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SYNTHESIS AND ANTIBACTERIAL ACTIVITY OF NEW (Z)-2-(BENZYLIDENE)-3,4-DIHYDRO-2H-[1,4]-BENZOTHIAZIN-3-ONE D-GLUCOSE DERIVATIVES

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Abstract: In this work, we are interested in the synthesis of new N-glucosyl 1,4-benzothiazin-3-ones by grafting 5,6-anhydro-3-O-alkyl-1,2-O-isopropylidene-α-D-glucofuranoses onto (Z)-2-benzylidene-3,4-dihydro-2H-1,4-benzothiazin-3-one **1**. Deprotection of compounds **3a-c** was performed in the mixture CF_3COOH-H_2O (9/1: v/v) leading to the corresponding compounds **4a-c** with good yields. The structures of the synthesized compounds have been characterized using ^1H NMR and ^{13}C NMR. Compounds **3a-c** and **4a-c** were subjected to the evaluation of antibacterial activity. Some compounds tested showed significant activity.

Keywords: *alkylation, anti-bacterial activities, 1,4-benzothiazin-3-one, D-glucofuranose, D-glucose*

INTRODUCTION

Compounds containing 1,4-benzothiazine backbone have been studied extensively both in academic, agricultural and industrial areas [1 – 6]. These molecules exhibit a wide range of biological applications indicating that the 1,4-benzothiazine moiety is a template potentially useful in medicinal chemistry research and biological activities as anti-inflammatory [7 – 9], analgesic [9], antibacterial [10 – 12], anticancer [13, 14], anticonvulsant [15], antidiabetic [16], antifungal [17], anthelmintic [18, 19], anti-HCV [20], antimalarial [21], antimicrobial [22 – 26], antiproliferative [27], antipsychotropic [28], antiviral agents [29], antinociceptive [30], and calcium antagonist [31]. Furthermore, 1,4-benzothiazine moiety is the core structure in many drugs such as antibiotic **type I** and blood cholesterol lowering drug **type II** (Figure 1) [32].
1,4-Benzothiazine derivatives have been shown to possess a number of effects in various *in vivo* and *in vitro* experimental systems [33].
In this work we describe the synthesis of **type III** compounds, obtained by grafting 5,6-anhydro-3-O-alkyl-1,2-O-isopropylidene -α-D-glucofuranoses onto (Z)-2-benzylidene-3,4-dihydro-2H-1,4-benzothiazin-3-one**1**.The glycosylation reaction involves regioselectively the nitrogen atom of the lactam function of compound 1 as observed in previous works done by Lakhrissi *et al.* [34] and Bouhlal *et al.* [35, 36] (Figure 1).

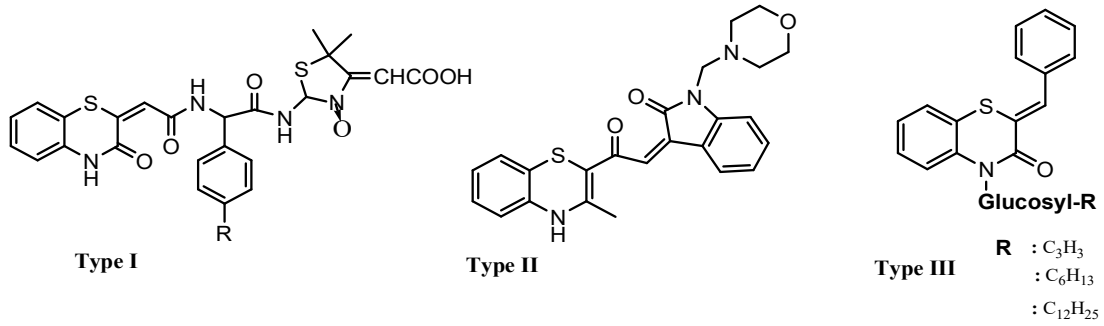


Figure 1. *Examples of bioactive molecules derived from 1,4-benzothiazin-3-one*

This work also aims to develop a new compound with antibacterial properties against gram-negative bacteria, *Escherichia coli ATCC 25922*, *Pseudomonas aeruginosa ATCC 27853*, *Acinetobacter ATCC 17978*, *Escherichia coli ESBL, Klebsiella pneumonia ESBL and Acinetobacter* ESBL and gram-positive bacteria, *Staphylococcus aureus ATCC 25923 and Staphylococcus aureus MLSB,* using disc diffusion method [37]. The MIC was measured in $\mu g \cdot mL^{-1}$ and the activity was compared with standard drug Chloramphenicol.

MATERIALS AND METHODS

Spectral data measurements

All chemicals were purchased from Sigma-Aldrich or Acros Organics (France). All solvents were distilled before use. The spectroscopic characterization of the synthesized compounds was achieved by recording NMR spectra, which were measured on a Bruker

Avance DPX 300 instrument. The chemical shifts (δ) were expressed in ppm and the coupling constants (J) in Hertz (Hz), down field from TMS (tetramethylsilane, ($Si(CH_3)_4$) which has been assigned a chemical shift of zero, TMS as an internal reference. Thin layer chromatography (TLC) and column chromatography were carried out on silica plates (Merck 60 F254) and silica gel (Merck 60, 230-400 mesh), respectively. Melting points of compounds (**3a–c**) and (**4a–c**) were determined in open capillaries.

Experimental part:

General procedure (step i)

To a solution of (Z)-2-(benzylidene)-3,4-dihydro-2H-[1,4]-benzothiazin-3-one **1** (0.50 g, 1.97 mmol), K_2CO_3 (0.54 g, 3.94 mmol) in DMSO or 4:1 toluene-DMSO (15 mL) at 100-110 °C, was added activated carbohydrate derivative (0.95 g, 3.94 mmol) of **2a**; (1.13 g, 3.94 mmol) of **2b**) and (1.46 g, 3.94 mmol) of **2c**. When no more starting material was detected by TLC, the mixture was concentrated under reduced pressure. The residue obtained was extracted with toluene-water mixture and the organic phase was separated, washed with a saturated aqueous solution of NaCl, dried on Na_2SO_4, and concentrated under reduced pressure. The crude isolated was chromatographed on silica gel (hexane-ethyl acetate, (9/1: v/v)).

General procedure for deprotection reaction (step ii)

The protected derivative **3a** (0.50 g, 1.01 mmol); or **3b** (0.50 g, 0.93 mmol); or **3c** (0.50 g, 0.80 mmol), was added to a stirred solution of 9:1 CF_3COOH-water (15 mL) at room temperature. When no more starting material was detected by TLC, the solution was concentrated to dryness under reduced pressure. The crude obtained was chromatographed on silica gel (hexane- ethyl acetate, (9/1: v/v)).

Antibacterial activity

Microorganisms used

The synthesized compounds were evaluated for their *in vitro* antibacterial activity against both $Gram^+$ bacteria: *Staphylococcus aureus ATCC 25923* and *Staphylococcus aureus MLSB* and six bacteria $Gram^-$: *Escherichia coli ATCC 25922*, *Pseudomonas aeruginosa ATCC 27853*, *Acinetobacter baumannii ATCC 19606*, *Escherichia coli ESBL, Klebsiella pneumonia ESBL and Acinetobacter baumannii ESBL*, the strains used in this work widely encountered in various pathologies in humans, were obtained from the Department of Microbiology, National Institute of Hygiene, Rabat, Morocco.

Antibacterial test by gel diffusion method

For testing antibacterial activity, we used the technique of diffusion by mid agar (MH) on Petri dish [37]. The media were inoculated with a few milliliters of the bacterial inoculum so as to cover the entire agar surface. The tests were performed according to the method of Vincent (aromatogram). The latter is to remove the filter paper discs impregnated our products dissolved in DMSO 1 % on the surface of agar in petri dishes previously seeded by inoculation. The dishes were then incubated in an oven at 37 °C for 24 hours. The biological activity manifests itself by the appearance of a halo of

inhibition of microbial growth around the discs containing the test product. The reading is performed by measuring the diameter of inhibition observed.

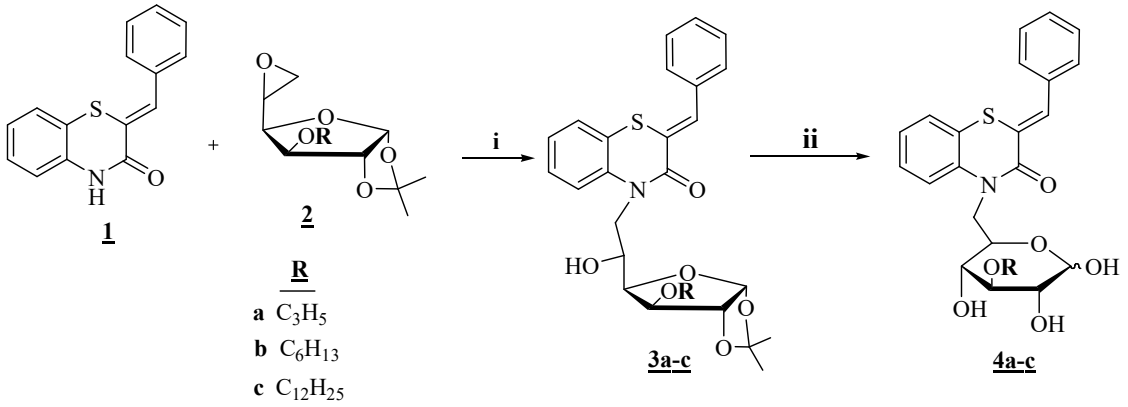
Minimum Inhibitory Concentration (MIC)

For MIC determination, we adopted the technique of sterile micro titer micro plates using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) as an indicator of sustainability. In each case, we poured 100 µL of liquid culture medium MH more 100 µL of the test product. Serial dilutions were then performed. Each well was then inoculated with10 µL of the bacterial suspension. At the end of the incubation period at the appropriate temperature, 10 µL of MTT (0.4 $mg{\cdot}mL^{-1}$) was added to each well. The plates were reincubated for 30 minutes at 37 °C. After incubation time, wells, where microbial growth occurred, showed a blue-violet color. The MIC was then determined and corresponded to the lowest concentration substance which produced no bacterial growth.

RESULTS AND DISCUSSIONS

Glycosylation of (Z)-2-benzylidene-2H-[1,4]benzothiazin-3(4H)-one (3a–c) and (4a–c)

The glucofuranosyl 1,4-benzothiazine derivatives **3a-c** (**type I**) were obtained by the regiospecific condensation of (Z)-2-(benzylidene)-3,4-dihydro-2H[1,4]-benzothiazin-3-one **1** with the anhydroglucosyl substrates **2a-c.** These precursors were synthesized according to the literature [34 – 40]. The reaction has been carried out in the presence of K_2CO_3 at 110 °C using DMSO as solvent. Under these conditions we obtained the desired products **3a-c** with (91-94 %) yield (Scheme 1).



Reagents and conditions: (**i**) K_2CO_3, DMSO, 100-110 °C, 2-4 h;
(**ii**) 9:1 $CF_3COOH–H_2O$, rt, 1 H.

Scheme 1. *Syntheses of [1,4]-benzothiazin-3-one derivatives:* ***3a–c*** *and* ***4a–c***

The progress of the reaction is monitored by CCM, eluent: hexane/ethyl acetate (9/1: v/v). The reaction crude obtained was chromatographed on a column silica gel: using as eluent hexane/ethyl acetate (9/1: v/v). The expected products **3a-c** were isolated in oily form. Their structures were identified on the basis of ^1H and ^{13}C NMR

spectral data. The ^{1}H and ^{13}C NMR spectra taken from the $CDCl_3$ of the compounds **3a-c** show the presence in particular of two singlets at 1.41 and 1.58 ppm corresponding to the two isopropyl CH_3 group of the sugar moiety; two signals at 26.33 and 26.88 ppm (compound **3a)**; 26.34 and 26.89 ppm (compounds **3b** and **3c**) corresponding to the two isopropyl CH_3 carbons. In order to obtain amphiphilic molecules, we restored the hydrophilic heads of compounds 4a-c by releasing the C-1, C-2 and C-4 protected hydroxyl groups from the glucofuranose pattern (Scheme 1). The deprotection of the isopropylidene groups compounds **3a-c** with 9:1 CF_3CO_2H–H_2O, afforded the expected glucopyranosyl benzothiazine derivatives **4a-c** (**type ii**) in good yields (79-82 %) (Scheme 1).

All these structures were confirmed by NMR spectroscopic analyses. The ^{1}H NMR and ^{13}C NMR spectra of these compounds showed in particular two doublets at 4.61 and 5.34 ppm (compound **4a**); 4.67 and 5.34 ppm (compound **4b**) and 4.64 and 5.32 ppm (compound **4c**) corresponding to the protons bound to α and β anomeric carbons, confirming that the glucosidic moieties are in pyranose forms. The ^{13}C NMR spectra highlight in particular the presence of two other signals at 92.43, 96.89 ppm (compound **4a**); 92.45, 96.97 ppm (compound **4b**) and 92.45, 96.97 ppm (compound **4c**) corresponding respectively to the α, β anomeric carbons.

(2Z)-2-benzylidene-4-(6-deoxy-1,2-O-isopropylidene-3-O-allyl-a-D-glucofuranos-6-yl)-2H-[1,4]-benzothiazin-3(4H)-one (3a):

Yield: 93 %; **brown oil**; **^{1}H NMR** ($CDCl_3$ with 0.05 % v/v TMS, 400 MHz): δ_H 7.97 (1H, s, CH_{vinyl}), 7.06-7.69 (9H, m, H_{arom}), 6.08 (1H, d, $J_{1,2}$ = 3.7 Hz, H-1), 5.92-6.05 (1H, m, H_{allyl}), 5.34-5.41 (1H, m, H_{allyl}), 5.24-5.28- (1H, m, H_{allyl}), 4.67 (1H, d, $J_{2,1}$ = 3.7 Hz, H-2), 4.59, 4.43 (2H, 2dd, $J_{6a,6b}$ = 14.6 Hz, $J_{6a,5}$ = 2.7 Hz, $J_{6b,5}$ = 5.7 Hz, H-6), 4.38-4.40 (1H, m, H-5), 4.23-4.26 (2H, m, O-CH_2), 4.21 (1H, dd, $J_{4,5}$ = 8.6 Hz, $J_{4,3}$ = 3.9 Hz, H-4), 4.15 (1H, d, $J_{3,4}$ = 3.9 Hz, H-3), 1.58 (s, 3H, CH_{3iso}), 1.41 (s, 3H, CH_{3iso}); **^{13}C NMR** (62.5 MHz, $CDCl_3$): δ_C 165.04 (CO), 137.11, 134.60, 120.47, 119.50, 135.81 (Cq), 134.26 (CH_{vinyl}), 130.35, 129.11, 128.46, 127.50, 126.30, 124.08, 111.93 (CH_{arom}), 117.95 (CH_{2allyl}), 117.66 (CH_{allyl}), 105.38 (C-1), 82.93 (C-2), 81.74 (C-4), 81.37 (C-3), 71.83 (C-6), 69.42 (C-5), 52.35 (O-CH_2), 26.88, 26.33 (CH_{3iso}).

(2Z)-2-benzylidene-4-(6-deoxy-1,2-O-isopropylidene-3-O-hexyl-α-D-glucofuranos-6-yl)-2H-[1,4]-benzothiazin-3(4H)-one (3b):

Yield: 91 %; **brown oil**; **^{1}H NMR** ($CDCl_3$ with 0.05 % v/v TMS, 400 MHz): δ_H 7.97 (1H, s, CH_{vinyl}), 7.06-7.69 (9H, m, H_{arom}), 6.07 (1H, d, $J_{1,2}$ = 3.7 Hz, H-1), 4.65 (1H, d, $J_{2,1}$ = 3.7 Hz, H-2), 4.57, 4.42 (2H, 2dd, $J_{6a,6b}$ = 14.5 Hz, $J_{6a,5}$ = 2.7 Hz, $J_{6b,5}$ = 5.6 Hz, H-6), 4.38-4.46 (1H, m, H-5), 4.25 (1H, dd, $J_{4,5}$ = 8.6 Hz, $J_{4,3}$ = 3.9 Hz, H-4), 4.14 (1H, d, $J_{3,4}$ = 3.9 Hz, H-3), 3.68 (3H, t, J = 7.1 Hz, O-CH_2), 1.58 (s, 3H, CH_{3iso}), 1.41 (s, 3H, CH_{3iso}), 1.28-1.39 (8H, m, CH_2), 0.92 (3H, t, J = 7.2 Hz, CH_3); **^{13}C-NMR** (62.5 MHz, $CDCl_3$): δ_C 164.83 (CO), 137.11 134.63, 120.47, 119.50, 111.86 (Cq), 135.71 (CH_{vinyl}), 130.35, 129.09, 128.45, 127.46, 126.32, 124.04, 117.68 (CH_{arom}), 105.37 (C-1), 82.83 (C-2), 82.35 (C-4), 81.59 (C-3), 71.26 (O-CH_2), 69.36 (C-5), 52.01 (C-6), 26.34-26.89 (CH_{3iso}), 31.94, 29.80, 29.69, 29.66, 29.63, 29.61, 29.43, 29.39, 26.05, 22.72 (CH_2), 14.15 (CH_3).

(2Z)-2-benzylidene-4-(6-deoxy-1,2-O-isopropylidene-3-O-dodecyl-α-D-glucofuranos-6-yl)-2H-[1,4]-benzothiazin-3(4H)-one (3c):

Yield: 91 %; **brown oil**; **^{1}H NMR** ($CDCl_3$ with 0.05 % v/v TMS, 400 MHz): δ_H 7.97 (1H, s, CH_{vinyl}), 7.06-7.69 (9H, m, H_{arom}), 6.07 (1H, d, $J_{1,2}$ = 3.7 Hz, H-1), 4.65 (1H, d, $J_{2,1}$ = 3.7 Hz, H-2), 4.57, 4.42 (2H, 2dd, $J_{6a,6b}$ = 14.5 Hz, $J_{6a,5}$ = 2.7 Hz, $J_{6b,5}$ = 5.7 Hz, H-6), 4.38-4.45 (1H, m, H-5), 4.25 (1H, dd, $J_{4,5}$ = 8.6 Hz, $J_{4,3}$ = 3.9 Hz, H-4), 4.14 (1H, d, $J_{3,4}$ = 3.9 Hz, H-3), 3.68 (3H, t, J = 7.0 Hz, O-CH_2), 1.58 (s, 3H, CH_{3iso}), 1.41 (s, 3H, CH_{3iso}), 1.28-1.39 (20H, m, CH_2), 0.92 (3H, t, J = 7.1 Hz, CH_3); **^{13}C-NMR** (62.5 MHz, $CDCl_3$): δ_C 164.83 (CO), 137.11, 134.63, 120.47, 119.50, 111.86 (Cq), 135.71 (CH_{vinyl}), 130.35, 129.09, 128.45, 127.46, 126.32, 124.04, 117.68 (CH_{arom}),105.37 (C-1), 82.83 (C-2), 82.35 (C-4), 81.59 (C-3), 71.83 (C-6), 71.26 (O-CH_2), 69.36 (C-5), 26.34-26.89 (CH_{3iso}), 31.94, 29.80, 29.69, 29.66, 29.63, 29.43, 29.39 (CH_2), 26.89 (CH_3), 26.05, 22.72 (CH_2), 14.15 (CH_3).

(2Z)-2-benzylidene-4-(((3R,4R,5S,6R)-3,5,6-trihydroxy-4-(prop-1-ynyloxy)-tetrahydro-2H-pyran-2-yl)methyl)-2H-[1,4]-benzothiazin-3(4H)-one (4a):

Yield: 80 %; **brown oil**; **^{1}H NMR** ($CDCl_3$ with 0.05 % v/v TMS, 400 MHz): δ_H 7.91 (1H, s, CH_{vinyl}), 7.03-7.66 (9H, m, H_{arom}), 5.97-6.12 (1H, m, CH_{allyl}), 5.35-540 (1H, m, H_{allyl}), 5.22-525 (1H, m, H_{allyl}), 4.61, 5.34 (1H, d, $J_{1,2}$ = 3.7 Hz, H-1), 4.58, 4.43 (2H, 2dd, $J_{6a,6b}$ = 14.6 Hz, $J_{6a,5}$ = 2.6 Hz, $J_{6b,5}$ = 5.5 Hz, H-6), 3.41-4.57 (4H, m, H-2,3,4 and 5), 4.41-4.45 (2H, m, CH_2-O); **^{13}C-NMR** (62.5 MHz, $CDCl_3$): δ_C 163.33 (CO), 135.33 (CH_{vinyl}), 136.94, 134.57, 120.26, 119.48 (Cq), 135.24, 130.30, 129.07, 128.46, 127.24, 127.13, 126.42, 124.16, 118.70 (CH_{arom}), 118.55 (CH_{2allyl}), 117.42 (CH_{allyl}), 96.89 (C-1b), 92.43 (C-1a), 83.35 (C-3b), 81.12(C-3a), 72.75 (C-6), 71.75–73.60 (C-2,4,5), 47.93(CH_2-O).

(2Z)-2-benzylidene-4-(((3R,4R,5S,6R)-4-(hexyloxy)-3,5,6-trihydroxy-tetrahydro-2H-pyran-2-yl)methyl)-2H-[1,4]-benzothiazin-3(4H)-one (4b):

Yield: 82 %; **brown oil**; **^{1}H NMR** ($CDCl_3$ with 0.05 % v/v TMS, 400 MHz): δ_H 7.93 (1H, s, CH_{vinyl}), 7.67-7.06 (9H, m, H_{arom}), 5.34, 4.67 (1H, d, $J_{1,2}$ = 3.6 Hz, H-1), 4.59, 4.44 (2H, 2dd, $J_{6a,6b}$ = 14.5 Hz, $J_{6a,5}$ = 2.7 Hz, $J_{6b,5}$ = 5.6 Hz, H-6), 4.43-3.57 (4H, m, H-2,3,4 and 5), 3.36 (2H, t, J = 7.0 Hz, CH_2-O), 1.28-1.37 (8H, m, CH_2), 0.95 (3H, t, J = 7.0 Hz, CH_3); **^{13}C-NMR** (62.5 MHz, $CDCl_3$): δ_C 163.28 (CO), 135.28 (CH_{vinyl}), 136.97, 134.61, 127.41, 124.52 (Cq), 130.29, 129.06, 128.46, 127.11, 126.25, 124.06, 120.28, 119.49, 118.65 (CH_{arom}), 96.97 (C-1b), 92.45 (C-1a), 83.93 (C-3b), 81.51 (C-3a), 73.61 (C-6), 73.60-71.98 (C-2,4,5), 47.93 (CH_2-O), 31.76, 30.32, 25.77, 22.67 (CH_2), 14.10 (CH_3).

(2Z)-2-benzylidene-4-(((3R,4R,5S,6R)-4-(dodecyloxy)-3,5,6-trihydroxy-tetrahydro-2H-pyran-2-yl)methyl)-2H-[1,4]-benzothiazin-3(4H)-one (4c):

Yield: 79 %; **brown oil**; **^{1}H NMR** ($CDCl_3$ with 0.05 % v/v TMS, 400 MHz): δ_H 7.92 (1H, s, CH_{vinyl}), 7.04-7.66 (9H, m, H_{arom}), 5.32, 4.64 (1H, d, $J_{1,2}$ = 3.7 Hz, H-1), 4.57, 4.43 (2H, 2dd, $J_{6a,6b}$ = 14.5 Hz, $J_{6a,5}$ = 2.7 Hz, $J_{6b,5}$ = 5.6 Hz, H-6), 4.57-3.43 (4H, m, H-2,3,4 and 5), 3.36 (2H, t, J = 7.0 Hz, CH_2-O), 1.28-1.36 (20H, m, CH_2), 0.94 (3H, t, J = 7.0 Hz, CH_3); **^{13}C-NMR** (62.5 MHz, $CDCl_3$): δ_C 163.25 (C=O), 135.28 (CH_{vinyl}), 136.96, 134.60, 127.40, 124.50 (Cq), 130.29, 129.04, 128.45, 127.12, 126.23, 124.05, 120.29, 119.47, 118.65 (CH_{arom}), 96.97 (C-1b), 92.45 (C-1a), 83.93 (C-3b), 81.54

(C-3a), 73.61 (C-6), 71.83-75.65 (C-2,4,5), 47.97 (CH_2-O), 31.97, 30.45, 30.38, 29.74, 29.70, 29.65, 29.61, 29.41, 26.13, 22.74 (CH_2), 14.17 (CH_3).

Pharmacological activity: antibacterial activity

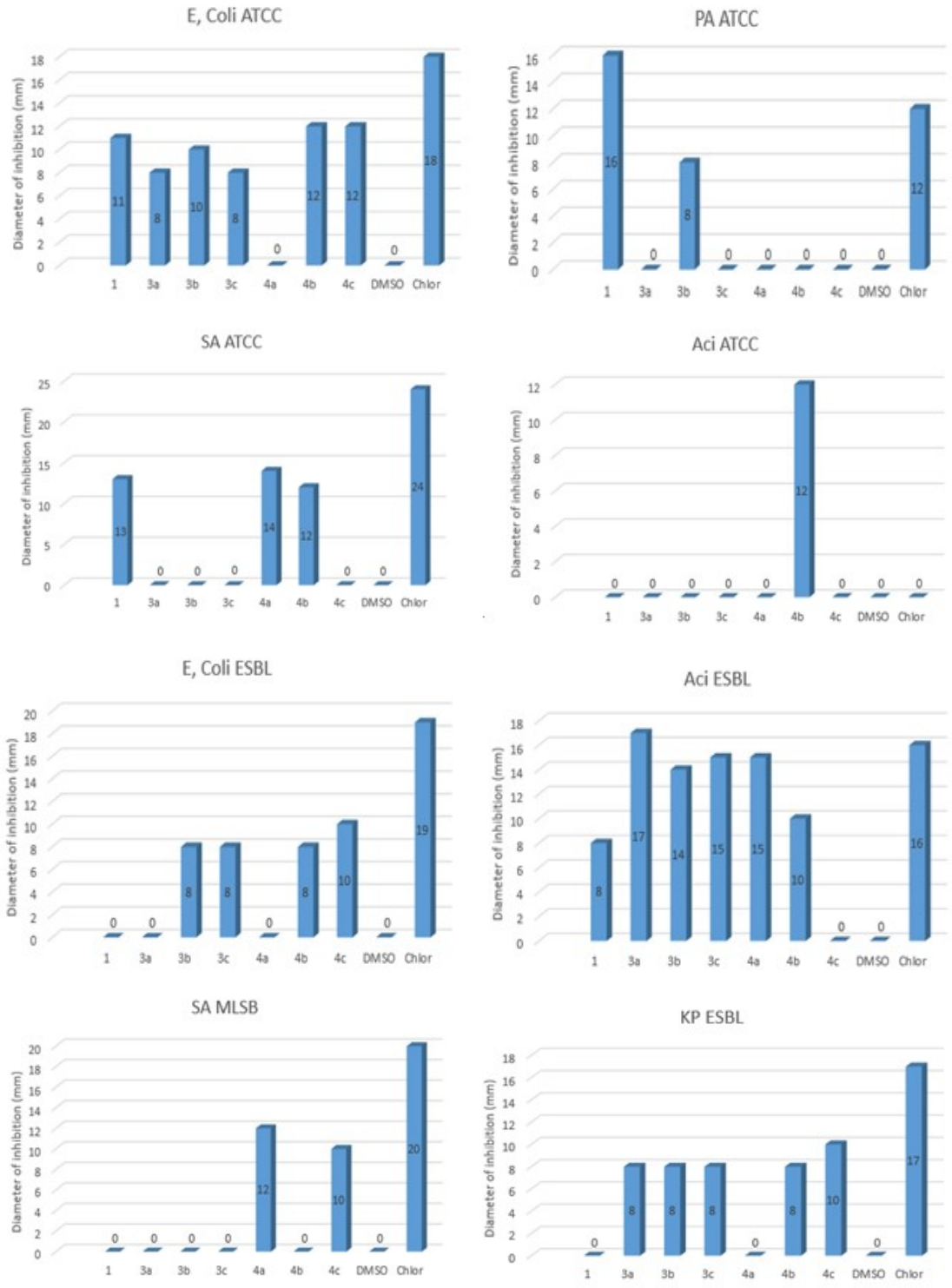
The results of the evaluation are shown in Table 1 and in Figure 2 (in the form of antibiograms).

Table 1. *Minimum Inhibitory Concentration (MIC) in µg·mL^{-1} of the compounds* ***1***, ***3a-c*** *and* ***4a-c***

Products / Strain	**1**	**3a**	**3b**	**3c**	**4a**	**4b**	**4c**	**DMSO**	**Chlor**
E. coli ATCC	250	250	250	125	-	250	125	-	4
P. aeruginosa ATCC	125	-	250	-	-	-	-	-	-
S. aureus ATCC	125	-	-	-	125	250	-	-	4
Acinetobacter *ATCC*	-	-	-	-	-	62.5	-	-	-
E. coli ESBL	-	-	125	62.5	-	125	250	-	5
Acinetobacter *ESBL*	>250	125	62.5	125	62.5	250	-	-	16
S. aureus MLSB	-	-	-	-	125	-	62.5	-	3
K. pneumonia ESBL	-	250	250	>250	-	250	125	-	2.5

This study determined the MIC of some synthesized 1,4-benzothiazine derivatives (Table 1). The results of the antibacterial activity of the products tested showed that compound **1** possess an activity with a MIC = 125 µg·mL^{-1} against *Pseudomonas Aeruginosa (ATCC*), *Staphylococcus aureus* (*ATCC*) and a MIC = 250 µg·mL^{-1} against *Escherichia coli* (*ATCC*) and *Acinetobacter baumannii* (*ESBL*). The studies achieved show the absence of growth inhibition of compound **1** in the case of four bacterial strains: *Acinetobacter baumannii* (*ATCC*), *Escherichia coli* (*ESBL*), *Staphylococcus aureus* (*MLSB*) and *Klebsiella pneumonia* (*ESBL*).

In order to increase the inhibitory activity of compound **1** we prepared three glucopyranosyl derivatives of benzothiazine **3a-c** obtained by the condensation reaction of compound **1** with anhydroglucose **2a-c**. It is worthy to note that compound **3a** shows activity with MIC of the order of 125 µg·mL^{-1} for *Acinetobacter baumannii* (*ESBL*) and 250 µg·mL^{-1} for *Escherichia coli (ATCC*) and *Klebsiella pneumoniae* (*ESBL*) and the absence of growth inhibition in the case of the bacterial strains tested *Pseudomonas Aeruginosa (ATCC*), *Staphylococcus aureus* (*ATCC*), *Acinetobacter baumannii* (*ATCC*), *Escherichia coli* (*ESBL*) and *Staphylococcus aureus* (*MLSB*).



Chlor= Chloramphenicol (30 µg·mL^{-1}), DMSO = Dimethylsulfoxyde (1 %)

Figure 2. *Results of the antibacterial activity of the synthesized compounds* ***1***, ***3a-c*** *and* ***4a-c*** *vis-a-vis bacteria tested (Escherichia coli ATCC, Pseudomonas aeruginosa ATCC, Staphylococcus aureus ATCC, Acinetobacter baumannii ATCC, Escherichia coli ESBL, Acinetobacter baumannii ESBL, Staphylococcus aureus MLSB and Klebsiella pneumonia ESBL)*

Although the compound **3b** shows no activity against the three bacterial strains tested: *Staphylococcus aureus* (*ATCC*), *Acinetobacter baumannii* (*ATCC*) and *Staphylococcus aureus* (*MLSB*), it presents a better activity with MIC 62.5 $\mu g \cdot mL^{-1}$ for *Acinetobacter baumannii* (*ESBL*), 125 $\mu g \cdot mL^{-1}$ for *Escherichia coli* (*ESBL*) and of the order of 250 $\mu g \cdot mL^{-1}$ for *Escherichia coli (ATCC*), *Pseudomonas Aeruginosa* (*ATCC*) and *Klebsiella pneumonia* (*ESBL*). However, the compound **3c** also presents an activity with MIC of 62.5 $\mu g \cdot mL^{-1}$ for *Escherichia coli* (*ESBL*), 125 $\mu g \cdot mL^{-1}$ for *Acinetobacter baumannii* (*ESBL*), *Escherichia coli* (ATCC) and >250 $\mu g \cdot mL^{-1}$ for *Klebsiella pneumoniae* (*ESBL*) and inactive in the other bacterial strains. On the other hand, the compounds **4a-b** obtained by deprotection of the isopropylidene groups **3a-c** with 9:1 CF_3CO_2H-H_2O exerted inhibitory activity toward different bacteria.
The compound **4a**, gives a better activity with MIC of about 62.5 $\mu g \cdot mL^{-1}$ for *Acinetobacter baumannii* (*ESBL*), 125 $\mu g \cdot mL^{-1}$ for *Staphylococcus aureus* (*ATCC*) and *Staphylococcus aureus* (*MLSB*). The compound **4b** gives an activity with MIC of 62.5 $\mu g \cdot mL^{-1}$ for *Acinetobacter baumannii* (*ESBL*), 125 $\mu g \cdot mL^{-1}$ for *Escherichia coli* (*ESBL*) and the order of 250 $\mu g \cdot mL^{-1}$ for *Escherichia coli (ATCC*), *Staphylococcus aureus (ATCC*), *Acinetobacter baumannii* (*ATCC*) and *Klebsiella pneumonia* (*ESBL*). Finally, the compound **4c**, has better activity with MIC of 62.5 $\mu g \cdot mL^{-1}$ for *Staphylococcus aureus* (*MLSB*), 125 $\mu g \cdot mL^{-1}$ for *Escherichia coli* (*ATCC*), *Klebsiella pneumoniae* (*ESBL*) and the order of 250 $\mu g \cdot mL^{-1}$ for *Escherichia coli* (*ESBL*).

CONCLUSION

In the development of this work, the syntheses of new N-glucosyl-1,4-benzothiazine derivatives **3a-c** and **4a-c** were carried out in satisfactory yields by grafting (Z)-2-benzylidene-2H-1,4-benzothiazin-3(4H)-one with activated D-glucose. The reactions studied are regioselective involving exclusively the nitrogen atom of the lactam function of the 1,4-benzothiazine moiety. The *N*-glucosyl 1,4-benzothiazine derivatives obtained were identified by ^1H and ^{13}C NMR spectral data. The NMR spectra of compounds **4a-c**, taken in $CDCl_3$, show that the carbohydrate moiety of the deprotected products has a pyranic structure with two α and β anomeric forms. All the six newly synthesized products were subjected to the evaluation of antibacterial activity. Some compounds tested showed significant activity.

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