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Original article

Microwave induced synthesis of novel 8,9-dihydro-7*H*-pyrimido[4,5-*b*][1,4]diazepines as potential antitumor agents

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Abstract

A series of new racemic 4-amino-6-aryl-8-(1,3-benzodioxol-5-yl)-8,9-dihydro-7*H*-pyrimido[4,5-*b*][1,4]diazepines **4a–f** and 4-amino-8-aryl-6-(1,3-benzodioxol-5-yl)-8,9-dihydro-7*H*-pyrimido[4,5-*b*][1,4]diazepines **5a–f** were obtained regioselectively from the reaction of 4,5,6-triaminopyrimidine **1** with 1 equiv of methylenedioxychalcones **2a–f** and **3a–f**, under microwave irradiation. Detailed NMR measurements confirm the high regioselectivity of this reaction. These compounds have been evaluated in the US National Cancer Institute (NCI) for their ability to inhibit approximately 60 different human tumor cell lines, where **4e**, **5a** and **5b** presented remarkable activity against 47, 11 and 37 cancer cell lines, respectively, with the most important GI_{50} values ranging from 0.068 to 0.35 μM, in vitro assay.

Keywords: Regioselective cyclocondensation; 4,5,6-Triaminopyrimidine; Methylenedioxychalcones; Pyrimido[4,5-*b*][1,4]diazepine; Antitumoral; Microwave

1. Introduction

The reaction of aromatic or heteroaromatic 1,2-diamines with α,β-unsaturated ketones (chalcones) is a useful method for the preparation of condensed 1,4-diazepine systems [1–8], because of its versatility and high regioselectivity.

Although benzodiazepines have been studied largely over decades because of their important pharmacological activity [9,10], in recent years, the fusion of a heterocyclic system to the seven-member diazepine ring has become, as shown in literature [11,12], a promising step for the synthesis of new compounds with a higher and more specific activity over different biological targets as the central nervous system (CNS) [13] and also as antitumorals in several cell lines [14].

On the other hand, microwave enhanced synthesis has attracted substantial attention in recent years, enabling many organic reactions to proceed much faster and with higher yields than when conventional heating is employed. Microwave irradiation has become a powerful synthetic tool for rapid synthesis of a variety of organic compounds [15–17]. Microwave-assisted reactions have attracted much interest because of the simplicity in operation, milder reaction conditions, increasing reaction rates and formation of cleaner products. Solvent-free microwave-assisted reactions have gained more popularity as they provide an opportunity to work with open vessels [18–23].

In this way, focusing on the preparation of such kind of bioactive nitrogen containing heterocycles, we report the regioselective synthesis and cytotoxic activities of 4-amino-6-aryl-8-(1,3-benzodioxol-5-yl)-8,9-dihydro-7*H*-pyrimido[4,5-*b*][1,4]diazepines **4a–f** and 4-amino-8-aryl-6-(1,3-benzodioxol-5-yl)-8,9-dihydro-7*H*-pyrimido[4,5-*b*][1,4]diazepines **5a–f**. These

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compounds were obtained as a racemic mixture using microwave-assisted reaction techniques.

The compounds were evaluated for their antitumoral activity against approximately 60 different human tumor cell lines, representing leukemia, melanoma and cancers of the lung, colon, brain, ovary, breast, prostate, and kidney. The antitumoral evaluation was performed by The Developmental Therapeutics Program (DTP) of the National Cancer Institute of the United States (U.S.), and the testing results are expressed according to the next three parameters: GI_{50} which is the molar concentration of the compounds required to inhibit the growth of that cell line by 50% (relative to untreated cells), TGI the molar concentration that causes total growth inhibition, and LC_{50} which is a parameter of cytotoxicity and reflects the molar concentration needed to kill 50% of the cells [24].

2. Results and discussion

2.1. Chemistry

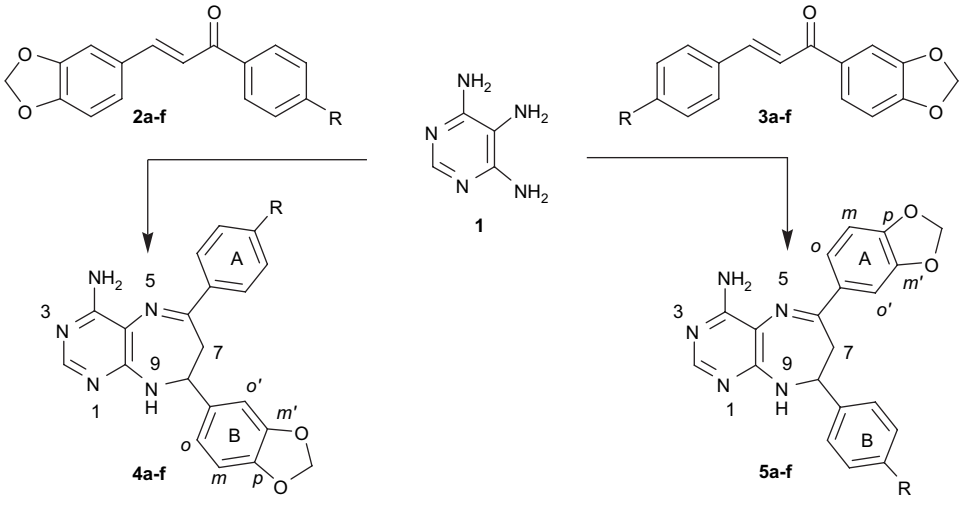
The microwave irradiation of an equimolar mixture of 4,5,6-triaminopyrimidine **1** and chalcones **2a–f** and **3a–f** in the presence of catalytic amounts of DMF (1 mL) affords the desired products **4a–f** and **5a–f** selectively and in good yields (see Scheme 1).

Due to the presence of non-equivalent amino groups at the *ortho*-position, regioisomeric cyclization products could be expected. However, in both series the formation of a single product was observed, possibly because of the electronic effect of the pyrimidine ring which enhances the nucleophilicity of the amino group on C-5 [25,26]. Therefore, condensation between such amino group and the carbonyl group of **2a–f** and **3a–f** can be followed by a Michael addition of the less nucleophilic amino group (the two equivalent amino groups on C-4 and C-6) to the C=C double bond.

The IR spectra show typical bands between 3083 and 3455 cm^{-1} (NH and NH_2 groups) and 1557–1657 cm^{-1} (C=N and C=C groups). Additionally, derivatives **4a** and **5a** show typical absorptions at 1342–1572 cm^{-1} due to NO_2 stretching vibrations.

The ^1H NMR data for all products are summarized in Section 4. The three protons of the 1,4-diazepine ring on methylene C-7 and the stereogenic center C-8 form an AMX spin system. The geminal diastereotopic protons on methylene group H_2C-7 are observed at $\delta = 2.78–3.05$ ppm (H_A, a doublet of doublets) and at $\delta = 3.54–3.99$ ppm (H_M, a doublet of doublets), the coupling constant between them is in the range $^2J_{AM} = 13.3–14.8$ Hz. The latter geminal system shows two vicinal couplings with C-8, characterized by $^3J_{AX} = 1.8–2.1$ Hz and $^3J_{MX} = 5.5–6.6$ Hz. The coupling of the proton on N-9 gives rise to a doublet ($\delta = 7.47–9.21$ ppm for **4** and 7.03–8.80 ppm for **5**, $^3J = 4.0–5.7$ Hz), indicating the vicinal position to the proton on C-8 (multiplet at $\delta = 5.03–5.43$ ppm, for both series). Protons in the NH_2 group at C-4 appear as a singlet at $\delta = 6.23–7.96$ ppm for all compounds, except for **4a** in which the signal is not present in the spectra. The characteristic signal of the proton on C-2 of the pyrimidodiazepine ring appears as a singlet at $\delta = 7.15–8.27$ ppm for all derivatives. Another characteristic singlet appearing at $\delta = 5.85–6.03$ ppm corresponds to protons on OCH_2O of 1,3-benzodioxol substituent for products **4a–f** and **5a–f**.

Analysis of ^{13}C, DEPT-135 and two dimensional heteronuclear NMR spectra provided the final structural elucidation of compounds **4a–f** and **5a–f**. So, the signal for C-7 is in the



Comp.	4a	4b	4c	4d	4e	4f	5a	5b	5c	5d	5e	5f
R	NO_2	Cl	Br	H	CH_3	OCH_3	NO_2	Cl	Br	H	CH_3	OCH_3
Yield (%)	55	62	68	35	51	42	77	67	65	38	72	57
mp (°C)	278-280	214	220	150-151	241-243	130-132	168-170	189-191	184-186	191	240	215

Scheme 1. Synthesis of novel 8,9-dihydro-7*H*-pyrimido[4,5-*b*][1,4]diazepines.

range δ 38.7–40.5 ppm whereas the signal of C-8 appears at δ 57.6–59.2 ppm. Quaternary carbon C-4a presents a peak at δ 105.1–107.9 ppm (except for the particularly high value of **4b** with δ 116.5 ppm). In contrast, signals for C-9a are displayed between δ 151.0 and 163.3 ppm. This behavior can be explained in terms of the strong push–pull effect of the NH_2 and C=N groups attached to the C=C double bond in structures **4** and **5**. HMBC experiments for all derivatives indicate three-bond correlations between 9-H (diazepine NH) and carbons C-4a and C-7, also observed between C-9a and protons 2-H and 8-H. Mass spectra of compounds **4** and **5** show well defined molecular ions with a characteristic fragmentation pattern involving the loss of styrene derivatives (according to the aryl group present in C-8) as neutral molecules, determining the *m/z* values of the resulting ions. This fragmentation occurs by type-B homolytic processes (excision on C–C bond in α position to N-9) with stabilization of the formed radical [27].

2.2. Anticancer activity

The two-stage screening process started with the evaluation of all compounds against the 60 cell lines at a single dose of 1.0 μM. The output from the single dose screen was reported as a mean graph available for analysis by the COMPARE program. This primary assay showed that compounds **4a**, **4b**, **4f** and **5f** were essentially inactive, while compounds **4e**, **5a** and **5b** were declared active (Table 1). Therefore, a secondary screening was performed, in order to determine their cytostatic activity, against the 60 cell panel representing leukemia, melanoma and cancers of the lung, colon, brain, ovary, breast, prostate, and kidney. The compounds were evaluated at five concentration levels (100, 10, 1.0, 0.1 and 0.01 μM). The test consisted of a 48 h continuous drug exposure protocol and sulforhodamide B (SRB) protein assay was used to estimate cell growth. Details of this evaluation method, and the complementary information related with the activity pattern over all cell lines, have been published [28–30]. As shown in Table 2, compound **4e** presented remarkable activity against 47 human tumor cell lines, being the most sensitive to growth inhibition HL-60 (TB) and SR of leukemia, with GI_{50} values of 0.72 and 0.25 μM, respectively (cytotoxicity expressed as LC_{50} with values >100 μM for both cell lines), while **5a** (active against 11 human tumor cell lines) exhibited a GI_{50} of 0.19 μM ($LC_{50} > 100$ μM) for RPMI-8226 cell line of the same panel. Compounds **5a** and **5b** (the latter active against 37 human tumor cell lines) were found especially effective against the cell line IGROV1 corresponding to ovarian cancer panel with GI_{50} of 0.068 and 0.35 μM, respectively (LC_{50} of 25.5 μM for **5a** and 42.8 μM for **5b**). The testing results show significant activities (Table 2), with GI_{50} ranges of 0.25–40.0 μM for **4e**, 0.068–92.6 μM for **5a** and 0.35–92.6 μM for **5b** against other cell lines from the different panels of cancer types. The cytotoxicities associated with compounds **4e**, **5a** and **5b**, and measured as LC_{50} go from 23.3 μM to >100 μM, for all cell lines.

Table 1
Results of primary anticancer assay for seven compounds selected by the NCI

Compound	Activity[a]
4a	NA
4b	NA
4e	A
4f	NA
5a	A
5b	A
5f	NA

[a] Activity denoted as: A = active; NA = not active.

3. Conclusion

We described herein the synthesis and biological activity of new 8,9-dihydro-7*H*-pyrimido[4,5-*b*][1,4]diazepine derivatives bearing a benzodioxole moiety and variously substituted aryl group. Investigation of anticancer screening data reveals that among the seven pyrimidodiazepines evaluated, derivatives **4a**, **5a** and **5b** showed high activity against different cancer cell lines with remarkable values in leukemia and ovarian cancer panels. Given the significant results obtained, chemical studies are being conducted to improve the antitumor activity of such compounds, as well as other biological studies focused on antiviral and antifungal activities.

4. Experimental

Melting points were determined in a Buchi Melting Point Apparatus and are uncorrected. The ^1H and ^{13}C NMR spectra were run on a Bruker DPX 400 spectrometer operating at 400 and 100 MHz, respectively, using dimethyl sulfoxide-d_6 as solvent and tetramethylsilane as internal standard. The mass spectra were obtained on a Hewlett Packard HP Engine-5989 spectrometer (equipped with a direct inlet probe) operating at 70 eV. The elemental analyses have been obtained using a LECO CHNS-900 elemental analyzer. Reactions under microwave irradiation were performed using the CEM Discover™ monomode system in open vessels under magnetic stirring for 2–5 min at 150 °C, and with power/time control using a ramp of 100–300 W.

4.1. General procedure for synthesis of 4-amino-6-aryl-8-(1,3-benzodioxol-5-yl)-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepines (*4a–f*) and 4-amino-8-aryl-6-(1,3-benzodioxol-5-yl)-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepines (*5a–f*)

A mixture of dihydrochloride of triamine **1** (1.53 mmol) and methylenedioxychalcones **2a–f** or **3a–f** (1.53 mmol) with 0.2 mL of dry dimethylformamide, was subjected to microwave irradiation in open vessels under magnetic stirring for 2–5 min. Reaction progress was monitored by TLC. After cooling, the reaction mixture was treated by addition of dry ethanol (5 mL) with mixing, giving rise to formation of precipitate which was filtered and washed with fractions of water and cold ethanol yielding compounds **4a–c** and **5a–c**. For compounds **4d–f** and **5d–f**, the resulting precipitate was filtered

Table 2
In vitro testing results expressed as growth inhibition of cancer cell lines for compounds **4e**, **5a** and **5b**[a]

Compound	Number of cell lines			Most sensitive cell lines ($GI_{50} < 20$ μM)			
	Investigated	Giving positive GI_{50}		Panel	Cell line	GI_{50}[b] (μM)	LC_{50}[c] (μM)
		GI_{50}[b] (μM)					
		No.	Range				
4e	53	50	0.25–40.0	Leukemia	HL-60 (TB)	0.72	>100
					K-562	4.19	>100
					MOLT-4	2.66	>100
					SR	0.25	>100
				Non-small cell lung cancer	A549/ATCC	7.09	>100
					EKVX	16.2	>100
					NCI-H226	12.6	>100
					NCI-H23	10.2	>100
					NCI-H322M	16.5	>100
					NCI-H460	3.59	>100
					NCI-H522	3.67	>100
				Colon cancer	COLO 205	3.47	>100
					HCC-2998	6.29	>100
					HCT-15	3.14	55.2
					HT29	3.81	>100
					KM12	3.79	75.1
					SW-620	3.56	>100
				CNS cancer	SF-268	4.06	52.2
					SF-295	3.45	>100
					SF-539	4.98	69.9
					SNB-75	4.55	>100
					U251	4.55	44.5
				Melanoma	LOX IMVI	6.38	81.5
					MALME-3M	3.01	>100
					SK-MEL-28	10.2	>100
					SK-MEL-5	3.85	>100
					UACC-62	4.09	>100
				Ovarian cancer	IGROV1	3.34	>100
					OVCAR-3	2.75	23.3
					OVCAR-4	17.7	>100
					OVCAR-8	13.5	>100
					SK-OV-3	15.8	>100
				Renal cancer	A498	6.33	>100
					ACHN	7.74	>100
					CAKI-1	1.44	>100
					RXF 393	3.50	>100
					SN12C	8.61	>100
					TK-10	4.47	69.7
					UO-31	10.8	65.2
				Prostate cancer	DU-145	4.52	46.7
				Breast cancer	MCF7	3.42	>100
					NCI/ADR-RES	3.52	>100
					MDA-MB-231/ATCC	4.37	55.6
					HS 578T	7.28	>100
					MDA-MB-435	1.15	90.8
					T-47D	5.72	>100
					MDA-MB-468	3.68	>100
5a	57	47	0.068–92.6	Leukemia	HL-60 (TB)	13.1	>100
					RPMI-8226	0.19	>100
					SR	2.37	>100
				Non-small cell lung cancer	NCI-H522	6.78	>100
				Colon cancer	HCT-116	18.2	>100
					HCT-15	9.55	>100
				Ovarian cancer	IGROV1	0.068	25.5
				Renal cancer	RXF 393	7.19	>100
					UO-31	18.1	>100
				Breast cancer	HS 578T	9.18	>100
					BT-549	1.68	>100

Table 2 (*continued*)

Compound	Number of cell lines			Most sensitive cell lines ($GI_{50} < 20$ μM)			
	Investigated	Giving positive GI_{50}		Panel	Cell line	GI_{50}[b] (μM)	LC_{50}[c] (μM)
		GI_{50}[b] (μM)					
		No.	Range				
5b	56	53	0.35–92.6	Leukemia	HL-60 (TB)	5.37	>100
					K-562	4.27	>100
					MOLT-4	5.85	>100
					RPMI-8226	1.50	>100
				Non-small cell	A549/ATCC	11.6	78.3
				lung cancer	HOP-62	16.6	>100
					NCI-H322M	16.9	>100
					NCI-H522	2.37	>100
				Colon cancer	COLO 205	15.0	>100
					HCC-2998	5.88	>100
					HCT-116	10.7	47.5
					HCT-15	3.05	>100
					HT29	17.2	>100
					KM12	2.03	>100
					SW-620	4.91	>100
				CNS cancer	SF-268	8.52	>100
					SF-295	14.7	>100
					SNB-75	5.22	>100
					U251	6.19	78.9
				Melanoma	LOX IMVI	10.1	>100
				Ovarian cancer	IGROV1	0.35	42.8
					OVCAR-3	3.71	83.0
					OVCAR-4	4.58	>100
					OVCAR-5	18.3	>100
					OVCAR-8	4.66	>100
				Renal cancer	A498	5.94	>100
					ACHN	8.25	>100
					RXF 393	2.51	59.3
					TK-10	8.23	>100
					UO-31	7.48	>100
				Prostate cancer	PC-3	9.06	50.8
					DU-145	16.8	65.9
				Breast cancer	MCF7	3.61	>100
					NCI/ADR-RES	5.35	>100
					MDA-MB-231/ATCC	2.70	>100
					HS 578T	1.07	>100
					MDA-MB-468	10.1	>100

[a] Data obtained from NCI's in vitro disease-oriented human tumor cell line screen [27–29].
[b] GI_{50} was the drug concentration resulting in a 50% reduction in the net protein increase (as measured by SRB staining) in control cells during the drug incubation. Determined at five concentration levels (100, 10, 1.0, 0.1 and 0.01 μM).
[c] LC_{50} is a parameter of cytotoxicity and reflects the molar concentration needed to kill 50% of the cells.

and purified by silica gel chromatography with methylene chloride:ethanol (30:1) as eluent.

4.1.1. 4-Amino-8-(1,3-benzodioxol-5-yl)-6-(4-nitrophenyl)-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepine (**4a**)

The compound was obtained as a yellow-red powder in a yield of 55%; IR (KBr, cm^{-1}): ν 3353, 3158 for NH and NH_2, 1572, 1342 for NO_2, 1657, 1597 for C=N and C=C; ^{1}H NMR (400 MHz, DMSO-d_6): δ 3.05 (dd, 2H, 7-H_A, $^2J_{AM} = 14.5$ Hz, $^3J_{AX} = 1.9$ Hz), 3.94 (dd, 2H, 7-H_M, $^2J_{AM} = 14.5$ Hz, $^3J_{MX} = 6.3$ Hz), 5.23 (m, 1H, 8-H_X), 5.90 (s, 2H, OCH_2O), 6.62 (s, 1H, H*o*′, Aryl B), 6.75 (d, 1H, H*m*, Aryl B, $J = 8.0$ Hz), 6.81 (d, 1H, H*o*, Aryl B, $J = 8.0$ Hz), 8.04 (d, 1H, H*o*, Aryl A, $J = 9.0$ Hz), 8.12 (d, 1H, H*m*, Aryl A, $J = 9.0$ Hz), 8.27 (s, 1H, 2-H), 9.21 (d, 1H, 9-H, $J = 5.3$ Hz); ^{13}C NMR (400 MHz, DMSO-d_6): δ 40.57 (C-7), 59.02 (C-8), 101.44 (OCH_2O), 105.29 (C-4a), 107.12 (C*o*, Aryl B), 108.42 (C*m*, Aryl B), 119.73 (C*o*′, Aryl B), 123.54 (C*m*, Aryl A), 128.94 (C*o*, Aryl A), 136.65 (C*i*, Aryl B), 145.17 (C*i*, Aryl A), 146.77 (C-2), 147.23 (C*m*′, Aryl B), 147.73 (C*p*, Aryl B), 148.30 (C*p*, Aryl A), 151.90 (C-9a), 156.50 (C-6), 162.92 (C-4); mass (*m*/*z*, %): 404 (M^+, 19), 389 (14), 256 (10), 148 (100), 109 (10), 76 (6), 43 (5). Anal. Calcd. for $C_{20}H_{16}N_6O_4$: C, 59.40; H, 3.99; N, 20.78. Found: C, 59.57; H, 3.96; N, 20.47.

4.1.2. 4-Amino-8-(1,3-benzodioxol-5-yl)-6-(4-chlorophenyl)-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepine (**4b**)

The compound was obtained as a white-yellow powder in a yield of 62%; IR (KBr, cm^{-1}): ν 3444, 3214, 3126 for NH

and NH_2, 1609, 1566 for C=N and C=C; ^{1}H NMR (400 MHz, DMSO-d_6): δ 2.79 (dd, 2H, 7-H_A, $^2J_{AM} = 14.4$ Hz, $^3J_{AX} = 2.0$ Hz), 3.69 (dd, 2H, 7-H_B, $^2J_{AM} = 14.4$ Hz, $^3J_{MX} = 6.2$ Hz), 5.04 (m, 1H, 8-H_X), 5.85 (s, 2H, OCH_2O), 6.51 (s, 2H, NH_2), 6.57 (d, 1H, H*o*, Aryl B, $J = 8.2$ Hz), 6.68 (d, 1H, H*m*, Aryl B, $J = 8.0$ Hz), 6.69 (s, 1H, H*o*′, Aryl B), 7.28 (d, 1H, H*o*, Aryl A, $J = 8.7$ Hz), 7.48 (d, 1H, 9-H, $J = 5.5$ Hz), 7.70 (d, 1H, H*m*, Aryl A, $J = 8.7$ Hz), 7.73 (s, 1H, 2-H); ^{13}C NMR (400 MHz, DMSO-d_6): δ 39.75 (C-7), 57.62 (C-8), 110.76 (OCH_2O), 116.52 (C*o*, Aryl B), 116.84 (C-4a), 117.78 (C*m*, Aryl B), 129.08 (C*o*′, Aryl B), 137.93 (C*o*, Aryl A), 138.54 (C*m*, Aryl A), 143.89 (C*p*, Aryl A), 148.03 (C*i*, Aryl B), 148.87 (C*i*, Aryl A), 155.87 (C*p*, Aryl B), 157.02 (C*m*′, Aryl B), 163.29 (C-9a), 165.45 (C-2), 169.79 (C-6), 172.29 (C-4); mass (*m*/*z*, %): 393 (M^+, 19), 378 (19), 245 (24), 148 (100), 109 (12), 76 (5), 43 (6). Anal. Calcd. for $C_{20}H_{16}ClN_5O_2$: C, 60.99; H, 4.09; N, 17.78. Found: C, 60.85; H, 4.12; N, 17.91.

4.1.3. 4-Amino-8-(1,3-benzodioxol-5-yl)-6-(4-bromophenyl)-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepine (**4c**)

The compound was obtained as a yellow powder in a yield of 68%; IR (KBr, cm^{-1}): ν 3455, 3211, 3083 for NH and NH_2, 1606, 1567 for C=N and C=C; ^{1}H NMR (400 MHz, DMSO-d_6): δ 2.78 (dd, 2H, 7-H_A, $^2J_{AM} = 14.5$ Hz, $^3J_{AX} = 1.9$ Hz), 3.69 (dd, 2H, 7-H_M, $^2J_{AM} = 14.5$ Hz, $^3J_{MX} = 5.6$ Hz), 5.03 (m, 1H, 8-H_X), 5.85 (s, 2H, OCH_2O), 6.50 (s, 2H, NH_2), 6.57 (s, 1H, H*o*′, Aryl B), 6.68 (d, 1H, H*m*, Aryl B, $J = 7.4$ Hz), 6.69 (d, 1H, H*o*, Aryl B, $J = 7.6$ Hz), 7.41 (d, 1H, H*o*, Aryl A, $J = 8.3$ Hz), 7.47 (d, 1H, 9-H, $J = 4.2$ Hz), 7.62 (d, 1H, H*m*, Aryl A, $J = 8.5$ Hz), 7.74 (s, 1H, 2-H); ^{13}C NMR (400 MHz, DMSO-d_6): δ 39.52 (C-7), 58.80 (C-8), 100.80 (OCH_2O), 106.55 (C*o*, Aryl B), 106.86 (C-4a), 107.82 (C*m*, Aryl B), 119.10 (C*o*′, Aryl B), 122.80 (C*p*, Aryl A), 128.43 (C*m*, Aryl A), 130.90 (C*o*, Aryl A), 138.07 (C*i*, Aryl B), 139.26 (C*i*, Aryl A), 145.90 (C*m*′, Aryl B), 147.05 (C*p*, Aryl B), 153.33 (C-9a), 155.52 (C-2), 159.87 (C-6), 162.34 (C-4); mass (*m*/*z*, %): 439 (M^+, 10), 424 (10), 291 (13), 148 (100), 109 (10), 76 (6), 43 (6). Anal. Calcd. for $C_{20}H_{16}BrN_5O_2$: C, 54.81; H, 3.68; N, 15.98. Found: C, 54.85; H, 3.48; N, 15.81.

4.1.4. 4-Amino-8-(1,3-benzodioxol-5-yl)-6-phenyl-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepine (**4d**)

The compound was obtained as a white powder in a yield of 35%; IR (KBr, cm^{-1}): ν 3475, 3414, 3230 for NH and NH_2, 1639 for C=N and C=C; ^{1}H NMR (400 MHz, DMSO-d_6): δ 3.03 (dd, 2H, 7-H_A, $^2J_{AM} = 14.4$ Hz, $^3J_{AX} = 2.0$ Hz), 3.83 (dd, 2H, 7-H_M, $^2J_{AM} = 14.4$ Hz, $^3J_{MX} = 6.2$ Hz), 5.18 (m, 1H, 8-H_X), 5.90 (s, 2H, OCH_2O), 6.65 (d, 1H, H*o*, Aryl B, $J = 7.8$ Hz), 6.76 (d, 1H, H*m*, Aryl B, $J = 8.0$ Hz), 6.82 (s, 1H, H*o*′, Aryl B), 7.29–7.38 (m, 3H, H*m* and H*p*, Aryl A), 7.78 (d, 1H, H*o*, Aryl A, $J = 7.3$ Hz), 7.96 (s, 2H, NH_2), 8.23 (s, 1H, 2-H), 8.95 (d, 1H, 9-H, $J = 4.9$ Hz); ^{13}C NMR (400 MHz, DMSO-d_6): δ 39.90 (C-7), 59.24 (C-8), 101.38 (OCH_2O), 105.42 (C-4a), 107.22 (C*o*, Aryl B), 108.35 (C*m*, Aryl B), 119.83 (C*o*′, Aryl B), 127.73 (C*o*, Aryl A), 128.55 (C*m*, Aryl A), 130.48 (C*p*, Aryl A), 136.91 (C*i*, Aryl B), 139.58 (C*i*, Aryl A), 146.34 (C-2), 146.72 (C*p*, Aryl B), 147.66 (C*m*′, Aryl B), 151.64 (C-9a), 156.33 (C-4), 165.40 (C-6); mass (*m*/*z*, %): 359 (M^+, 28), 344 (22), 211 (47), 148 (100), 109 (13), 76 (9), 43 (13). Anal. Calcd. for $C_{20}H_{17}N_5O_2$: C, 66.84; H, 4.77; N, 19.49. Found: C, 66.63; H, 4.82; N, 19.43.

4.1.5. 4-Amino-8-(1,3-benzodioxol-5-yl)-6-(4-methylphenyl)-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepine (**4e**)

The compound was obtained as a beige powder in a yield of 51%; IR (KBr, cm^{-1}): ν 3462, 3412 for NH and NH_2, 1640 for C=N and C=C; ^{1}H NMR (400 MHz, DMSO-d_6): δ 2.24 (s, 3H, *p*-CH_3), δ 2.96 (dd, 2H, 7-H_A, $^2J_{AM} = 14.1$ Hz, $^3J_{AX} = 2.0$ Hz), 3.76 (dd, 2H, 7-H_M, $^2J_{AM} = 14.1$ Hz, $^3J_{MX} = 5.8$ Hz), 5.11 (m, 1H, 8-H_X), 5.87 (s, 2H, OCH_2O), 6.61 (d, 1H, H*o*, Aryl B, $J = 8.0$ Hz), 6.72 (d, 1H, H*m*, Aryl B, $J = 8.0$ Hz), 6.78 (s, 1H, H*o*′, Aryl B), 7.08 (d, 1H, H*o*, Aryl A, $J = 8.1$ Hz), 7.66 (d, 1H, H*m*, Aryl A, $J = 8.2$ Hz), 7.91 (s, 2H, NH_2), 8.17 (s, 1H, 2-H), 8.84 (d, 1H, 9-H, $J = 5.0$ Hz); ^{13}C NMR (400 MHz, DMSO-d_6): δ 20.85 (*p*-CH_3), 39.52 (C-7), 58.79 (C-8), 100.93 (OCH_2O), 105.06 (C-4a), 106.79 (C*o*, Aryl B), 107.91 (C*m*, Aryl B), 119.40 (C*o*′, Aryl B), 127.31 (C*m*, Aryl A), 128.74 (C*o*, Aryl A), 136.40 (C*i*, Aryl A), 136.49 (C*i*, Aryl B), 139.92 (C*p*, Aryl A), 145.86 (C-2), 146.25 (C*p*, Aryl B), 147.20 (C*m*′, Aryl B), 151.14 (C-9a), 155.99 (C-4), 164.80 (C-6); mass (*m*/*z*, %): 373 (M^+, 47), 358 (43), 225 (100), 148 (86), 109 (21), 76 (8), 43 (17). Anal. Calcd. for $C_{21}H_{19}N_5O_2$: C, 67.55; H, 5.13; N, 18.76. Found: C, 67.36; H, 5.16; N, 18.57.

4.1.6. 4-Amino-8-(1,3-benzodioxol-5-yl)-6-(4-methoxyphenyl)-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepine (**4f**)

The compound was obtained as a yellow powder in a yield of 42%; IR (KBr, cm^{-1}): ν 3460, 3415, 3213 for NH and NH_2, 1591, 1566 for C=N and C=C; ^{1}H NMR (400 MHz, DMSO-d_6): δ 2.96 (dd, 2H, 7-H_A, $^2J_{AM} = 14.5$ Hz, $^3J_{AX} = 1.9$ Hz), 3.74 (s, 3H, *p*-OCH_3), 3.86 (dd, 2H, 7-H_M, $^2J_{AM} = 14.5$ Hz, $^3J_{MX} = 5.5$ Hz), 5.09 (m, 1H, 8-H_X), 5.93 (s, 2H, OCH_2O), 6.64 (d, 1H, H*o*, Aryl B, $J = 8.3$ Hz), 6.74 (d, 1H, H*m*, Aryl B, $J = 8.1$ Hz), 6.80 (s, 1H, H*o*′, Aryl B), 7.20 (d, 1H, H*o*, Aryl A, $J = 8.6$ Hz), 7.70 (d, 1H, H*m*, Aryl A, $J = 8.6$ Hz), 7.89 (s, 2H, NH_2), 8.20 (s, 1H, 2-H), 8.78 (d, 1H, 9-H, $J = 5.6$ Hz); ^{13}C NMR (400 MHz, DMSO-d_6): δ 39.60 (C-7), 55.27 (*p*-OCH_3), 58.65 (C-8), 100.95 (OCH_2O), 106.09 (C-4a), 106.33 (C*o*, Aryl B), 108.40 (C*m*, Aryl B), 119.43 (C*o*′, Aryl B), 127.02 (C*m*, Aryl A), 127.74 (C*o*, Aryl A), 131.97 (C*p*, Aryl A), 136.42 (C*i*, Aryl B), 136.76 (C*i*, Aryl A), 145.70 (C-2), 146.24 (C*p*, Aryl B), 147.23 (C*m*′, Aryl B), 151.02 (C-9a), 155.80 (C-4), 164.99 (C-6); mass (*m*/*z*, %): 389 (M^+, 42), 374 (32), 241 (100), 148 (73), 109 (17), 76 (7), 43 (11). Anal. Calcd. for $C_{21}H_{19}N_5O_3$: C, 64.77; H, 4.92; N, 17.98. Found: C, 64.68; H, 5.07; N, 17.84.

*4.1.7. 4-Amino-6-(1,3-benzodioxol-5-yl)-8-(4-nitrophenyl)-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepine (**5a**)*

The compound was obtained as a red powder in a yield of 77%; IR (KBr, cm^{-1}): ν 3374, 3179 for NH and NH_2, 1642, 1583 for C=N and C=C; ^1H NMR (400 MHz, DMSO-d_6): δ 2.99 (dd, 2H, 7-H_A, $^2J_{AM} = 14.4$ Hz, $^3J_{AX} = 1.9$ Hz), 3.99 (dd, 2H, 7-H_M, $^2J_{AM} = 14.4$ Hz, $^3J_{MX} = 6.12$ Hz), 5.43 (m, 1H, 8-H_X), 6.00 (s, 2H, OCH_2O), 6.77 (d, 1H, H*o*, Aryl A, $J = 8.3$ Hz), 7.15 (d, 1H, H*m*, Aryl A, $J = 8.3$ Hz), 7.48 (d, 2H, H*o*, Aryl B, $J = 9.0$ Hz), 7.50 (s, 1H, H*o*′, Aryl A), 7.58 (s, 2H, NH_2), 8.08 (d, 2H, H*m*, Aryl B, $J = 8.6$ Hz), 8.11 (s, 1H, 2-H), 8.59 (d, 1H, 9-H, $J = 5.7$ Hz); ^{13}C NMR (400 MHz, DMSO-d_6): δ 38.71 (C-7), 59.07 (C-8), 101.48 (OCH_2O), 105.94 (C-4a), 107.10 (C*o*′, Aryl A), 107.39 (C*o*, Aryl A), 122.29 (C*m*, Aryl A), 123.35 (C*m*, Aryl B), 127.55 (C*o*, Aryl B), 133.60 (C*i*, Aryl A), 146.51 (C*p*, Aryl B), 147.55 (C*p*, Aryl A), 148.32 (C-2), 149.05 (C*m*′, Aryl A), 149.88 (C*i*, Aryl B), 152.14 (C-9a), 157.25 (C-4), 162.77 (C-6); mass (*m*/*z*, %): 404 (M^+, 100), 389 (46), 255 (63), 149 (49), 109 (28), 89 (34), 63 (28), 43 (13). Anal. Calcd. for $C_{20}H_{16}N_6O_4$: C, 59.40; H, 3.99; N, 20.78. Found: C, 59.35; H, 4.02; N, 20.71.

*4.1.8. 4-Amino-6-(1,3-benzodioxol-5-yl)-8-(4-chlorophenyl)-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepine (**5b**)*

The compound was obtained as a white powder in a yield of 67%; IR (KBr, cm^{-1}): ν 3414, 3256 for NH and NH_2, 1630, 1585 for C=N and C=C; ^1H NMR (400 MHz, DMSO-d_6): δ 2.98 (dd, 2H, 7-H_A, $^2J_{AM} = 14.4$ Hz, $^3J_{AX} = 2.0$ Hz), 3.90 (dd, 2H, 7-H_M, $^2J_{AM} = 14.8$ Hz, $^3J_{MX} = 6.12$ Hz), 5.27 (m, 1H, 8-H_X), 6.03 (s, 2H, OCH_2O), 6.79 (d, 1H, H*o*, Aryl A, $J = 8.3$ Hz), 7.18 (d, 1H, H*m*, Aryl A, $J = 9.0$ Hz), 7.21 (d, 2H, H*o*, Aryl B, $J = 8.3$ Hz), 7.29 (d, 2H, H*m*, Aryl B, $J = 8.3$ Hz), 7.54 (s, 1H, H*o*′, Aryl A), 7.92 (s, 2H, NH_2), 8.20 (s, 1H, 2-H), 8.52 (d, 1H, 9-H, $J = 5.6$ Hz); ^{13}C NMR (400 MHz, DMSO-d_6): δ 39.17 (C-7), 58.58 (C-8), 101.51 (OCH_2O), 105.19 (C-4a), 107.27 (C*o*′, Aryl A), 107.39 (C*o*, Aryl A), 122.51 (C*m*, Aryl A), 128.02 (C*o*, Aryl B), 128.16 (C*m*, Aryl B), 131.82 (C*p*, Aryl B), 133.52 (C*i*, Aryl A), 141.13 (C*i*, Aryl B), 145.80 (C-2), 147.57 (C*m*′, Aryl A), 149.17 (C*p*, Aryl A), 151.51 (C-9a), 155.60 (C-4), 163.88 (C-6); mass (*m*/*z*, %): 393 (M^+, 75), 378 (50), 255 (100), 147 (31), 109 (32), 89 (38), 63 (24), 43 (15). Anal. Calcd. for $C_{20}H_{16}ClN_5O_2$: C, 60.99; H, 4.09; N, 17.78. Found: C, 60.95; H, 4.03; N, 17.86.

*4.1.9. 4-Amino-6-(1,3-benzodioxol-5-yl)-8-(4-bromophenyl)-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepine (**5c**)*

The compound was obtained as a white-yellow powder in a yield of 65%; IR (KBr, cm^{-1}): ν 3449, 3413, 3257 for NH and NH_2, 1641, 1630, 1585 for C=N and C=C; ^1H NMR (400 MHz, DMSO-d_6): δ 2.97 (dd, 2H, 7-H_A, $^2J_{AM} = 14.6$ Hz, $^3J_{AX} = 1.9$ Hz), 3.88 (dd, 2H, 7-H_M, $^2J_{AM} = 14.5$ Hz, $^3J_{MX} = 5.8$ Hz), 5.24 (m, 1H, 8-H_X), 6.03 (s, 2H, OCH_2O), 6.79 (d, 1H, H*o*, Aryl A, $J = 8.2$ Hz), 7.15 (d, 2H, H*o*, Aryl B, $J = 8.3$ Hz), 7.19 (d, 1H, H*m*, Aryl A, $J = 8.6$ Hz), 7.42 (d, 2H, H*m*, Aryl B, $J = 8.4$ Hz), 7.54 (s, 1H, H*o*′, Aryl A), 7.84 (s, 2H, NH_2), 8.18 (s, 1H, 2-H), 8.80 (d, 1H, 9-H, $J = 5.2$ Hz); ^{13}C NMR (400 MHz, DMSO-d_6): δ 39.93 (C-7), 58.59 (C-8), 101.47 (OCH_2O), 105.30 (C-4a), 107.23 (C*o*′, Aryl A), 107.36 (C*o*, Aryl A), 120.33 (C*p*, Aryl B), 122.42 (C*m*, Aryl A), 128.33 (C*o*, Aryl B), 131.04 (C*m*, Aryl B), 133.57 (C*i*, Aryl A), 141.62 (C*i*, Aryl B), 146.39 (C-2), 147.54 (C*m*′, Aryl A), 149.10 (C*p*, Aryl A), 151.59 (C-9a), 156.06 (C-4), 163.59 (C-6); mass (*m*/*z*, %): 439 (M^+, 41), 424 (25), 255 (100), 147 (32), 109 (37), 89 (41), 63 (34), 43 (30). Anal. Calcd. for $C_{20}H_{16}BrN_5O_2$: C, 54.81; H, 3.68; N, 15.98. Found: C, 54.79; H, 3.76; N, 15.87.

*4.1.10. 4-Amino-6-(1,3-benzodioxol-5-yl)-8-phenyl-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepine (**5d**)*

The compound was obtained as a white powder in a yield of 38%; IR (KBr, cm^{-1}): ν 3470, 3415, 3242 for NH and NH_2, 1645, 1587 for C=N and C=C; ^1H NMR (400 MHz, DMSO-d_6): δ 2.95 (dd, 2H, 7-H_A, $^2J_{AM} = 13.3$ Hz, $^3J_{AX} = 1.8$ Hz), 3.81 (dd, 2H, 7-H_M, $^2J_{AM} = 13.5$ Hz, $^3J_{MX} = 5.8$ Hz), 5.21 (m, 1H, 8-H_X), 5.97 (s, 2H, OCH_2O), 6.72 (d, 1H, H*o*, Aryl A, $J = 7.6$ Hz), 7.10–7.20 (m, 6H, H*m*, Aryl A; H*o*, H*m*, and H*p*, Aryl B), 7.15 (s, 1H, 2-H), 7.45 (s, 1H, H*o*′, Aryl A), 7.84 (s, 2H, NH_2), 8.80 (d, 1H, 9-H, $J = 4.0$ Hz); ^{13}C NMR (400 MHz, DMSO-d_6): δ 39.93 (C-7), 59.16 (C-8), 101.42 (OCH_2O), 105.10 (C-4a), 107.26 (C*o*′, Aryl A), 107.31 (C*o*, Aryl A), 122.43 (C*m*, Aryl A), 126.02 (C*o*, Aryl B), 127.31 (C*p*, Aryl B), 128.20 (C*m*, Aryl B), 133.67 (C*i*, Aryl A), 142.16 (C*i*, Aryl B), 145.92 (C-2), 147.44 (C*m*′, Aryl A), 149.00 (C*p*, Aryl A), 151.59 (C-9a), 155.78 (C-4), 164.01 (C-6); mass (*m*/*z*, %): 359 (M^+, 100), 344 (64), 255 (79), 147 (28), 109 (24), 89 (25), 63 (14), 43 (10). Anal. Calcd. for $C_{20}H_{17}N_5O_2$: C, 66.84; H, 4.77; N, 19.49. Found: C, 66.71; H, 4.72; N, 19.52.

*4.1.11. 4-Amino-6-(1,3-benzodioxol-5-yl)-8-(4-methylphenyl)-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepine (**5e**)*

The compound was obtained as a beige powder in a yield of 72%; IR (KBr, cm^{-1}): ν 3468, 3414 for NH and NH_2, 1643 for C=N and C=C; ^1H NMR (400 MHz, DMSO-d_6): δ 2.20 (s, 3H, *p*-CH_3), 2.93 (dd, 2H, 7-H_A, $^2J_{AM} = 14.5$ Hz, $^3J_{AX} = 2.1$ Hz), 3.57 (dd, 2H, 7-H_M, $^2J_{AM} = 14.5$ Hz, $^3J_{MX} = 6.6$ Hz), 5.05 (m, 1H, 8-H_X), 5.98 (s, 2H, OCH_2O), 6.25 (s, 2H, NH_2), 6.76 (d, 2H, H*m*, Aryl A, $J = 8.3$ Hz), 7.02 (d, 2H, H*m*, Aryl B, $J = 8.1$ Hz), 7.03 (s, 1H, 9-H), 7.09 (d, 2H, H*o*, Aryl B, $J = 8.1$ Hz), 7.15 (d, 1H, H*o*, Aryl A, $J = 8.3$ Hz), 7.32 (s, 1H, H*o*′, Aryl A), 7.76 (s, 1H, 2-H); ^{13}C NMR (400 MHz, DMSO-d_6): δ 20.10 (*p*-CH_3), 39.27 (C-7), 58.86 (C-8), 100.83 (OCH_2O), 106.44 (C-4a; C*o*′, Aryl A), 107.02 (C*m*, Aryl A), 121.10 (C*o*, Aryl A), 125.54 (C*o*, Aryl B), 128.28 (C*m*, Aryl B), 134.64 (C*i*, Aryl A), 135.63 (C*i*, Aryl B), 140.54 (C*p*, Aryl B), 148.24 (C*m*′, Aryl A), 148.00 (C*p*, Aryl A), 152.90 (C-9a), 154.59 (C-2), 160.41 (C-6), 161.76 (C-4); mass (*m*/*z*, %): 373 (M^+, 79), 358 (54), 255 (100), 147 (17), 109 (21), 89 (17), 63 (10), 43

(8). Anal. Calcd. for $C_{21}H_{19}N_5O_2$: C, 67.55; H, 5.13; N, 18.76. Found: C, 67.42; H, 5.18; N, 18.69.

*4.1.12. 4-Amino-6-(1,3-benzodioxol-5-yl)-8-(4-methoxyphenyl)-8,9-dihydro-7H-pyrimido[4,5-b][1,4]diazepine (**5f**)*

The compound was obtained as a white-yellow powder in a yield of 57%; IR (KBr, cm^{-1}): ν 3457, 3417, 3219 for NH and NH_2, 1589, 1568 for C=N and C=C; ^1H NMR (400 MHz, DMSO-d_6): δ 2.20 (s, 3H, *p*-OCH_3), 2.93 (dd, 2H, 7-H_A, $^2J_{AM} = 14.5$ Hz, $^3J_{AX} = 1.9$ Hz), 3.54 (dd, 2H, 7-H_M, $^2J_{AM} = 14.5$ Hz, $^3J_{MX} = 6.6$ Hz), 5.04 (m, 1H, 8-H_X), 5.98 (s, 2H, OCH_2O), 6.23 (s, 2H, NH_2), 6.77 (s, 2H, H*m*, Aryl A), 6.78 (d, 2H, H*m*, Aryl B, $J = 8.1$ Hz), 6.97 (d, 1H, 9-H, $J = 3.3$ Hz), 7.12 (d, 2H, H*o*, Aryl B, $J = 8.7$ Hz), 7.15 (d, 1H, H*o*, Aryl A, $J = 8.3$ Hz), 7.33 (s, 1H, H*o*′, Aryl A), 7.76 (s, 1H, 2-H); ^{13}C NMR (400 MHz, DMSO-d_6): δ 39.37 (C-7), 54.78 (*p*-OCH_3), 58.68 (C-8), 100.82 (OCH_2O), 106.44 (C*o*′, Aryl A), 107.02 (C-4a; C*m*, Aryl A), 113.33 (C*o*, Aryl B), 121.08 (C*o*, Aryl A), 126.76 (C*m*, Aryl B), 134.61 (C*i*, Aryl A), 138.76 (C*i*, Aryl B), 147.09 (C*m*′, Aryl A), 148.02 (C*p*, Aryl A), 152.81 (C-9a), 154.61 (C-2), 158.04 (C*p*, Aryl B), 160.49 (C-6), 161.75 (C-4); mass (*m*/*z*, %): 389 (M^+, 40), 374 (34), 255 (100), 147 (10), 109 (16), 89 (12), 63 (8), 43 (6). Anal. Calcd. for $C_{21}H_{19}N_5O_3$: C, 64.77; H, 4.92; N, 17.98. Found: C, 64.70; H, 4.98; N, 17.89.

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References

[1] A. Nawojski, W. Nawrocka, Rocz. Chem. 51 (1977) 2117–2120; Chem. Abstr. 88 (1978) 136578.
[2] F.G. Yaremenko, V.D. Orlov, N.N. Kolos, V.F. Lavrushin, Khim. Geterotsikl. Soedin. (1979) 848–851; Chem. Abstr. 91 (1979) 123717.
[3] V.D. Orlov, J. Quiroga, N.N. Kolos, Khim. Geterotsikl. Soedin. (1987) 363–369; Chem. Abstr. 107 (1987) 217603.
[4] B. Insuasty, R. Abonia, J. Quiroga, An. Quim. 88 (1992) 718–721; Chem. Abstr. 119 (1992) 49356.
[5] V.D. Orlov, N.N. Kolos, J. Quiroga, Z. Kaluski, E. Figas, A. Potekhin, Khim. Geterotsikl. Soedin. (1992) 506–510; Chem. Abstr. 118 (1993) 38896.
[6] B. Insuasty, M. Ramos, J. Quiroga, A. Sánchez, M. Nogueras, N. Hanold, H. Meier, J. Heterocycl. Chem. 31 (1994) 61–64.
[7] B. Insuasty, M. Ramos, R. Moreno, J. Quiroga, A. Sánchez, M. Nogueras, N. Hanold, M. Meier, J. Heterocycl. Chem. 32 (1995) 1229–1233.
[8] B. Insuasty, R. Abonia, J. Quiroga, H. Meier, J. Heterocycl. Chem. 30 (1993) 229–231.
[9] J.K. Landquist, in: A.R. Katritzky, C.N. Russ (Eds.), Comprehensive Heterocyclic Chemistry, Pergamon, Oxford, 1984, p. 166.
[10] H. Schutz, Benzodiazepines, Springer, Heidelberg, 1982.
[11] A. Constanzo, F. Bruni, G. Auzzi, S. Selleri, L.P. Vettori, J. Heterocycl. Chem. 27 (1990) 695–697.
[12] T.A. Kelly, D.W. McNeil, J.M. Rose, E. David, C.K. Shih, P.M. Grob, J. Med. Chem. 40 (1977) 2430–2433 and references therein.
[13] H.A. DeWald, S. Lobbestaell, B.P.H. Poschel, J. Med. Chem. 24 (1981) 982–987.
[14] B. Insuasty, H. Insuasty, J. Quiroga, M. Nogueras, A. Sánchez, M.D. López, J. Heterocycl. Chem. 36 (1999) 933–936.
[15] A. Loupy, A. Petit, J. Hamelin, F. Texier-Boullet, P. Jacquault, D. Mathe, Synthesis (1998) 1213–1234.
[16] R.S. Varma, Green Chem. (1999) 43–55.
[17] S. Deshayes, M. Liagre, A. Loupy, J.-L. Luche, A. Petit, Tetrahedron 55 (1999) 10851–10870.
[18] K. Tanaka, F. Toda, Chem. Rev. 100 (2000) 1025–1074.
[19] R.S. Varma, Clean Prod. Process. 1 (1999) 132–147.
[20] R.S. Varma, Pure Appl. Chem. 73 (2001) 193–198.
[21] J. Quiroga, S. Cruz, B. Insuasty, R. Abonia, Heterocycl. Commun. 6 (2000) 275–282.
[22] J. Quiroga, C. Cisneros, B. Insuasty, R. Abonia, M. Nogueras, A. Sánchez, Tetrahedron Lett. 42 (2001) 5625–5627.
[23] J. Quiroga, A. Rengifo, B. Insuasty, R. Abonía, M. Nogueras, A. Sánchez, Tetrahedron Lett. 43 (2002) 9061–9063.
[24] M.R. Boyd, K.D. Paull, Drug Dev. Res. 34 (1995) 91–109.
[25] L.A. Yanovskaya, G.V. Krishtal, V.V. Kulganek, Usp. Khim. 53 (1984) 1280–1303; Chem. Abstr. 101 (1984) 190667x.
[26] V.D. Orlov, I.Z. Papiashvili, P.A. Grigorov, Khim. Geterotsikl. Soedin (1983) 671–676; Chem. Abstr. 99 (1983) 53717q.
[27] J. Quiroga, B. Insuasty, G. Gallo, Bol. Soc. Chil. Quím. 41 (1996) 415–421.
[28] M.R. Boyd, K.D. Paull, Drug Dev. Res. 34 (1995) 91–109.
[29] A.P. Monks, D.A. Scudiero, P. Skehan, R.H. Shoemaker, K.D. Paull, C. Vistica, C. Hose, J. Langley, P. Cronise, A. Vaigro-Wolff, M. Gray-Goodrich, H. Campbell, M. Mayo, M. Boyd, J. Natl. Cancer Inst. 83 (1991) 757–766.
[30] J.N. Weinstein, T.G. Myers, P.M. O'Connor, S.H. Friend, A.J. Fornace Jr., K.W. Kohn, T. Fojo, S.E. Bates, L.V. Rubinstein, N.L. Anderson, J.K. Buolanwini, W.W. van Osdol, A.P. Monks, D.A. Scudiero, E.A. Sausiville, D.W. Zaharevitz, B. Bunow, V.N. Viswanadhan, G.S. Jhonson, R.E. Wittes, K.D. Paull, Science 275 (1997) 343–349.