = 1.8$ Hz), 7.45 (d, 1H, aromatic H, $J = 7.9$ Hz). ^{13}C NMR (DMSO-d_6, BB decoupling and DEPT): δ 12.9 (CH_3), 13.6 (CH_3), 19.1 (CH_2), 22.5 (CH_3), 28.8 (CH_2), 30.3 (CH_2), 42.3 (CH_2), 42.8 (CH_2), 118.5 (CH), 122.6 (CH), 122.9 (C), 128.8 (CH), 139.4 (C), 141.5 (C), 164.5 (CO), 168.4 (CO). MS; *m*/*z* (%): 306 (89.7), 264 (16), 249 (21.1), 193 (36.4), 179 (26.6), 84 (22.6), 70 (100), 43 (51.7).

3.7. 6-Acetybutylamino-4-butyl-2H-benzo[1,4]thiazin-3-one (9)

Yield 36%. M.p. 117–118°C. TLC: ($CHCl_3$/C_2H_5OH 9:1) R_f 0.87. IR (KBr): *v* 2930, 2860, 1650 (large) cm^{-1}. ^1H NMR (DMSO-d_6): δ 0.83 (t, 6H, CH_3-4, $J = 7.1$ Hz), 1.16–1.45 (m, 8H, $4CH_2$), 1.79 (s, 3H, $COCH_3$), 3.51 (s, 2H, CH_2-2), 3.61 (t, 2H, CH_2-6, $J = 7.1$ Hz), 4.03 (t, 2H, CH_2-4, $J = 7.1$ Hz), 6.96 (dd, 1H, aromatic H, $J = 8.1$ and 1.8 Hz), 7.27 (d, 1H, aromatic H, $J = 1.8$ Hz), 7.45 (d, 1H, aromatic H, $J = 8.1$ Hz). ^{13}C NMR (DMSO-d_6, BB decoupling and DEPT): δ 13.6 (CH_3), 13.7 (CH_3), 19.0 (CH_2), 19.4 (CH_2), 22.5 (CH_3), 28.8 (CH_2), 29.4 (CH_2), 30.4 (CH_2), 42.3 (CH_2), 47.6 (CH_2), 118.3 (CH), 122.5 (CH), 122.8 (C), 128.8 (CH), 139.4 (C), 141.7 (C), 164.5 (CO), 168.6 (CO). MS; *m*/*z* (%): 334 (49.4), 291 (11.6), 249 (46.2), 193 (26.9), 163 (25.3), 151 (28.6), 135 (12.8), 98 (36.8), 43 (100).

3.8. 6-Benzoylbutylamino-4-butyl-2H-benzo[1,4]-thiazin-3-one (**10**)

Yield 41%. M.p. 110–113°C. TLC: ($C_6H_5CH_3$/$CH_3COOC_2H_5$ 6:4) R_f 0.69. IR (KBr): ν 2930, 2860, 1660, 1630 cm^{-1}. ^1H NMR (DMSO-d_6): δ 0.77 (t, 3H, CH_3-6, $J = 6.1$ Hz), 0.86 (t, 3H, CH_3-4, $J = 7.1$ Hz), 0.97–1.11 (m, 4H, $2CH_2$), 1.20–1.39 (m, 2H, CH_2), 1.41–1.57 (m, 2H, CH_2), 3.40 (s, 2H, CH_2-2), 3.73–3.85 (m, 4H, $2CH_2$), 6.90 (dd, 1H, aromatic H, $J = 8.2$ and 1.7 Hz), 7.00 (d, 1H, aromatic H, $J = 1.7$ Hz), 7.20–7.32 (m, 6H, aromatic H). ^{13}C NMR (DMSO-d_6, BB decoupling and DEPT): δ 13.5 (CH_3), 13.6 (CH_3), 19.0 (CH_2), 19.5 (CH_2), 28.4 (CH_2), 29.2 (CH_2), 30.4 (CH_2), 42.3 (CH_2), 49.0 (CH_2), 118.7 (CH), 121.7 (C), 122.1 (CH), 127.7 (2CH), 128.0 (2CH), 128.5 (CH), 129.3 (CH), 136.5 (C), 138.8 (C), 142.0 (C), 164.3 (CO), 169.1 (CO). MS; m/z (%): 396 (19.4), 291 (8.4), 249 (6.3), 177 (9.9), 160 (38.1), 105 (100), 77 (50.1).

3.9. 6-Benzoyloctylamino-4-butyl-2H-benzo[1,4]-thiazin-3-one (**11**): liquid compound

Yield 83%. TLC: ($C_6H_5CH_3$/$CH_3COOC_2H_5$ 8:2) R_f 0.50. IR (KBr): ν 2930, 2860, 1670, 1640 cm^{-1}. ^1H NMR (DMSO-d_6): δ 0.83 (t, 3H, CH_3-4, $J = 6.1$ Hz), 0.77 (t, 3H, CH_3-6, $J = 5.9$ Hz), 1.05–1.08 (m, 4H, $2CH_2$), 1.22 (large s, 10H, $5CH_2$), 1.45–1.55 (m, 2H, CH_2-4), 3.40 (s, 2H, CH_2-2), 3.75–3.80 (m, 4H, CH_2-4), 6.90 (dd, 1H, aromatic H, $J = 8.2$ and 1.9 Hz), 6.99 (d, 1H, aromatic H, $J = 1.9$ Hz), 7.20–7.32 (m, 6H, aromatic H). ^{13}C NMR (DMSO-d_6, BB decoupling and DEPT): δ 13.5 (CH_3), 13.9 (CH_3), 19.0 (CH_2), 22.0 (CH_2), 26.2–28.6 ($5CH_2$), 30.4 (CH_2), 31.1 (CH_2), 42.3 (CH_2), 49.2 (CH_2), 118.6 (CH), 121.7 (C), 122.1 (CH), 127.7 (2CH), 128.0 (2CH), 128.5 (CH), 129.3 (CH), 136.5 (C), 138.5 (C), 142.0 (C), 164.3 (CO), 169.1 (CO). MS; m/z (%): 452 (0.6), 347 (8.5), 249 (13.1), 177 (12.8), 105 (100), 77 (48.7), 43 (13.3), 41 (19.3).

3.10. Hydrolysis of benzoyl compounds

A suspension of benzoyl compound (2 mmol) in 5 ml of 70% sulfuric acid is refluxed at 147–150°C for 30 min. After cooling, 6 ml of water is added and the mixture poured into 20 ml of ice water. The precipitate is extracted twice by 20 ml of chloroform. Ammonium hydroxide (28% solution, 10 ml) is added to the organic phase, which is washed with water, dried over magnesium sulfate and vacuum evaporated. The crude product is treated with petroleum ether and ethyl acetate and then recrystallised from methanol.

3.11. 4-Butyl-6-butylamino-2H-benzo[1,4]thiazin-3-one (**12**)

Yield 60%. M.p. 73–74°C. TLC: ($C_6H_5CH_3$/$CH_3COOC_2H_5$ 8:2) R_f 0.59. IR (KBr): ν 3360, 2940, 2860, 1665 cm^{-1}. ^1H NMR (DMSO-d_6): δ 0.88 (t, 3H, CH_3-4, $J = 7.0$ Hz), 0.90 (t, 3H, CH_3-6, $J = 6.9$ Hz), 1.22–1.37 (m, 4H, $2CH_2$), 1.44–1.57 (m, 4H, $2CH_2$), 2.98 (dt, 2H, CH_2-6), 3.32 (s, 2H, CH_2-2), 3.86 (t, 2H, CH_2-4, $J = 7.2$ Hz), 5.73 (t, 1H, NH-6, $J = 4.8$ Hz), 6.27 (dd, 1H, aromatic H, $J = 8.1$ and 1.9 Hz), 6.47 (d, 1H, aromatic H, $J = 1.9$ Hz), 7.03 (d, 1H, aromatic H, $J = 8.1$ Hz). The NH proton at 5.73 ppm was confirmed by the irradiation of the double triplet at 2.98 ppm of 6-NCH_2. ^{13}C NMR (DMSO-d_6, BB decoupling and DEPT): δ 13.6 (CH_3), 13.7 (CH_3), 19.3 (CH_2), 19.7 (CH_2), 29.1 (CH_2), 30.7 (CH_2), 31.5 (CH_2), 42.4 (CH_2), 43.1 (CH_2), 101.7 (CH), 107.1 (CH), 107.3 (C), 128.5 (CH), 139.8 (C), 148.7 (C), 165.1 (CO). MS; m/z (%): 292 (100), 249 (90.2), 207 (21.1), 193 (56.2), 177 (16.7), 163 (51.9), 151 (26.5), 41 (33.9).

3.12. 4-Butyl-6-octylamino-2H-benzo[1,4]thiazin-3-one (**13**)

Yield 33%. M.p. 69–71°C. TLC: ($C_6H_5CH_3$/$CH_3COOC_2H_5$ 6:4) R_f 0.71. IR (KBr): ν 3320, 2900, 2840, 1660 cm^{-1}. ^1H NMR (DMSO-d_6): δ 0.81–0.91 (m, 6H, $2CH_3$), 1.22–1.37 (m, 12H, $6CH_2$), 1.47–1.54 (m, 4H, $2CH_2$), 2.97 (dt, 2H, CH_2-6, $J = 6.6$ Hz), 3.31 (s, 2H, CH_2-2), 3.86 (t, 2H, CH_2-4, $J = 7$ Hz), 5.72 (t, 1H, NH-6), 6.26 (dd, 1H, aromatic H, $J = 8.4$ and 2.1 Hz), 6.46 (d, 1H, aromatic H, $J = 2.1$ Hz), 7.03 (d, 1H, aromatic H, $J = 8.4$ Hz). ^{13}C NMR (DMSO-d_6, BB decoupling and DEPT): δ 13.6 (CH_3), 13.8 (CH_3), 19.3 (CH_2), 22.0 (CH_2), 26.6–29.1 ($5CH_2$), 31.2 (CH_2), 31.6 (CH_2), 42.7 (CH_2), 43.1 (CH_2), 101.5 (CH), 107.2 (CH, C), 128.5 (CH), 139.8 (C), 148.7 (C), 165.1 (CO). MS; m/z (%): 348 (68.9), 249 (100), 193 (39.2), 177 (12.7), 163 (27.9), 151 (25.8), 43 (26.8), 41 (24.3).

4. Biological activity

The calculation of minimal inhibitory concentration (MIC) was performed in vitro against six microorganisms (cocci, Gram-positive and Gram-negative bacilli): *Micrococcus flavus* — DAUFPE 323, *Bacillus cereus* — DAUFPE 11, *Salmonella enteritidis* — DAUFPE 415, come from the collection of the Department of Antibiotics of the Federal University of Pernambuco; *Staphylococus aureus*, *Escherichia coli* and *Proteus vulgaris* are wild strains isolated from contaminated food.

The Mueller–Hinton agar has been used as a medium of reference for antimicrobial activity tests [13]. Inocula were prepared from 18 h-old subcultures at

Table 1
Antimicrobial activity of investigated compounds (MIC in μg/ml)

Microorganism	Compounds					
	4	**6**	**7**	**9**	**12**	Ciprofloxacin
M. flavus	2	64	[a]	32	64	0.25
S. aureus	[a]	[a]	[a]		[a]	4
B. cereus	4	[a]	[a]	[a]	[a]	2
S. enteriditis	[a]	64	16	[a]	[a]	8
E. coli	[a]	[a]	16	[a]	[a]	2
P. vulgaris	[a]	64	[a]	[a]	[a]	4

[a] ≥128 μg/ml.

37°C in this broth. The turbidity of culture suspension was adjusted to obtain 0.5 on the Mac Farland scale, i.e. 10^8 UFC/ml. The solutions of each drug were prepared at the concentration of 1280 μg/ml in a mixture of DMSO/Tween 80/distilled water (1:1:8) and then in distilled water from mother solutions. Serial dilutions were made with sterile distilled water according to a geometric progression of ratio 2, such as the concentrations of Petri plates which were in the range of 128–0.5 μg/ml. In the same way, it was verified that DMSO, at the 1/10 dilution, was completely inactive on the tested microorganisms.

Plates of Petri are prepared by mixing one part of each dilution of drug with nine parts of the Mueller–Hinton agar medium. Each plate, including a drug-free control plate, is inoculated by streak using a calibrated loop (0.05 ml). The plates are examined for the presence or absence of growth after 18 h of incubation at 37°C. The MIC is considered to be the lowest drug concentration for which there is no microbial growth [14]. Ciprofloxacin was used as the reference antibiotic. The antimicrobial activity of investigated compounds is shown in Table 1.

5. Results

Some new 2*H*-benzo[1,4]thiazin-3-one derivatives have been synthesised and characterised by their physical and analytical properties. The fragments observed by electronic impact mass spectrometry are in agreement with the proposed structures.

Five compounds were evaluated as potential antimicrobial agents against six microorganisms. Their activity is inferior to that of ciprofloxacin, the reference antibiotic.

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