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Short communication

Synthesis and cAMP-dependent phosphodiesterase inhibition of novel thiazoloquinazoline derivatives

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The series of 6,7,8,9-tetrahydro-5*H*-5-(2′-hydroxyphenyl)--2-(4′-substituted benzylidine)thiazolo(2,3-*b*)quinazolin--3(2*H*)-ones (**4a-j**) and 6,7,8,9-tetrahydro-5*H*-5-(2′-hydroxyphenyl)-2-(4′-substituted benzylidine)-3-(4-nitrophenyl amino)thiazoloquinazolines (**5a-j**) were synthesized by the reported method and evaluated for their phosphodiesterase inhibitory activity. All test compounds exhibited good activity. The structure-activity relationships were also studied. In both series of compounds, electron-withdrawing substitutions showed higher activity. Among the tested compounds, 6,7,8,9-tetrahydro-5*H*-5-(2′-hydroxyphenyl)-2--(4′-fluorobenzylidine)-3-(4-nitrophenylamino)thiazoloquinazoline (**5e**), 6,7,8,9-tetrahydro-5*H*-5-(2′-hydroxyphenyl)-2--(4′-nitrobenzylidine)-3-(4-nitrophenylamino)thiazoloquinazoline (**5j**) and 6,7,8,9-tetrahydro-5*H*-5-(2′-hydroxyphenyl)-2--(4′-chlorobenzylidine)-3-(4-nitrophenylamino)thiazoloquinazoline (**5f**) were found to be more potent than theophylline (IC_{50} in mmol L^{-1} of 1.34 ± 0.09 for **5f**, 1.44 ± 0.02 for **5e**, 1.52 ± 0.05 for **5j** *vs*. 1.72 ± 0.09 for theophylline).

Keywords: thiazoloquinazoline, benzylidinethiazoloquinazoline, nitrophenylaminothiazoloquinazoline, phosphodiesterase inhibitions, SAR

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Cyclic 3,5-adenosine monophosphate (cAMP) and cyclic 3,5-guanosine monophosphate (cGMP) are important second messengers that play a central role in mediating a variety of functional responses to hormones and other cellular transmitters (1–4). Sensitivity of the physiological processes regulated by cyclic nucleotides requires precise and rapid regulation of the level of these second messengers according to the requirements of the physiological status of the cell. Precise modulation of the phosphodiesterase (PDE) function is critical for maintaining cyclic nucleotide levels within a narrow concentration range. Due to their key role in the regulation of physiological processes, inhibitors of PDEs can be used as therapeutic tools for various diseases. Relatively less emphasis, however, has been placed on the development of agents that interfere with the

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catabolism of cAMP and cGMP *via* inhibition of phosphodiesterase. A variety of nitrogen and sulfur containing heterocycles are reported to inhibit PDE (5–10). Quinazolines and condensed quinazoline such as hoquizil, prazosin and buqineran, possess PDE inhibitory (11, 12) antimicrobial (13), antiinflammatory (14), anticonvulsant (15) and antihypertensive (16) activity (Fig. 1). Based on these observations, a hypothetical model has been proposed, taking into account the following broad objectives: to develop new fused heterocycles expected to exert PDE inhibitory activity similar to that of theophylline.

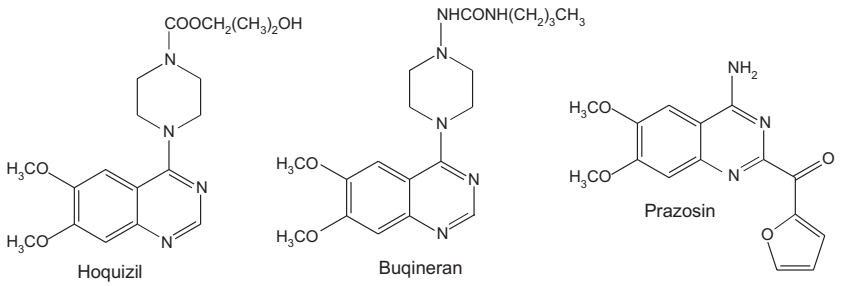


Fig. 1. Quinazoline bearing drugs.

Based on this approach, we attempted to synthesize 6,7,8,9-tetrahydro-5*H*-5-(2′-hydroxyphenyl)-2-(4′-substituted benzylidine)thiazolo(2,3-*b*)quinazolin-3(2*H*)-one (**4a-j**) and 6,7,8,9-tetrahydro-5*H*-5-(2′-hydroxyphenyl)-2-(4′-substituted benzylidine)-3-(4-nitrophenylamino)thiazoloquinazolines (**5a-j**) analogues. The PDE inhibitory activity for all title compounds (**4a-j** and **5a-j**) was tested using a standard technique (4).

EXPERIMENTAL

Bovine heart phosphodiesterase sample was obtained from Sigma (India) and theophylline, as a reference compound, from German Remedies (India).

Melting points were taken in an open capillary tube and were uncorrected. IR spectra were recorded in KBr pellets (ABB Bomem FT-IR spectrometer MB 104, ABB Limited, India). The ^{1}H-NMR spectra were recorded in $CDCl_3$ (Bruker 400 NMR spectrometer, IET Limited) with TMS as internal reference. Mass spectral data were recorded with a quadrupol mass spectrometer (Shimadzu GC MS QP 5000, Japan). Microanalyses were performed using a Vario EL V300 elemental analyzer (Elemental Analysensysteme GmbH, Germany). The purity of the compounds was checked by TLC on pre-coated SiO_2 gel (HF_{254}) aluminium plates (Merck, USA) using ethyl acetate/benzene (1:3) and visualized in a UV chamber. Physico-chemical and spectral data results of these compounds are shown in Table I.

Table I. Spectral data of synthesized compounds

Compd.	Yield (%)	M. p. (°C)	IR (KBr) (ν, cm^{-1})	^1H NMR (δ, ppm)	*m/z*	Molecular formula Analysis: calcd./found (%)		
						C	H	N
3	71	153–155	3402 (O-H), 3046 (Ar-CH), 1719 (C=O), 1462 (C=C)	6.61–6.89 (m, 4H Ar-H), 5.71 (s, 1H, -CH) 9.91 (s, 1H, Ar-OH), 3.76 (s, 2H, $-CH_2$) 1.6–2.42 (m, 8H, 4 × CH_2)	300 [M+] (06)	$C_{16}H_{16}N_2O_2S$		
						63.98	5.37	9.32
						63.92	5.28	9.30
4a	82	159–161	3474 (O-H), 3093 (Ar-CH), 1725 (C=O), 1461 (C=C)	6.92–7.56 (m, 9H, Ar-H), 6.63 (s, 1H, =CH), 5.81 (s, 1H, H-5), 9.74 (s, 1H, Ar-OH), 1.58–2.67 (m, 8H, 4 × CH_2)	377 [M+] (38)	$C_{23}H_{20}N_2O_2S$		
						71.11	5.19	7.21
						71.19	5.26	7.14
4b	79	176–178	3442 (O-H), 3041 (Ar-CH), 1723 (C=O), 1431 (C=C)	6.81–7.71 (m, 8H, Ar-H), 6.73 (s, 1H, =CH), 5.71 (s, 1H, H-5), 9.76 (s, 2H, Ar-OH), 1.61–2.35 (m, 8H, 4 × CH_2)	404 [M+] (49)	$C_{23}H_{20}N_2O_3S$		
						68.30	4.98	6.93
						68.37	4.87	6.99
4c	78	183–185	3476 (O-H), 3096 (Ar-CH), 1728 (C=O), 1468 (C=C)	6.96–7.54 (m, 8H, Ar-H), 6.67 (s, 1H, =CH), 5.83 (s, 1H, H-5), 9.84 (s, 1H, Ar-OH), 3.75 (s, 3H $-OCH_3$), 1.58–2.62 (m, 8H, 4 × CH_2)	418 [M+] (67)	$C_{24}H_{22}N_2O_3S$		
						68.88	5.30	6.69
						68.90	5.33	6.72
4d	76	186–188	3448 (O-H), 3049 (Ar-CH), 1721 (C=O), 1434 (C=C)	6.86–7.74 (m, 8H, Ar-H), 6.72 (s, 1H, =CH), 5.76 (s, 1H, H-5), 9.76 (s, 1H, Ar-OH), 2.20 (s, 3H $-CH_3$), 1.62–2.32 (m, 8H, 4 × CH_2)	402 [M+] (62)	$C_{24}H_{22}N_2O_2S$		
						71.00	5.51	6.96
						69.87	5.32	6.74
4e	69	153–155	3437 (O-H), 3026 (Ar-CH), 1729 (C=O), 1522 (C=C)	6.63–7.32 (m, 8H, Ar-H), 6.38 (s, 1H, =CH), 5.87 (s, 1H, H-5), 9.94 (s, 1H, Ar-OH), 1.34–2.33 (m, 8H, 4 × CH_2)	406 [M+] (52)	$C_{23}H_{19}FN_2O_2S$		
						67.96	4.71	6.89
						67.97	4.73	6.87
4f	65	157–159	3431 (O-H), 3021 (Ar-CH), 1722 (C=O), 1527 (C=C), 816 (C-Cl)	6.61–7.32 (m, 8H, Ar-H), 6.32 (s, 1H, =CH), 5.89 (s, 1H, H-5), 9.97 (s, 1H, Ar-OH), 1.32–2.37 (m, 8H, 4 × CH_2)	424 [M+2] (73)	$C_{23}H_{19}ClN_2O_2S$		
						65.32	4.53	6.62
						65.44	4.41	6.67
4g	75	184–186	3447 (O-H), 3025 (Ar-CH), 1716 (C=O), 1523 (C=C), 823 (C-Br)	6.73–7.29 (m, 8H, Ar-H), 6.48 (s, 1H, =CH), 5.73 (s, 1H, H-5), 9.89 (s, 1H, Ar-OH), 1.26–2.32 (m, 8H, 4 × CH_2)	468 [M+2] (91)	$C_{23}H_{19}BrN_2O_2S$		
						59.11	4.10	5.99
						59.14	4.13	5.97
4h	81	185–187	3452 (O-H), 3059 (Ar-CH), 1727 (C=O), 1439 (C=C)	6.82–7.75 (m, 6H, Ar-H), 6.71 (s, 1H, =CH), 5.73 (s, 1H, H-5), 9.84 (s, 1H, Ar-OH), 2.23 (s, 9H $-OCH_3$), 1.64–2.38 (m, 8H, 4 × CH_2)	478 [M+] (100)	$C_{26}H_{26}N_2O_5S$		
						65.25	5.48	5.85
						65.29	5.32	5.89

						$C_{25}H_{24}N_2O_2S$		
4i	79	181–183	3450 (O-H), 3051 (Ar-CH), 1724 (C=O), 1437 (C=C)	6.89–7.76 (m, 7H, Ar-H), 6.74 (s, 1H, =CH), 5.78 (s, 1H, H-5), 9.84 (s, 1H, Ar-OH), 2.23 (s, 6H -CH_3), 1.62–2.32 (m, 8H, 4 × CH_2)	416 [M+] (09)	72.09 72.12	5.81 5.79	6.73 6.75
						$C_{23}H_{19}N_3O_4S$		
4j	79	184–186	3441 (O-H), 3042 (Ar-CH), 1723 (C=O), 1435 (C=C)	6.82–7.71 (m, 8H, Ar-H), 6.77 (s, 1H, =CH), 5.72 (s, 1H, H-5), 9.71 (s, 1H, Ar-OH), 1.64–2.36 (m, 8H, 4 × CH_2)	433 [M+] (74)	63.73 63.81	4.42 4.54	9.69 9.52
						$C_{29}H_{26}N_4O_3S$		
5a	78	157–159	3461 (O-H), 3029 (Ar-CH), 1492 (C=C) 1316 (N-H bending), 3391 (N-H stretching)	6.74–7.13 (m, 13H, Ar-H), 6.32 (s, 1H, =CH), 5.59 (s, 1H, H-5), 9.81 (s, 1H, Ar-OH), 4.42 (s, 1H, thiazole), 7.26 (s, 1H, N-H), 1.46–2.42 (m, 8H, 4 × CH_2)	510 [M+] (36)	68.21 68.26	5.13 5.19	10.97 10.82
						$C_{29}H_{26}N_4O_4S$		
5b	72	151–153	3467 (O-H), 3021 (Ar-CH), 1497 (C=C) 1312 (N-H bending), 3391 (N-H stretching)	6.74–7.29 (m, 12H, Ar-H), 6.36 (s, 1H, =CH),5.62 (s, 1H, H-5), 9.87 (s, 2H, Ar-OH), 4.46 (s, 1H, thiazole), 7.23 (s, 1H, N-H), 1.46–2.42 (m, 8H, 4 × CH_2)	526 [M+] (21)	66.14 66.22	4.98 4.88	10.64 10.69
						$C_{30}H_{28}N_4O_4S$		
5c	76	156–158	3464 (O-H), 3027 (Ar-CH), 1494 (C=C) 1306 (N-H bending), 3396 (N-H stretching)	6.72–7.23 (m, 12H, Ar-H), 6.36 (s, 1H, =CH),5.62 (s, 1H, H-5), 9.87 (s, 1H, Ar-OH), 4.46 (s, 1H, thiazole), 3.78 (s, 3H -OCH_3), 7.29 (s, 1H, N-H), 1.46–2.42 (m, 8H, 4 × CH_2)	540 [M+] (76)	66.65 66.67	5.22 5.25	10.36 10.38
						$C_{30}H_{28}N_4O_3S$		
5d	76	192–193	3438 (O-H), 3024 (Ar-CH), 1412 (C=C) 1322 (N-H bending), 3310 (N-H stretching)	6.69–7.24 (m, 12H, Ar-H), 6.28 (s, 1H, =CH), 5.72 (s, 1H, H-5), 9.82 (s, 1H, Ar-OH), 4.45 (s, 1H, thiazole), 2.28 (s, 3H, -CH_3), 7.69 (s, 1H, N-H), 1.36–2.41 (m, 8H, 4 × CH_2)	524 [M+] (82)	68.68 68.65	5.38 5.36	10.68 10.70
						$C_{29}H_{25}FN_4O_3S$		
5e	89	184–186	3449 (O-H), 3026 (Ar-CH), 1524 (C=C), 1316 (N-H bending), 3319 (N-H stretching), 821 (C-F)	6.74–7.32 (m, 12H, Ar-H), 6.23 (s, 1H, =CH), 5.84 (s, 1H, H-5), 9.96 (s, 1H, Ar-OH), 4.42 (s, 1H, thiazole), 7.34 (s,1H, N-H), 1.24–2.32 (m, 8H, 4 × CH_2)	528 [M+] (100)	65.89 65.91	4.77 4.79	10.60 10.62
						$C_{29}H_{25}ClN_4O_3S$		
5f	72	164–166	3445 (O-H), 3025 (Ar-CH), 1523 (C=C), 1315 (N-H bending), 3320 (N-H stretching), 829 (C-Cl)	6.71–7.35 (m, 12H, Ar-H), 6.23 (s, 1H, =CH), 5.84 (s, 1H, H-5), 9.96 (s, 1H, Ar-OH), 4.42 (s, 1H, thiazole), 7.16 (s, 1H, N-H), 1.24–2.32 (m, 8H, 4 × CH_2)	547 [M+2] (41)	63.90 63.84	4.62 4.67	10.28 10.30

						$C_{29}H_{25}BrN_4O_3S$		
						59.09	4.27	9.50
5g	82	183–185	3447 (O-H), 3021 (Ar-CH), 1519 (C=C), 1327 (N-H bending), 3319 (N-H stretching), 818 (C-Br)	6.81–7.36 (m, 12H, Ar-H), 6.49 (s, 1H, =CH), 5.84 (s, 1H, H-5), 9.81 (s, 1H, Ar-OH), 4.46 (s, 1H, thiazole), 7.79 (s, 1H, N-H), 1.29–2.34 (m, 8H, 4 × CH_2)	590 [M+2] (53)	59.11	4.29	9.57
						$C_{32}H_{32}N_4O_6S$		
						63.98	5.37	9.33
5h	71	187–189	3429 (O-H), 3027 (Ar-CH), 1413 (C=C), 1334 (N-H bending), 3311 (N-H stretching)	6.72–7.21 (m, 10H, Ar-H), 6.27 (s, 1H, =CH), 5.72 (s, 1H, H-5), 9.91 (s, 1H, Ar-OH), 4.32 (s, 1H, thiazole), 3.32 (s, 9H, -OCH_3), 7.67 (s, 1H, N-H), 1.34–2.46 (m, 8H, 4 × CH_2)	600 [M+] (69)	63.81	5.39	9.37
						$C_{31}H_{31}N_4O_3S$		
						69.12	5.61	10.40
5i	77	181–183	3429 (O-H), 3027 (Ar-CH), 1413 (C=C), 1334 (N-H bending), 3313 (N-H stretching)	6.79–7.24 (m, 12H, Ar-H), 6.26 (s, 1H, =CH), 5.74 (s, 1H, H-5), 9.93 (s, 1H, Ar-OH), 4.39 (s, 1H, thiazole), 2.34 (s, 6H, -CH_3), 7.42 (s, 1H, N-H), 1.36–2.41 (m, 8H, 4 × CH_2)	538 [M+] (08)	69.14	5.63	10.43
						$C_{29}H_{25}N_5O_5S$		
						62.69	4.54	12.60
5j	71	152–154	3461 (O-H), 3021 (Ar-CH), 1493 (C=C), 1309 (N-H bending), 3392 (N-H stretching)	6.73–7.21 (m, 12H, Ar-H), 6.33 (s, 1H, =CH), 5.67 (s, 1H, H-5), 9.81 (s, 1H, Ar-OH), 4.46 (s, 1H, thiazole), 7.19 (s, 1H, N-H), 1.46–2.42 (m, 8H, 4 × CH_2)	555 [M+] (89)	62.62	4.51	12.67

Syntheses

The target novel thiazoloquinazoline derivatives were synthesized by the previously reported method (17). The key intermediate and target compounds are illustrated in Scheme 1.

*2-Hydroxybenzylidine cyclohexanone (**1**).* – Equimolar quantities of each cyclohexanone (0.039 mol) and salicylaldehyde (0.039 mol) were taken in a beaker; to this, sodium hydroxide solution was added to make the solution alkaline. This was shaken and kept aside. The solid thus obtained was filtered, washed with water and recrystallized from absolute ethanol.

*4-Hydroxy phenyl 3,4,5,6,7,8-hexahydroquinazolin-2-thione (**2**).* – A mixture of 2-hydroxybenzylidine cyclohexanone (**1**) (0.039 mol), thiourea (0.03 mol) and potassium hydroxide (2.5 g) in 95 % ethanol (100 mL) was heated under reflux for 3 h. The reaction mixture was concentrated to half its volume, diluted with water, then acidified with glacial acetic acid and kept overnight. The solid thus obtained was filtered, washed with water and recrystallized from ethanol to give **2**.

*6,7,8,9-Tetrahydro-5*H*-5-(2′-hydroxyphenyl)thiazolo(2,3-*b*)quinazolin-3(*2H*)-one* (***3***). – Chloroacetic acid (0.096 mol) was melted on a water bath and **2** (0.009 mol) was added portionwise to maintain homogeneity. The homogeneous mixture was heated on a water bath for 30 min and kept overnight. The solid thus obtained was washed with water and recrystallized from ethanol to give **3**.

*6,7,8,9-Tetrahydro-5*H*-5-(2′-hydroxyphenyl)-2-(4′-substituted benzylidine)thiazolo(2,3-*b*) quinazolin-3(*2H*)-ones* (***4a-j***). – A mixture of **3** (0.002 mol), substituted benzaldehyde (0.002 mol) and anhydrous sodium acetate (0.002 mol) in glacial acetic acid (10 mL) was heated under reflux for 4 h. The reaction mixture was kept overnight and the solid thus separated was filtered, washed with water and recrystallized from ethanol to furnish 6,7,8,9-tetrahydro-5*H*-5-(2′-hydroxyphenyl)-2-(4′-substituted benzylidine) thiazolo (2,3-*b*)quinazolin-3(2*H*)-ones (**4a-j**).

*6,7,8,9-Tetrahydro-5*H*-5-(2′-hydroxyphenyl)-2-(4′-substituted benzylidine)-3-(4-nitrophenylamino)thiazoloquinazolines* (***5a-j***). – A reaction mixture of the respective quinazolin-3(2*H*)-one **4a-j** (0.004 mol) was dissolved in (0.004 mol) thionyl chloride. To this, DMF (0.004 mol) was added to get chloro derivates. The reaction mixture was then coupled with *p*-nitroanilines (0.004 mol) in DMF at 80 °C for 3 h. After the mixture was cooled, the precipitate was filtered, recrystallized from ethanol, and dried to give the target compounds 6,7,8,9--tetrahydro-*5H*-5-(2′-hydroxyphenyl)-2-(4′-substituted benzylidine)-3-(4-nitrophenylamino)thiazoloquinazolines (**5a-j**).

PDE inhibition assay

Inhibition of bovine heart phosphodiesterase was determined by the known method (2).

PDE activity was calculated by measuring the production of inorganic phosphates in the presence of an excess of 5-nucleotidase. Each sample contained 1 mmol L^{-1} cAMP, 3 mmol L^{-1} $MgSO_4$, 5-nucleotidase (0.02 units mL^{-1}) and bovine heart phosphodiesterase (0.002 units mL^{-1}), and appropriate concentration of the test compound in ethanol and Tris-HCl buffer (pH 7.5, 50 mmol L^{-1}), in a final volume of 1 mL. The reagents comprising 0.4 mL acid molybdate solution, 0.4 mL Elon reducing agent, and 1.0 mL H_2O, to give a final volume of 3.0 mL, were mixed in an incubation vessel, and the reaction was initiated by addition of cAMP and allowed to proceed for 20 min over a 30 °C water bath. The reaction was terminated by addition of 0.2 mL 5 % trichloroacetic acid. The solution was then vortexed and centrifuged at 800×*g* for 10 min. Color was allowed to develop for 20 min and absorbance was measured at 660 nm. The results given in Table II are expressed as the concentration of inhibitor giving 50 % inhibition (IC_{50}) of the cAMP--dependent PDE activity. The IC_{50} value was determined from plot of percentage inhibition *vs.* varying concentration of the inhibitor compounds **4a-j** and **5a-j**. To validate the enzyme assay method, the IC_{50} value of theophylline as a standard inhibitor was also determined.

RESULTS AND DISCUSSION

Chemistry

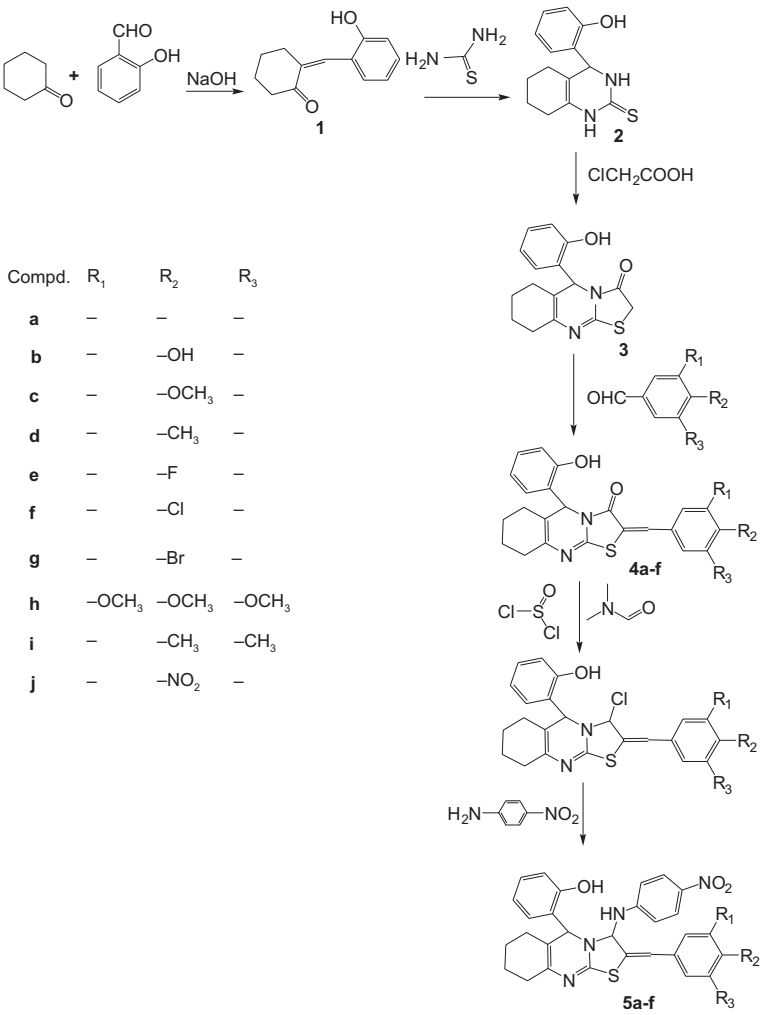
The series of heterocycles **4a-j** and **5a-j** were synthesized by the reaction of **3** with appropriate aromatic aldehyde and *p*-nitroaniline in the presence of anhydrous sodium acetate and DMF as it is presented in Scheme 1. The IR, ^{1}H NMR, mass spectroscopy and elemental analyses for the new compounds were in accord with the assigned structures. The IR spectra of compounds **4a-j** showed stretching bands of the keto group at 1715–1740 cm^{-1}. In **5a-j**, stretching and bending NH bands of thiazoloquinazoline moiety appear at 3300–3400 and 1300–1350 cm^{-1}, respectively. The recorded IR spectra of compounds **5a-j** showed no keto group bands. This clearly envisages that the keto group of **4a-j** is converted into secondary NH. In **5a-j**, the NH signal of 3-(4-nitrophenyl)amino-thiazoloquinazoline moiety appear at δ 7.26 (s), 7.23 (s), 7.29 (s), 7.69 (s), 7.34 (s), 7.16 (s),

Table II. PDE inhibition and relative data of synthesized compounds

Compd.	IC_{50} (mmol L^{-1})	Relative activity
4a	2.10 ± 0.09	1.22
4b	2.40 ± 0.02	1.35
4c	2.33 ± 0.09	1.34
4d	2.28 ± 0.02	1.27
4e	2.09 ± 0.08	1.22
4f	2.11 ± 0.08	1.21
4g	2.13 ± 0.08	1.28
4h	2.32 ± 0.08	1.32
4i	2.41 ± 0.03	1.35
4j	2.64 ± 0.02	1.47
5a	1.98 ± 0.03	1.09
5b	2.12 ± 0.04	1.20
5c	2.09 ± 0.05	1.18
5d	2.03 ± 0.04	1.14
5e	1.44 ± 0.02	0.87
5f	1.34 ± 0.09	0.84
5g	1.64 ± 0.04	0.93
5h	2.07 ± 0.06	1.18
5i	2.07 ± 0.03	1.17
5j	1.52 ± 0.05	0.87
Theophylline	1.72 ± 0.09	1

Solvent used: DMSO
IC_{50} values were determined as described in the experimental section. Mean ± SD, n = 3.

7.79 (s), 7.67 (s), 7.42 (s) and 7.19 (s) ppm, respectively. The presence and position of NH signal in the ^{1}H NMR spectra of final compounds conform with the secondary NH proton in thiazoloquinazoline moiety. This clearly envisages that thiazole-3-one moiety was involved in 3-(4-nitrophenyl)amino formation. Mass spectra showed accurate molecular ion peaks at m/z 300, 478 and 528 for **3**, **4h** and **5e**, resp. All these facts clearly demonstrated that the 3rd position of keto group in thiazole ring was converted into secondary amino group, as indicated in Sheme 1 and conforming with the proposed structures **5a-j**.



Compd.	R_1	R_2	R_3
a	–	–	–
b	–	–OH	–
c	–	$-OCH_3$	–
d	–	$-CH_3$	–
e	–	–F	–
f	–	–Cl	–
g	–	–Br	–
h	$-OCH_3$	$-OCH_3$	$-OCH_3$
i	–	$-CH_3$	$-CH_3$
j	–	$-NO_2$	–

Scheme 1

PDE inhibition activity

The target compounds were evaluated for their ability to inhibit bovine heart phosphodiesterase with theophylline chosen as a standard for comparison (Table II). All the compounds were found to exhibit PDE inhibitory activity. Despite the fact that this assay is less sensitive compared to radioisotope PDE assays and likely to underestimate the relative inhibitory potencies of the test compounds, the results of this evaluation indicate that the quinazoline derivatives are still more potent inhibitors of PDE than theophylline.

Compounds having thiazolo(2,3-*b*)quinazolin-3(2*H*)-one (**4a-j**) and thiazoloquinazoline (**5a-j**) ring systems were all found to exhibit promising PDE inhibitory activity with IC_{50} in the range 1.34–2.64 (±0.09) mmol L^{-1} and activity relative to theophylline of 0.84–1.47. Among them 6,7,8,9-tetrahydro-5*H*-5-(2′-hydroxyphenyl)-2-(4′-chlorobenzyline)-3-(4-nitrophenylamino)thiazoloquinazoline (**5f**) was found to be most potent, with relative activity of 0.8 compared to the standard, whereas for both compounds 6,7,8,9-tetrahydro-5*H*-5-(2′-hydroxyphenyl)-2-(4′-fluorobenzylidine)-3-(4-nitrophenylamino)thiazoloquinazoline (**5e**) and 6,7,8,9-tetrahydro-5*H*-5-(2′-hydroxyphenyl)-2-(4′-nitrobenzylidine)--3-(4-nitrophenylamino)thiazoloquinazoline (**5j**) relative reactivity was found to be 0.87.

The electronic nature of the substituent groups at second positions in thiazole nucleus led to significant variation in PDE inhibitory activity. In both series the presence of an electron-withdrawing group (chloro, fluoro, nitro, bromo) in the thiazoloquinazoline nucleus resulted in higher activity. For example, 4-nitrophenyl amino substitution in thiazoloquinazoline nucleus (**5a-j**) increased activity; the order of activity was Cl > F > NO_2 > Br > electron donating group. Compound **5f**, **5e**, **5j** and **5g** showed higher activity than the reference drug theophylline.

CONCLUSION

In summary, the present study has demonstrated that fused thiazoloquinazolines are novel inhibitors of phosphodiesterase and may potentially act as smooth muscle relaxatory agents. Further work is in progress to estabilish this observation in different animal models and find selective inhibitors of PDE 4, which may impact strategies for bronchodilator activity.

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S A Ž E T A K

Sinteza i cAMP-ovisna inhibicija fosfodiesteraze novih derivata tiazolokinazolina

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U radu je opisana sinteza serije 6,7,8,9-tetrahidro-5*H*-5-(2′-hidroksifenil)-2-(4′-supstituiranih benzilidin)tiazolo(2,3-*b*)kinazolin-3(2*H*)-ona (**4a-j**) i 6,7,8,9-tetrahidro-5*H*-5-(2′-hidroksifenil)-2-(4′-supstituiranih benzilidin)-3-(4-nitrofenilamino)tiazolokinazolina (**5a-j**) prema objavljenoj metodi te ispitano njihovo inhibitorno djelovanje na fosfodiesterazu. Svi testirani spojevi pokazuju dobro djelovanje. Proučavan je i odnos strukture i djelovanja. U obje serije spojeva, elektron-odvlačeći supstituenti doprinose jačem djelovanju. Među ispitivanim spojevima pronađeno je da 6,7,8,9-tetrahidro-5*H*-5-(2′-hidroksifenil)-2-(4′-fluorobenzilidine)-3-(4-nitrofenilamino)tiazolokinazolin (**5e**), 6,7,8,9-tetrahidro-5*H*-5-(2′-hidroksifenil)-2-(4′-nitrobenzilidine)-3-(4-nitrofenilamino)tiazolokinazolin (**5j**) i 6,7,8,9-tetrahidro-5*H*-5-(2′-hidroksifenil)-2-(4′-klorobenzilidin)-3-(4-nitrofenilamino)tiazolokinazolin (**5f**) imaju jače djelovanje od teofilina (IC_{50} u mmol L^{-1} 1,34 ± 0,09 za **5f**, 1,44 ± 0,02 za **5e**, 1,52 ± 0,05 za **5j** nasuprot 1,72 ± 0,09 za teofilin).

Ključne riječi: tiazolokinazolin, benzilidinetiazolokinazolin, nitrofenilaminotiazolokinazolin, inhibicija fosfodiesteraze, SAR

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