https://doi.org/10.31925/farmacia.2021.3.15 **ORIGINAL ARTICLE**

SYNTHESIS, CHARACTERIZATION AND CYTOTOXICITY ASSESSMENT OF NEW 4-BENZYL-1,3-OXAZOLE DERIVATIVES INCORPORATING 4-[(4-BROMOPHENYL)SULFONYL]PHENYL FRAGMENT

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Abstract

Herein we present the design, synthesis, characterization, and cytotoxicity assessment of seven compounds derived from phenylalanine that incorporate a 4-[(4-bromophenyl)sulfonyl]phenyl fragment: four open-chain products (one *N*-acyl-α-amino acid, one *N*-acyl-α-amino acyl chloride, two *N*-acyl-α-amino ketones), and three five-membered heterocycles with two heteroatoms (O and N): one 2-aryl-4-benzyl-1,3-oxazol-5(4*H*)-one, and two 2-aryl-4-benzyl-1,3-oxazoles with *p*-tolyl, and *m*-xylyl substituent, respectively, in position 5. Structures of new derivatives were assigned by elemental analysis, NMR spectroscopy, and other spectral methods (FT-IR, MS, UV-Vis). Evaluation of the purity of the compounds was realized by reversed-phase high-performance liquid chromatography. *Daphnia magna* toxicological test was used to assess the cytotoxicity of new compounds.

Rezumat

Studiul prezintă proiectarea, sinteza, caracterizarea și evaluarea citotoxicității a șapte compuși derivați de la fenilalanină, care incorporează un fragment 4-[(4-bromofenil)sulfonil]fenil: patru produși cu catenă deschisă (*N*-acil-α-aminoacid, o clorură de *N*-acil-α-aminoacil, două *N*-acil-α-aminocetone) și trei heterocicluri pentaatomice cu doi heteroatomi (O și N): o 2-aril-4-benzil-1,3-oxazol-5(4*H*)-onă și 2-aril-4-benzil-1,3-oxazoli cu substituentul *p*-tolil și respectiv, *m*-xilil, în poziția 5. Structurile noilor derivați au fost atribuite prin analiză elementală, spectroscopie RMN și alte metode spectrale (FT-IR, SM, UV-Viz). Evaluarea purității compușilor a fost realizată prin cromatografie de lichide de înaltă performanță cu fază inversă. Testul toxicologic *Daphnia magna* a fost utilizat pentru a se evalua citotoxicitatea noilor compuși.

Keywords: *N*-acyl-α-amino acid, 4-benzyl-1,3-oxazol-5(4*H*)-one, 4-benzyl-1,3-oxazole, cytotoxic effect

Introduction

1,3-Oxazoles have increasing importance in heterocyclic chemistry and drew the researchers' attention due to their biological and medicinal applications. Though parent 1,3-oxazole does not occur naturally, numerous natural derivatives containing a 1,3-oxazole core have been isolated, especially from marine invertebrates and microorganisms, some of them exhibiting remarkable therapeutical effects [1-4]. Additionally, a series of synthetic pharmaceutical molecules bearing 1,3-oxazole scaffold were reported to have antimicrobial [5], anti-diabetic [6], analgesic, anti-inflammatory [7], anticancer activity [8], etc. 5-Hydroxy-1,3-oxazoles exist in their corresponding keto tautomer: 1,3-oxazol-5(4*H*)-ones which present cytotoxic [9, 10], antiviral [11], plant growth regulating properties [12]. Some representants of *N*-acyl-α-amino acids class show biological activities, such as anti-hypertensive [13], anticancer [14], mucolytic [15], antianaemic [16], anti-ulcer effect [17], and are specific antidotes in acute intoxications [18, 19]. *N*-Acyl-α-amino-ketones display anti-inflammatory [20], antiviral [21, 22] and antithrombotic action [23]. Besides, is known that numerous diaryl sulfones are used in therapy for their properties [24-26]. In an attempt to find new effective drugs, diaryl sulfone pharmacophore has been incorporated into a large number of derivatives with potential biological value [9, 10, 12, 27-29].

Given the scientific relevance of compounds from these classes and our previous researches [9, 10, 12, 30], in this article, we report the design, synthesis, and characterization of new *N*-acyl-α-amino acids, 1,3-oxazol-5(4*H*)-ones, *N*-acyl-α-amino acyl chlorides, *N*-acyl-α-amino ketones, and 1,3-oxazoles analogues derived from phenylalanine containing a 4-[(4-bromophenyl)sulfonyl]phenyl substituent into their structure with the aim of obtaining new bioactive products and to explore their biological action. For this purpose, the cytotoxic effect of compounds was assessed on *Daphnia magna* crustacean. Amidst the various methods of screening, this assay is fast, reproducible, cost-efficient, simple and can predict biological activity [27].

Materials and Methods

Chemistry

General: Chemicals and reagents were acquired from common commercial suppliers. Dichloromethane was anhydrized over anhydrous calcium chloride. Uncorrected melting points were measured on a Boëtius apparatus. UV-Vis spectra were registered for methanolic solutions (≈ 0.025 mM) on an Analytik Jena AG Specord 40 spectrophotometer. FT-IR spectra were recorded in KBr pellets on a Bruker Vertex 70 spectrometer, absorption peaks being described as very strong, vs; strong, s; medium, m; weak, w. NMR spectra were acquired on a Varian Gemini 300 BB instrument at 300 MHz for ^{1}H and 75 MHz for ^{13}C. Combined 2D spectra (COSY, HETCOR) were also recorded. DMSO-d_6 or $CDCl_3$ as deuterated solvents were used. Chemical shifts, δ, are in parts *per* million (ppm), relative to the reference standard tetramethylsilane (TMS) signal. Coupling constants, *J*, are expressed in hertz (Hz). In the ^{1}H-NMR spectra, signal multiplicity was assigned as singlet (s), doublet (d), doublet of doublets (dd), doublet of doublets of doublets (ddd), triplet (t), multiplet (m); broad is abbreviated b. GC-EI-MS analysis was registered on a Fisons Instruments GC 8000 with an electron impact quadrupole, MD 800 mass spectrometer detector, and a fused-silica capillary column coated with poly(5% diphenyl – 95% dimethylsiloxane) (SLB-5ms, 30 m × 0.32 mm, d_f 0.25 μm). Dichloromethane was used as solvent and the helium flow rate was 2 mL/min. RP-HPLC chromatograms were performed on a Beckman System Gold 126 liquid chromatograph, with a System Gold 166 UV-Vis detector, a non-polar chromatography column (LiChrosorb RP-18, 25 cm × 4 mm, 5 μm particle size), and a Rheodyne injection system. New compounds' purity and retention time, t_R, in minutes (min) are indicated. Elemental analysis was carried out on a Costech ECS 4010 instrument.

Synthesis and characterization of compounds

*Synthesis of 4-[(4-bromophenyl)sulfonyl]benzoyl chloride **2***

4-[(4-Bromophenyl)sulfonyl]benzoic acid **1** [31] (6.82 g, 20 mmol) was heated under reflux with an excess of thionyl dichloride (35 mL, 57.10 g, 480 mmol). Unreacted $SOCl_2$ was distilled off the reaction mixture under *vacuum*. Obtained colourless crystals were used crude in the following reaction; yield = 99% (7.12 g), m.p. = 154 - 155ºC (lit. [32] 154ºC).

FT-IR (KBr, ν cm^{-1}): 3097s; 3040m; 1779vs; 1740vs; 1591m; 1572vs; 1470m; 1333vs; 1302s; 1287s; 1162vs; 888vs; 851s; 753vs; 730vs; 647vs; 575vs.

*Synthesis of 2-{4-[(4-bromophenyl)sulfonyl]benzamido}-3-phenylpropanoic acid **3***

Phenylalanine (3.30 g, 20 mmol) was dissolved in 1 N NaOH solution (20 mL). To this solution cooled to 0 - 5ºC, a solution of raw 4-[(4-bromophenyl)sulfonyl]-benzoyl chloride **2** (7.19 g, 20 mmol) in anhydrous dichloromethane (45 mL), and a 2 N NaOH solution (10 mL), respectively were added simultaneously, dropwise, under stirring, for 30 min. Reaction mixture stirring was continued for 1 h at room temperature. The separated aqueous layer was acidified with 2 N HCl. Precipitated solid was isolated by filtration, washed with water, dried, and recrystallized as white acicular crystals; yield = 92% (8.98 g); m.p. = 179 - 180ºC (water).

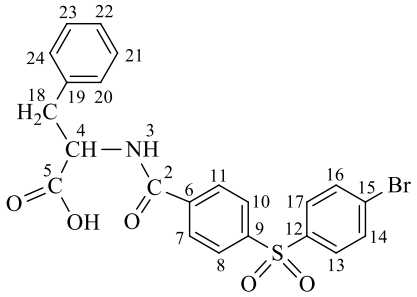


Figure 1.
Atoms numbering of compound **3** used for the assignment of NMR signals

UV-Vis (CH_3OH, λ nm) (lg ε): 202.6 (4.47); 250.9 (4.17).

FT-IR (KBr, ν cm^{-1}): 3356s; 3085m; 3058m; 3027m; 2978m; 2938m; 2869m; 2703w; 2640w; 2590w; 2530w; 1734vs; 1622vs; 1574s; 1547vs; 1492m; 1456m; 1447m; 1323vs; 1310s; 1295vs; 1161vs; 852m; 621s; 570s.

^{1}H-NMR (DMSO-d_6, δ ppm, *J* Hz): 3.04 (dd, 13.7, 10.4, 1H, H-18); 3.20 (dd, 13.7, 4.4, 1H, H-18); 4.64 (m, 1H, H-4); 7.10-7.30 (m, 5H, H-20 – H-24); 7.84 (d, 8.8, 2H, H-14, H-16); 7.91 (d, 8.8, 2H, H-13, H-17); 7.96 (d, 8.5, 2H, H-8, H-10); 8.06 (d, 8.5, 2H, H-7, H-11); 8.99 (d, 8.0, 1H, H-3).

^{13}C-NMR (DMSO-d_6, δ ppm): 36.18 (C-18); 54.21 (C-4); 126.32 (C-22); 127.54 (C-8, C-10); 128.13 (C-20, C-24); 128.35 (C-15); 128.65 (C-7, C-11); 128.94

(C-21, C-23); 129.42 (C-13, C-17); 132.88 (C-14, C-16); 137.88 (C-19); 138.63 (C-6); 139.82 (C-9); 142.74 (C-12); 164.95 (C-2); 172.68 (C-5).

RP-HPLC (methanol:water 30:70, v/v; 1 mL/min; 250 nm): purity = 99.63%; t_R = 4.57 min.

Anal. (%): Calcd. for $C_{22}H_{18}BrNO_5S$ (488.35 g/mol): C, 54.11; H, 3.72; N, 2.87; S, 6.57. Found: C, 54.06; H, 3.71; N, 2.86; S, 6.57.

*Synthesis of 4-benzyl-2-{4-[(4-bromophenyl)sulfonyl]-phenyl}-1,3-oxazol-5(4H)-one **4***

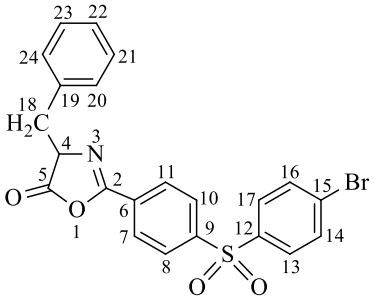


Figure 2.
Atoms numbering of compound **4** used for the assignment of NMR signals

2-{4-[(4-Bromophenyl)sulfonyl]benzamido}-3-phenylpropanoic acid **3** (5.13 g, 10.5 mmol) was suspended with stirring in anhydrous CH_2Cl_2 (50 mL) and an equimolar quantity of 4-methylmorpholine (1.15 mL) was added. Then, 1 mL (10.5 mmol) of ethyl chloroformate was slowly added to the reaction mixture. The solution was stirred for another 30 min and then poured over a mixture of ice and water (100 mL). The organic phase was isolated, washed with 5% $NaHCO_3$ solution, with water, and dried over anhydrous $MgSO_4$. After *in vacuo* concentration, solid product **4** was recrystallized from cyclohexane as white crystals; yield = 93% (4.59 g); m.p. = 161 - 162ºC.

UV-Vis (CH_3OH, λ nm) (lg ε): 202.6 (4.48); 252.9 (4.21).

FT-IR (KBr, ν cm^{-1}): 3086m; 3062m; 3031m; 2988w; 2930w; 2851w; 1822vs; 1651vs; 1600m; 1574s; 1496m; 1473m; 1455m; 1327vs; 1297vs; 1278s; 1159vs; 1044vs; 852s; 614vs; 570vs.

^{1}H-NMR ($CDCl_3$, δ ppm, J Hz): 3.19 (dd, 14.0, 6.6, 1H, H-18); 3.38 (dd, 14.0, 4.9, 1H, H-18); 4.72 (dd, 6.6, 4.9, 1H, H-4); 7.15-7.24 (m, 5H, H-20 – H-24); 7.67 (d, 8.8, 2H, H-14, H-16); 7.81 (d, 8.8, 2H, H-13, H-17); 7.99 (d, 8.5, 2H, H-8, H-10); 8.04 (d, 8.5, 2H, H-7, H-11).

^{13}C-NMR ($CDCl_3$, δ ppm): 37.24 (C-18); 66.84 (C-4); 127.44 (C-22); 128.11 (C-8, C-10); 128.58 (C-7, C-11); 128.92 (C-20, C-24); 129.25 (C-15); 129.50 (C-13, C-17); 129.62 (C-21, C-23); 130.37 (C-6); 132.93 (C-14, C-16); 134.89 (C-19); 139.82 (C-9); 144.90 (C-12); 160.34 (C-2); 176.78 (C-5).

GC-EI-MS (*m/z*, rel. abund. %): 425 (^{79}Br)/427 (^{81}Br) (13.36/15.27) $[M\text{-}CO_2]^{+\cdot}$; 309/311 (26.72/26.72) $[^{79}BrC_6H_4SO_2C_6H_4CH_2]^+$/$[^{81}BrC_6H_4SO_2C_6H_4CH_2]^+$; 203/205 (100, BP/93.13) $[^{79}BrC_6H_4SO]^+$/$[^{81}BrC_6H_4SO]^+$; 177 (10.69); 165 (10.69); 155/157 (9.16/6.87) $[^{79}BrC_6H_4]^+$/$[^{81}BrC_6H_4]^+$; 152 (3.82); 116 (45.80) $[C_6H_5C{=}C{=}NH]^+$; 96 (11.45); 90 (16.79) $[C_6H_4CH_2]^{+\cdot}$; 89 (30.53); 76 (4.20) $[C_6H_4]^{+\cdot}$; 63 (4.58) $[C_5H_3]^+$; 50 (4.58) $[C_4H_2]^{+\cdot}$; t_R = 56.97 min.

RP-HPLC (methanol:water 60:40, v/v; 1 mL/min; 250 nm): purity = 90.20%; t_R = 5.52 min.

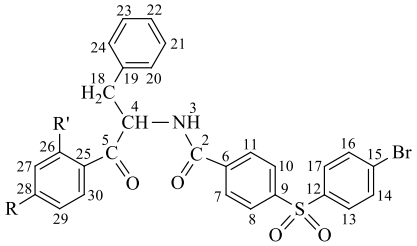
Anal. (%): Calcd. for $C_{22}H_{16}BrNO_4S$ (470.34 g/mol): C, 56.18; H, 3.43; N, 2.98; S, 6.82. Found: C, 56.25; H, 3.42; N, 2.97; S, 6.84.

*Synthesis of 2-{4-[(4-bromophenyl)sulfonyl]benzamido}-3-phenylpropanoyl chloride **5***

2-{4-[(4-Bromophenyl)sulfonyl]benzamido}-3-phenylpropanoic acid **3** (2.69 g, 5.5 mmol) was heated under reflux with an excess amount of thionyl dichloride (10 mL, 16.31 g, 137.09 mmol) until the emissions of gaseous hydrogen chloride and sulphur dioxide ceased. Unreacted $SOCl_2$ was distilled off under *vacuum* and the obtained yellow crystals was subsequently used without purification; yield = 99% (2.76 g); m.p. = 98 - 100ºC; $C_{22}H_{17}BrClNO_4S$ (506.80 g/mol).

FT-IR (KBr, ν cm^{-1}): 3374m; 3089m; 3064m; 3032m; 2969w; 2948w; 2843w; 1823s; 1794s; 1651s; 1598m; 1573s; 1517m; 1496m; 1472m; 1455m; 1328vs; 1291s; 1161vs; 891m; 850m; 613vs; 574s.

*Synthesis of N-(1-aryl-1-oxo-3-phenylpropan-2-yl)-4-[(4-bromophenyl)sulfonyl]benzamides **6a,b***



6a: R = CH_3, R' = H; **6b**: R, R' = CH_3

Figure 3.
Atoms numbering of compounds **6a,b** used for the assignment of NMR signals

A 3-fold molar excess of anhydrous aluminium trichloride (2.00 g, 15 mmol) was gradually added under magnetic stirring, at room temperature, to crude 4-benzyl-2-{4-[(4-bromophenyl)sulfonyl]phenyl}-1,3-oxazol-5(4*H*)-one **4** (2.35 g, 5 mmol) in 25 mL of anhydrous aromatic hydrocarbon (toluene or *m*-xylene). The reaction mixture was further stirred for 20 h until the release of hydrochloric acid ceased and then poured over 100 mL of ice-water mixture acidulated with 5 mL of 37% HCl. The precipitated solid was filtered off, washed with cold water, and then with a cold ethanol-water mixture (1:1, v/v). The

separated aqueous layer was extracted twice with 15 mL of CH_2Cl_2. Combined organic phases were washed with water, dried (Na_2SO_4), and concentrated by *vacuum* distillation, leaving the second fraction of the raw product. Purification by recrystallization from ethanol gives **6** as colourless crystals.

4-[(4-Bromophenyl)sulfonyl]-N-[1-oxo-3-phenyl-1-(p-tolyl)propan-2-yl]benzamide ***6a***, obtained by reaction of **4** with toluene (21.63 g, 234.75 mmol); yield = 87% (2.45 g); m.p. = 222 - 224ºC.

UV-Vis (CH_3OH, λ nm) (lg ε): 203.5 (4.49); 255.5 (4.26).

FT-IR (KBr, ν cm^{-1}): 3392s; 3084w; 3060w; 3028m; 2970w; 2930m; 2863w; 1682vs; 1650vs; 1604s; 1573s; 1513vs; 1483s; 1455m; 1329vs; 1299s; 1293s; 1162vs; 855m; 617vs; 578vs.

^{1}H-NMR (DMSO-d_6, δ ppm, J Hz): 2.35 (s, 3H, CH_3); 3.01 (dd, 13.9, 8.9, 1H, H-18); 3.19 (dd, 13.9, 4.4, 1H, H-18); 5.66 (m, 1H, H-4); 7.15 (t, 7.4, 1H, H-22); 7.29-7.37 (m, 4H, H-20, H-21, H-23, H-24); 7.32 (d, 8.2, 2H, H-27, H-29); 7.84 (d, 8.5, 2H, H-14, H-16); 7.90 (d, 8.5, 2H, H-13, H-17); 7.93 (d, 8.5, 4H, H-8, H-10, H-26, H-30); 8.04 (d, 8.5, 2H, H-7, H-11); 9.21 (d, 8.0, 1H, H-3).

^{13}C-NMR (DMSO-d_6, δ ppm): 21.25 (CH_3); 36.11 (C-18); 55.84 (C-4); 126.46 (C-22); 127.71 (C-8, C-10); 128.33 (C-15); 128.48 (C-21, C-23, C-27, C-29); 128.80 (C-7, C-11); 129.31 (C-20, C-24); 129.46 (C-13, C-17); 129.58 (C-26, C-30); 132.62 (C-25); 133.04 (C-14, C-16); 137.97 (C-19); 138.60 (C-6); 139.92 (C-9); 142.94 (C-12); 144.02 (C-28); 164.81 (C-2); 197.61 (C-5).

RP-HPLC (methanol:water 60:40, v/v; 1 mL/min; 250 nm): purity = 94.53%; t_R = 6.82 min.

Anal. (%): Calcd. for $C_{29}H_{24}BrNO_4S$ (562.47 g/mol): C, 61.92; H, 4.30; N, 2.49; S, 5.70. Found: C, 61.97; H, 4.29; N, 2.49; S, 5.72.

4-[(4-Bromophenyl)sulfonyl]-N-[1-(2,4-dimethyl-phenyl)-1-oxo-3-phenylpropan-2-yl]benzamide ***6b***, obtained by reaction of **4** with *m*-xylene (21.70 g, 204.39 mmol); yield = 90% (2.59 g); m.p. = 209 - 210ºC.

UV-Vis (CH_3OH, λ nm) (lg ε): 203.5 (4.48); 254.6 (4.19).

FT-IR (KBr, ν cm^{-1}): 3394vs; 3084m; 3063m; 3053m; 3031m; 2962m; 2927m; 2863w; 1682vs; 1654vs; 1610s; 1572vs; 1513vs; 1481vs; 1454s; 1325vs; 1294vs; 1162vs; 835m; 615vs; 578vs.

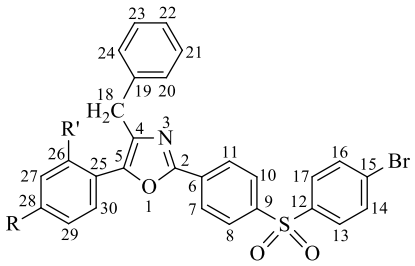
^{1}H-NMR (DMSO-d_6, δ ppm, J Hz): 2.27 (s, 3H, CH_3); 2.36 (s, 3H, CH_3); 2.99 (dd, 14.0, 10.0, 1H, H-18); 3.16 (dd, 14.0, 4.4, 1H, H-18); 5.45 (ddd, 10.0, 8.0, 4.4, 1H, H-4); 7.00-7.30 (m, 6H, H-20 – H-24, H-29); 7.09 (bs, 1H, H-27), 7.77 (d, 8.5, 1H, H-30); 7.83 (d, 8.8, 2H, H-14, H-16); 7.90 (d, 8.8, 2H, H-13, H-17); 7.90 (d, 8.5, 2H, H-8, H-10); 8.04 (d, 8.5, 2H, H-7, H-11); 9.15 (d, 8.0, 1H, H-3).

^{13}C-NMR (DMSO-d_6, δ ppm): 20.30 (CH_3); 20.84 (CH_3); 35.36 (C-18); 58.10 (C-4); 126.12 (C-29); 126.26 (C-22); 127.57 (C-8, C-10); 128.12 (C-21, C-23); 128.13 (C-7, C-11); 128.18 (C-30); 128.51 (C-15); 129.02 (C-20, C-24); 129.43 (C-13, C-17); 132.26 (C-27); 132.89 (C-14, C-16); 133.35 (C-25); 137.89 (C-26); 137.97 (C-19); 138.49 (C-6, C-28); 139.77 (C-9); 141.36 (C-12); 164.80 (C-2); 201.33 (C-5).

RP-HPLC (methanol:water 60:40, v/v; 1 mL/min; 250 nm): purity = 92.40%; t_R = 8.00 min.

Anal. (%): Calcd. for $C_{30}H_{26}BrNO_4S$ (576.50 g/mol): C, 62.50; H, 4.55; N, 2.43; S, 5.56. Found: C, 62.56; H, 4.54; N, 2.44; S, 5.54.

Synthesis of 5-aryl-4-benzyl-2-{4-[(4-bromophenyl)-sulfonyl]phenyl}-1,3-oxazoles ***7a,b***



7a: R = CH_3, R' = H; **7b**: R, R' = CH_3

Figure 4.
Atoms numbering of compounds **7a,b** used for the assignment of NMR signals

Raw *N*-(1-aryl-1-oxo-3-phenylpropan-2-yl)-4-[(4-bromophenyl)sulfonyl]benzamide **6** (10 mmol) in phosphoryl trichloride (20 mL, 217.83 mmol) was heated at reflux temperature for 4 h. The excess of $POCl_3$ was subsequently distilled off *in vacuo*. The oily residue was poured over an ice-water mixture and extracted with CH_2Cl_2 (2 × 20 mL). Combined organic phases were washed with 5% $NaHCO_3$ solution, then with water and dried over anhydrous Na_2SO_4. The solvent was removed under *vacuum* and the obtained crude solid **7** was recrystallized from ethanol as colourless crystals.

4-Benzyl-2-{4-[(4-bromophenyl)sulfonyl]phenyl}-5-(p-tolyl)-1,3-oxazole ***7a***, obtained from 5.62 g of **6a**; yield = 87% (4.74 g); m.p. = 211 - 213ºC.

UV-Vis (CH_3OH, λ nm) (lg ε): 203.5 (4.49); 252.0 (4.24); 340.1 (4.24).

FT-IR (KBr, ν cm^{-1}): 3086m; 3065m; 3027m; 2919m; 2860w; 1597s; 1574s; 1542m; 1509s; 1495m; 1473m; 1453m; 1326vs; 1293s; 1158vs; 1089vs; 844s; 618vs; 566vs.

^{1}H-NMR ($CDCl_3$, δ ppm, J Hz): 2.38 (s, 3H, CH_3); 4.18 (s, 2H, H-18); 7.20-7.30 (m, 5H, H-20 – H-24); 7.27 (d, 8.2, 2H, H-27, H-29); 7.54 (d, 8.2, 2H, H-26, H-30); 7.65 (d, 8.6, 2H, H-14, H-16); 7.82 (d, 8.6, 2H, H-13, H-17); 7.99 (d, 8.8, 2H, H-8, H-10); 8.21 (d, 8.8, 2H, H-7, H-11).

^{13}C-NMR ($CDCl_3$, δ ppm): 21.44 (CH_3); 33.19 (C-18); 125.52 (C-6); 125.96 (C-26, C-30); 126.61 (C-22); 127.10 (C-8, C-10); 128.30 (C-7, C-11); 128.51 (C-27, C-29); 128.73 (C-20, C-24); 128.81 (C-15); 129.34 (C-13, C-17); 129.79 (C-21, C-23); 132.11 (C-28); 132.83 (C-14, C-16); 136.12 (C-25); 138.87 (C-9); 140.62 (C-12); 142.00 (C-4); 142.01 (C-19); 148.20 (C-5); 157.71 (C-2).

RP-HPLC (methanol:water 70:30, v/v; 1 mL/min; 335 nm): purity = 97.70%; t_R = 4.98 min.

Anal. (%): Calcd. for $C_{29}H_{22}BrNO_3S$ (544.46 g/mol): C, 63.97; H, 4.07; N, 2.57; S, 5.89. Found: C, 63.94; H, 4.06; N, 2.58; S, 5.87.

4-Benzyl-2-{4-[(4-bromophenyl)sulfonyl]phenyl}-5-(2,4-dimethylphenyl)-1,3-oxazole ***7b***, obtained from 5.77 g of **6b**; yield = 86% (4.81 g); m.p. = 155 - 157ºC.

UV-Vis (CH_3OH, λ nm) (lg ε): 203.5 (4.49); 249.3 (4.19); 322.5 (4.19).

FT-IR (KBr, ν cm^{-1}): 3085w; 3065w; 3026m; 2923m; 2857w; 1602m; 1573s; 1558m; 1494m; 1472m; 1454m; 1327vs; 1289m; 1280m; 1158vs; 1101s; 843m; 625s; 570s.

^{1}H-NMR ($CDCl_3$, δ ppm, J Hz): 2.32 (s, 3H, CH_3); 2.38 (s, 3H, CH_3); 3.94 (s, 2H, H-18); 7.07 (bd, 7.6, 1H, H-29); 7.15 (bs, 1H, H-27); 7.20-7.30 (m, 5H, H-20 – H-24); 7.21 (d, 7.6, 1H, H-30); 7.65 (d, 8.7, 2H, H-14, H-16); 7.82 (d, 8.7, 2H, H-13, H-17); 7.99 (d, 8.8, 2H, H-8, H-10); 8.18 (d, 8.8, 2H, H-7, H-11).

^{13}C-NMR ($CDCl_3$, δ ppm): 20.46 (CH_3); 21.41 (CH_3); 32.46 (C-18); 124.48 (C-6); 126.48 (C-22); 126.78 (C-29); 127.03 (C-8, C-10); 128.32 (C-7, C-11); 128.59 (C-20, C-24); 128.66 (C-21, C-23); 128.80 (C-15); 129.34 (C-13, C-17); 130.17 (C-30); 131.80 (C-27); 132.83 (C-14, C-16); 137.79 (C-26); 137.82 (C-25); 138.96 (C-28); 139.98 (C-9), 140.65 (C-12); 142.01 (C-4, C-19); 148.60 (C-5); 158.43 (C-2).

RP-HPLC (methanol:water 70:30, v/v; 1 mL/min; 335 nm): purity = 99.90%; t_R = 5.03 min.

Anal. (%): Calcd. for $C_{30}H_{24}BrNO_3S$ (558.49 g/mol): C, 64.52; H, 4.33; N, 2.51; S, 5.74. Found: C, 64.57; H, 4.32; N, 2.52; S, 5.76.

Cytotoxicity assessment

Daphnia magna bioassay was used for the cytotoxicity evaluation. The procedure was described in our previous works [27, 33]. The daphnids were exposed to six concentrations ranging from 3.6 to 130 µg/mL, for a period of 48 h. The test was carried out in duplicate, and phenylalanine, compound **1**, and 1% DMSO were used as controls. The bioassay was performed in a controlled environment at 25 ± 1°C, 75% RH (climatic chamber Sanyo MLR-351 H, USA).

At 24 and 48 h, the lethality was recorded, and the LC50 values were calculated by interpolation on the lethality curves. GUSAR software was applied for the prediction of $LC50_{48\ h}$.

Results and Discussion

Chemistry

Synthetic transformations into title derivatives started from the following raw materials: 4-[(4-bromophenyl)-sulfonyl]benzoic acid **1** and corresponding acyl chloride **2** which are literature-known compounds [31, 32]. Carboxylic acid **1** was obtained from commercially available 4-methylbenzene-1-sulfonyl chloride and bromobenzene by two successive reactions [31]. Then, compound **1** was converted into 4-[(4-bromophenyl)-sulfonyl]benzoyl chloride **2**, the method being adapted based on previously described procedures [12, 30]. Further, phenylalanine was acylated with raw material **2** to new 2-{4-[(4-bromophenyl)sulfonyl]benzamido}-3-phenylpropanoic acid **3**. Subsequently, *N*-acyl-α-amino acid **3** was cyclodehydrated, under the action of ethyl carbonochloridate in presence of 4-methyl-morpholine to 4-benzyl-2-{4-[(4-bromophenyl)-sulfonyl]phenyl}-1,3-oxazol-5(4*H*)-one **4**. The 2-{4-[(4-bromophenyl)sulfonyl]benzamido}-3-phenyl-propanoyl chloride **5** was prepared by reacting *N*-acylated phenylalanine **3** with thionyl dichloride. By $AlCl_3$-catalyzed acylation of arenes (toluene, *m*-xylene) with 2-aryl-4-benzyl-1,3-oxazol-5(4*H*)-one **4**, the 4-aryl-2-aza-3-benzyl-1-{[(4-bromophenyl)-sulfonyl]phenyl}-1,4-butanediones **6a,b** were obtained. Further, the intramolecular condensation and dehydration of *N*-(1-aryl-1-oxo-3-phenylpropan-2-yl)-4-[(4-bromo-phenyl)sulfonyl]benzamides **6a,b** under the action of phosphoryl trichloride, known as Robinson-Gabriel reaction, yielded 5-aryl-4-benzyl-1,3-oxazoles containing 4-[(4-bromophenyl)sulfonyl]phenyl moiety in 2-position **7a,b**.

Depicted chemical structures of new derivatives were established based on NMR spectroscopy (^{1}H, ^{13}C, COSY, HETCOR), UV-Vis, IR, MS data and elemental analysis.

The *UV-Vis spectra* of the synthesized compounds presented E band at λ_{max} = 202.6 or 203.5 nm, and B band in 249.3 - 255.5 nm range. In addition, 1,3-oxazoles **7** spectra showed a third absorption maximum at longer wavelengths (322.5 or 340.1 nm), due to the appearance of the heterocyclic chromophore, which determines the extent of π electrons conjugation.

The *FT-IR spectra* of the new compounds presented all the expected characteristic absorption bands. In the case of acyclic precursors **3** and **6a,b**, main peaks were due to N-H valence vibration, ν(N-H), in 3356 - 3394 cm^{-1} range, ketonic carbonyl valence vibration, ν(O=C-C), at 1682 or 1734 cm^{-1}, and stretching vibration of amidic carbonyl, ν(O=C-N), in 1622 - 1654 cm^{-1} interval. Representative peaks for the hydrogen-bonded dimer of *N*-acyl phenylalanine **3** were also recorded: a broad absorption band due to O-H stretch, ν(O-H), between 2500 and 3300 cm^{-1}, and four broad bands in 2530 - 2703 cm^{-1} interval. The IR spectrum of acyl chloride **5** showed a fundamental band due

to ν(O=C-Cl) at 1823 cm^{-1}, Fermi resonance peak at 1794 cm^{-1}, and band due to C-Cl valence vibration, ν(Cl-C=O), at 891 cm^{-1}. The IR spectral data of five-membered-ring systems **4** and **7a,b** demonstrated that cyclocondensation of open-chain intermediates **3** and **6a,b**, respectively occurred. In compound **4** IR spectrum, carbonyl valence vibration, ν(C=O), was shifted at higher wavenumber (1822 cm^{-1}); besides, ν(N-H), ν(O-H), and ν(O=C-N) peaks were not recorded. Also, absorption bands were not observed in the N-H and C=O regions of 1,3-oxazoles **7** spectra. The IR spectra of pentaatomic heterocycles **4** and **7a,b** showed C=N valence vibration, ν(C=N), at 1651 cm^{-1} (**4**), and 1597 or 1602 cm^{-1} (**7a,b**). Furthermore, C-O-C symmetrical stretching vibration, ν_{sym}(C-O-C), appeared at 1044 cm^{-1} in saturated azlactone **4** spectrum, and at 1089 cm^{-1} or 1101 cm^{-1} in 1,3-oxazoles **7a,b** spectra, and C-O-C asymmetrical stretching vibration, ν_{as}(C-O-C), was registered at 1278 cm^{-1} (**4**), and 1280 cm^{-1} (**7b**).

The *^{1}H-NMR spectra* of the new compounds confirmed proposed chemical structures. Moreover, ^{1}H-^{1}H COSY spectra facilitated assigning the signals. In ^{1}H-NMR spectra of compounds **3** and **6a,b**, H-3 signal was recorded as a doublet at δ values of 8.99 ppm (**3**) and 9.15 or 9.21 ppm (**6a,b**), due to the coupling with H-4 (^3J = 8.0 Hz). As proof that cyclization of acyclic intermediates **3** and **6a,b** have taken place, in ^{1}H-NMR spectra of heterocycles **4** and **7a,b**, the signal assigned to NH proton (observed in the structure of the two raw materials) was absent. In ^{1}H-NMR spectra of **3** and **6a,b**, the H-4 signal appeared as a doublet of doublets of doublets or multiplet at δ = 4.64 ppm (**3**) and 5.45 or 5.66 ppm (**6a,b**), due to H-4 coupling with the two nonequivalent H-18 protons and one NH proton. For azlactone **4**, the H-4 signal was registered at δ = 4.72 ppm as a doublet of doublets due to coupling with the nonequivalent H-18 protons. The signal of H-4 (that appeared in ^{1}H-NMR spectra of **6a,b**), was not observed in spectra of heterocyclic analogues **7a,b** as proof that cyclocondensation occurred. The ^{1}H-NMR spectrum of **4** displayed two signals due to nonequivalent H-18 protons as a doublet of doublets, which showed discernible downfield shifts of 0.15 and 0.18 ppm, compared to corresponding protons signals in the spectrum of intermediate **3**. The ^{1}H-NMR spectra of 1,3-oxazoles **7a,b** highlighted a downfield shift of signal assigned to two magnetically equivalent H-18 protons as a singlet at δ = 3.94 or 4.18 ppm relative to the two signals registered for *N*-acyl-α-amino ketones **6a,b** as a doublet of doublets (due to germinal coupling between methylene non-equivalent protons and vicinal coupling between H-18 protons and H-4 proton) at 3.01 and 3.19 ppm (**6a**) and 2.99 and 3.16 ppm (**6b**).

The *^{13}C-NMR spectra* also proved the syntheses of the compounds took place. Besides, ^{1}H-^{13}C COSY spectra allowed the assignments of ^{13}C peaks. The C-4 signal, which was recorded at δ = 54.21 ppm in the ^{13}C-NMR spectrum of *N*-acylated phenylalanine **3**, was shifted downfield with 12.63 ppm after cyclization to **4**.

Moreover, in the case of 1,3-oxazol-5(4*H*)-one **4**, the C-2 resonated at 160.34 ppm (being shifted up-field with 4.61 ppm than C-2 of intermediate **3**), and the C-5 at 176.78 ppm (being deshielded with 4.10 ppm than C-5 of precursor **3**). The signal assigned to C-2 of 2,4,5-trisubstituted 1,3-oxazoles **7a,b** was registered at δ = 157.71 or 158.43 ppm, while the peak attributed to C-2 from -CONH- group of *N*-acyl-α-amino ketones **6a,b** (from δ = 164.80 or 164.81 ppm) was absent in aromatic heterocycles **7a,b** spectra. C-5 of 1,3-oxazoles **7a,b** resonated at δ = 148.20 or 148.60 ppm, whereas the corresponding atom of **6a,b** at δ = 197.61 or 201.33 ppm, revealing a high-field shift for this carbon from cyclization products **7a,b** structure, as a confirmation that the reaction took place.

The *mass spectrum* of 1,3-oxazol-5(4*H*)-one **4** obtained by GC-EI-MS analysis played an important role in elucidating the structure of this compound. The two very unstable molecular ions of **4** that correspond to bromine isotopes (^{79}Br/^{81}Br) did not show peaks in the mass spectrum. They began to fragment at the level of the heterocyclic nucleus by removing a molecule of carbon dioxide with the obtaining of two cation-radicals corresponding to isotopes of bromine with *m/z* = 425/427 (with relative abundances: 13.36% and 15.27%, respectively).

The base peak (PB) $[^{79}BrC_6H_4SO]^+$ with *m/z* = 203 and corresponding cation $[^{81}BrC_6H_4SO]^+$ with *m/z* = 205 (with relative abundance = 93.13%) were formed in accordance with ^{79}Br/^{81}Br isotopic ratio of about 1:1. Other main fragments of **4** corresponding to ^{79}Br and ^{81}Br isotopes are indicated in section Materials and Methods.

Cytotoxicity assessment

Daphnia magna bioassay results are presented in Table I. At 24 h, at the highest concentrations, with the exception of **6a** which exhibited at 24 h an L% of 45%, all tested compounds did not exceed 20%. At 48 h, the highest toxicity was induced by compound **7a**. However, this toxicity wasn't dependent on the concentrations. For this compound, at all concentrations was observed an L% between 30 and 60%, and thus, lower goodness of fit. For all other compounds, the correlation between the concentrations and the L% was satisfactory (for compound **4** and phenylalanine between 0.6 and 0.69 and for other compounds over 0.7) (Figure 5). The cytotoxicity on *D. magna* decrease in the following order: **7a**, **6a**, **7b**, **6b**, **3**, **1**, and **4**. Phenylalanine induced a maximum of 30% lethality at 48 h. The 95% CI values for compounds **6a**, **7b** and **6b** indicate similar toxicities of these three compounds. The values obtained experimentally were significantly higher than those predicted using GUSAR software.

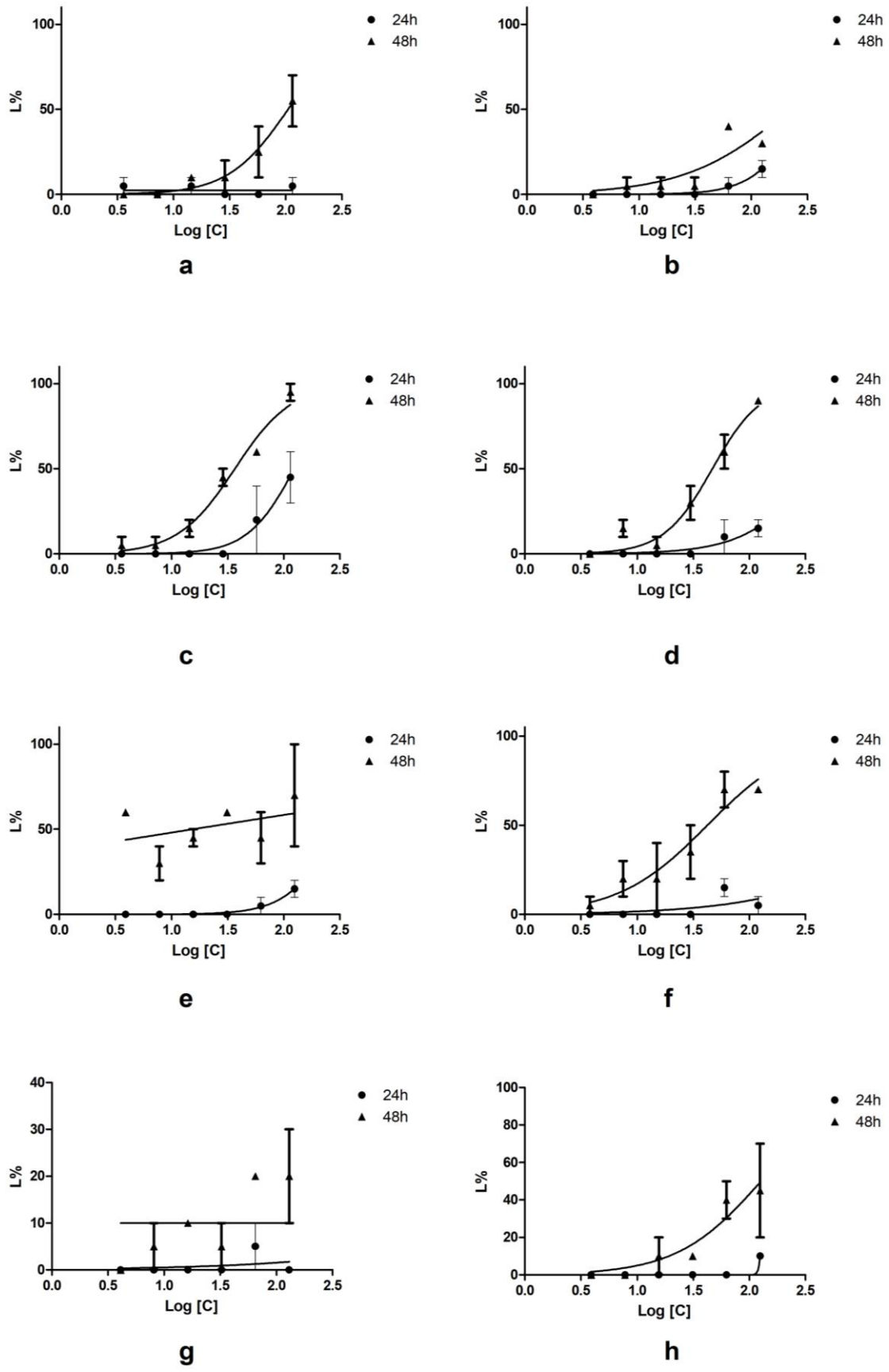


Figure 5.
Lethality curves on *Daphnia magna* for selected compounds;
a – **3**, b – **4**, c – **6a**, d – **6b**, e – **7a**, f – **7b**, g – phenylalanine, h – **1**

Table I

Results of *Daphnia magna* bioassay

Compounds	Predicted $LC50_{48\ h}$ (µg/mL)	Determined $LC50_{48\ h}$ (µg/mL)	95% CI of $LC50_{48\ h}$ (µg/mL)
3	0.44	105.00	72.59 - 151.80
4	0.28	222.2	83.99 - 587.80
6a	0.11	36.67	30.37 - 44.27
6b	0.04	46.85	37.80 - 58.07
7a	0.03	~ 15.49	NC
7b	0.02	42.34	26.08 - 68.74
phenylalanine	170.79	NC	NC
1	11.80	127.3	63.08 - 256.9

LC50 – 50% lethal concentration; 95% CI – 95% confidence interval; NC – not calculated due to obtained results

Conclusions

Seven new compounds, derived from phenylalanine, which incorporate in the structure the 4-[(4-bromophenyl)sulfonyl]phenyl fragment, were designed, synthesized, and physicochemically characterized. 1,3-Oxazol-5(4*H*)-one **4** was produced by Steiger acylation of phenylalanine with acyl chloride **2**, followed by intramolecular cyclodehydration of acyclic precursor **3**. *N*-Acyl phenylalanyl chloride **5** was obtained by reaction of **3** with $SOCl_2$. *N*-Acyl-α-amino ketones **6a,b** were generated by the reaction of 1,3-oxazol-5(4*H*)-one **4** with aromatic hydrocarbons in presence of aluminium trichloride. The 2,4,5-trisubstituted 1,3-oxazoles **7a,b** were synthesized by cyclization of open-chain intermediates **6a,b** under the action of phosphoryl trichloride. Compounds structure was elucidated through spectral methods and elemental analysis.

Daphnia magna bioassay revealed that the newly synthesized compounds **6a**, **6b**, **7a**, and **7b** exhibited high cytotoxicity being promising candidates for future biological investigations.

Conflict of interest

The authors declare no conflict of interest.

References

1. Mhlongo JT, Brasil E, de la Torre BG, Albericio F, Naturally Occurring Oxazole-Containing Peptides. *Mar Drugs*, 2020; 18(4): 203: 1-25.
2. Ke S, Zhang Z, Shi L, Liu M, Fang W, Zhang Y, Wu Z, Wan Z, Long T, Wang K, An efficient synthesis and bioactivity evaluation of oxazole-containing natural hinduchelins A-D and their derivatives. *Org Biomol Chem.*, 2019; 17(14): 3635-3639.
3. Bai R, Cruz-Monserrate Z, Fenical W, Pettit GR, Hamel E, Interaction of diazonamide A with tubulin. *Arch Biochem Bioph.*, 2020; 680: 108217: 1-24.
4. Jin Z, Muscarine, imidazole, oxazole and thiazole alkaloids. *Nat Prod Rep.*, 2016; 33(11): 1268-1317.
5. Zhang HZ, Zhao ZL, Zhou CH, Recent advance in oxazole-based medicinal chemistry. *Eur J Med Chem.*, 2018; 144: 444-492.
6. Han CL, Qu CZ, Cardiovascular Risk and Safety Evaluation of a Dual Peroxisome Proliferator-Activated Receptor-Alpha/Gamma Agonist, Aleglitazar, in Patients With Type 2 Diabetes: A Meta-analysis. *J Cardiovasc Pharmacol.*, 2020; 75(4): 351-357.
7. Furihata K, Nagasawa K, Hagino A, Kumagai Y, A drug-drug interaction study of a novel, selective urate reabsorption inhibitor dotinurad and the non-steroidal anti-inflammatory drug oxaprozin in healthy adult males. *Clin Exp Nephrol.*, 2020; 24(Suppl 1): 36-43.
8. Chiacchio MA, Lanza G, Chiacchio U, Giofrè SV, Romeo R, Iannazzo D, Legnani L, Oxazole-Based Compounds As Anticancer Agents. *Curr Med Chem.*, 2019; 26(41): 7337-7371.
9. Apostol TV, Socea LI, Drăghici C, Olaru OT, Șaramet G, Enache-Preoteasa C, Bărbuceanu ȘF, Design, synthesis, characterization, and cytotoxicity evaluation of new 4-benzyl-1,3-oxazole derivatives bearing 4-(4-chlorophenylsulfonyl)phenyl moiety. *Farmacia*, 2021; 69(2): 314-324.
10. Apostol TV, Barbuceanu SF, Olaru OT, Draghici C, Saramet G, Socea B, Enache C, Socea LI, Synthesis, Characterization and Cytotoxicity Evaluation of New Compounds from Oxazol-5(4H)-ones and Oxazoles Class Containing 4-(4-Bromophenylsulfonyl)phenyl Moiety. *Rev Chim.*, 2019; 70(4): 1099-1107.
11. Pinto IL, West A, Debouck CM, Dilella AG, Gorniak JG, O'Donnell KC, O'Shannessy DJ, Patel A, Jarvest RL, Novel, selective mechanism-based inhibitors of the herpes proteases. *Bioorg Med Chem Lett.*, 1996; 6(20): 2467-2472.
12. Apostol TV, Draghici C, Dinu M, Barbuceanu SF, Socea LI, Saramet I, Synthesis, Characterization and Biological Evaluation of New 5-Aryl-4-methyl-2-[para-(phenylsulfonyl)phenyl]oxazoles. *Rev Chim.*, 2011; 62(2): 142-148.
13. Ortega CA, Favier LS, Cianchino VA, Cifuente DA, New Orodispersible Mini Tablets of Enalapril Maleate by Direct Compression for Pediatric Patients. *Curr Drug Deliv.*, 2020; 17(6): 505-510.
14. Wei CW, Yu YL, Chen YH, Hung YT, Yiang GT, Anticancer effects of methotrexate in combination with α tocopherol and α tocopherol succinate on triple negative breast cancer. *Oncol Rep.*, 2019; 41(3): 2060-2066.
15. Ehre C, Rushton ZL, Wang B, Hothem LN, Morrison CB, Fontana NC, Markovetz MR, Delion MF, Kato T, Villalon D, Thelin WR, Esther CR Jr, Hill DB, Grubb BR, Livraghi-Butrico A, Donaldson SH, Boucher RC, An Improved Inhaled Mucolytic to Treat Airway Muco-obstructive Diseases. *Am J Respir Crit Care Med.*, 2019; 199(2): 171-180.

16. Berry RJ, Lack of historical evidence to support folic acid exacerbation of the neuropathy caused by vitamin B12 deficiency. *Am J Clin Nutr*., 2019; 110(3): 554-561.
17. Sharma P, Singh S, Siddiqui TI, Singh VS, Kundu B, Prathipati P, Saxena AK, Dikshit DK, Rastogi L, Dixit C, Gupta MB, Patnaik GK, Dikshit M, α-Amino acid derivatives as proton pump inhibitors and potent anti-ulcer agents. *Eur J Med Chem.*, 2007; 42(3): 386-393.
18. Schaper A, Ebbecke M, Intox, detox, antidotes – Evidence based diagnosis and treatment of acute intoxications. *Eur J Intern Med.*, 2017; 45: 66-70.
19. Pohanka M, Antidotes against methanol poisoning: A review. *Mini Rev Med Chem.*, 2019; 19(14): 1126-1133.
20. Semple G, Ashworth DM, Batt AR, Baxter AJ, Benzies DWM, Elliot LH, Evans DM, Franklin RJ, Hudson P, Jenkins PD, Pitt GR, Rooker DP, Yamamoto S, Isomura Y, Peptidomimetic aminomethylene ketone inhibitors of interleukin-1β-converting enzyme (ICE). *Bioorg Med Chem Lett.*, 1998; 8(8): 959-964.
21. Zhang L, Liu J, Cao R, Xu M, Wu Y, Shang W, Wang X, Zhang H, Jiang X, Sun Y, Hu H, Li Y, Zou G, Zhang M, Zhao L, Li W, Guo X, Zhuang X, Yang XL, Shi ZL, Deng F, Hu Z, Xiao G, Wang M, Zhong W, Comparative Antiviral Efficacy of Viral Protease Inhibitors against the Novel SARS-CoV-2 *In Vitro*. *Virol Sin.*, 2020; 35(6): 776-784.
22. Fear G, Komarnytsky S, Raskin I, Protease inhibitors and their peptidomimetic derivatives as potential drugs. *Pharmacol Ther.*, 2007; 113(2): 354-368.
23. Deng H, Bannister TD, Jin L, Babine RE, Quinn J, Nagafuji P, Celatka CA, Lin J, Lazarova TI, Rynkiewicz MJ, Bibbins F, Pandey P, Gorga J, Meyers HV, Abdel-Meguid SS, Strickler JE, Synthesis, SAR exploration, and X-ray crystal structures of factor XIa inhibitors containing an α-ketothiazole arginine. *Bioorg Med Chem Lett.*, 2006; 16(11): 3049-3054.
24. Evernden C, Dowhan M, Dabas R, Chaudhry A, Kalra A, Dharmani-Khan P, Gregson D, Johnson A, Jupp J, Jimenez-Zepeda V, Jamani K, Duggan P, Tay J, Khan F, Daly A, Storek J, High incidence of *Pneumocystis jirovecii* pneumonia in allogeneic hematopoietic cell transplant recipients in the modern era. *Cytotherapy*, 2020; 22(1): 27-34.
25. Bera S, Mondal D, Insights of synthetic analogues of anti-leprosy agents. *Bioorg Med Chem.*, 2019; 27(13): 2689-2717.
26. Al-Said MS, Ghorab MM, Nissan YM, Dapson in heterocyclic chemistry, part VIII: synthesis, molecular docking and anticancer activity of some novel sulfonylbiscompounds carrying biologically active 1,3-dihydropyridine, chromene and chromenopyridine moieties. *Chem Cent J.*, 2012; 6(1): 64: 1-14.
27. Roșca EV, Apostol TV, Chifiriuc MC, Grădișteanu Pîrcălăbioru G, Drăghici C, Socea LI, Olaru OT, Nițulescu GM, Pahonțu EM, Hrubaru M, Bărbuceanu ȘF, *In silico* and experimental studies for the development of novel oxazol-5(4H)-ones with pharmacological potential. *Farmacia*, 2020; 68(3): 453-462.
28. Mady MF, Saleh TS, El-Kateb AA, Abd El-Rahman NM, Abd El-Moez SI, Microwave-assisted synthesis of novel pyrazole and pyrazolo[3,4-d]pyridazine derivatives incorporating diaryl sulfone moiety as potential antimicrobial agents. *Res Chem Intermed.*, 2016; 42(2): 753-769.
29. Ahmad I, Shagufta, Sulfones: an important class of organic compounds with diverse biological activities. *Int J Pharm Pharm Sci.*, 2015; 7(3): 19-27.
30. Apostol TV, Saramet I, Draghici C, Barbuceanu SF, Socea LI, Almajan GL, Synthesis and Characterization of New 5-Aryl-2-[para-(4-chlorophenylsulfonyl)phenyl]-4-methyloxazoles. *Rev Chim.*, 2011; 62(5): 486-492.
31. Mavrodin A, Oteleanu D, Stoenescu MV, Zotta V, Studies on the sulfone group. IV. New sulfone-hydrazide derivatives. *Pharm Zentralhalle Dtschl.*, 1956; 95(9): 353-361.
32. Schiketanz I, Draghici C, Saramet I, Balaban AT, Aminoketone, oxazole and thiazole synthesis. Part 15.1 2-[4-(4-Halobenzenesulphonyl)-phenyl]-5-aryloxazoles. *Arkivoc*, 2002; (ii): 64-72.
33. Costea T, Hudiță A, Olaru OT, Gălățeanu B, Gîrd CE, Mocanu MM, Chemical composition, antioxidant activity and cytotoxic effects of Romanian *Craterellus cornucopioides* (L.) Pers. mushroom. *Farmacia*, 2020; 68(2): 340-347.