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SILYL MODIFICATION OF BIOLOGICALLY ACTIVE COMPOUNDS. 8*. TRIMETHYLSILYL ETHERS OF HYDROXYL-CONTAINING THIAZOLE DERIVATIVES

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Trimethylsilyl ethers of various hydroxyl-containing thiazole derivatives have been synthesized. The psychotropic activity (in vivo) and the cytotoxicity (in vitro on tumor cell lines HT-1080 and MG-22A) of these ethers and of their unsilylated precursors have been studied. It was discovered that the obtained compounds possess a sedative action. A moderate cytotoxic effect was detected for piperidine-containing thiazoles, displayed most strongly in relation to MG-22A cells.

Keywords: piperidine, silyl group, thiazole, psychotropic activity, silylation, cytotoxicity.

The role of the thiazole ring in biological processes is well known. A thiazole ring is the active part of thiamine (vitamin B_1) and thiamine pyrophosphate (cocarboxylase), which are coenzymes in the composition of certain enzymes (dehydrogenase and transkelase of γ-hydroxyketoglutarate) or are an important group for certain multienzymes (complexes of pyruvate dehydrogenase and α-ketoglutarate). Due to the effective influence on various functions of the organism and participation in metabolism and neuroregulation, thiamine displays a positive action on various pathological processes. Thiamine and its derivatives are therefore accepted pharmaceutical agents. In addition, compounds containing a thiazole ring possess psychotropic activity [2-7], and a series of bisthiazolium salts have the properties of neuromuscular blocking agents [8].

One of the problems of designing new drugs is the preparation of biologically active substances with increased lipophilicity, in order to provide penetration of the drug through the plasma membrane (lipid bilayer) and, for substances influencing the central nervous system, passage through the blood–brain barrier. A series of investigations has shown that silyl modification is one of the most effective means of solving this problem. Our investigations [1,9-14] also confirm the promise of such an approach. The O(N)-silyl derivatives are inclined towards chemical and metabolic hydrolysis with the formation of the initial compound, being so-called prodrugs in this respect. The expected byproducts of hydrolysis (the triorganosilanols and disiloxanes) are mainly nontoxic or weakly toxic compounds.

We have obtained trimethylsilyl ethers (**2,4,6,13-15**) of various hydroxyl-containing thiazoles (**1,3,5,10-12**) and also a C-silyl derivative, an organosilicon salt of 2-aminothiazole (**16**). The psychotropic activity and also the cytotoxicity of these compounds has been studied in comparison with their unsilylated precursors.

* For Part 7 see [1].

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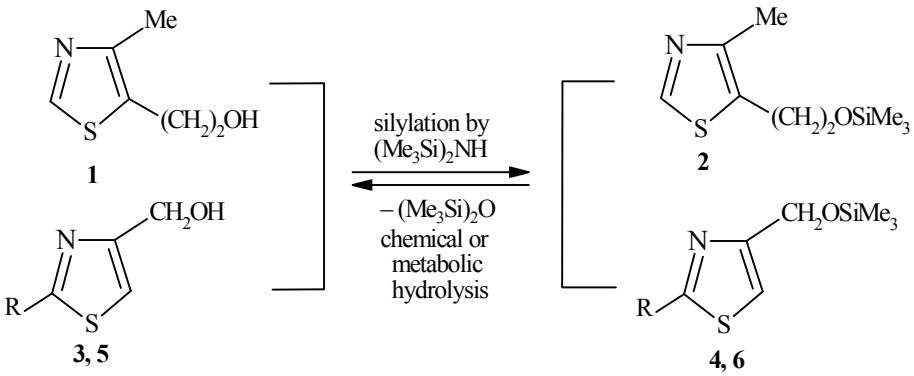
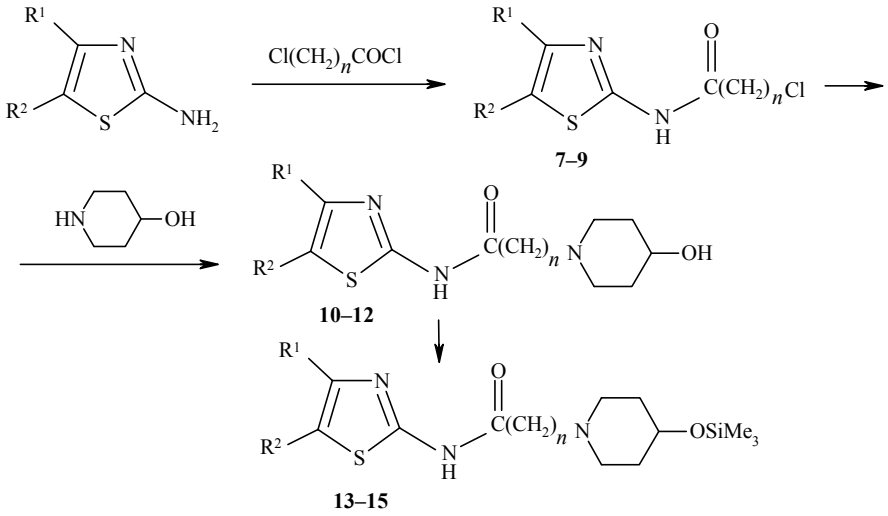
The organosilicon derivatives of 5-(β-hydroxyethyl)-4-methylthiazole **2**, and of 2-amino- and 4-hydroxymethyl-2-phenylthiazole **4** and **6** were synthesized by heating the initial thiazoles **1**, **3**, and **5** with hexamethyldisilazane for several hours.



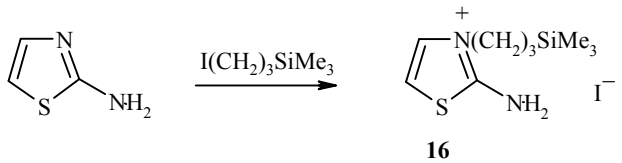
3, **4** R = NH_2; **5**, **6** R = Ph

The N-(2-thiazolyl)amides **10-12** were synthesized by the condensation of 2-aminothiazole (substituted or unsubstituted) with the appropriate acyl chloride (chloroacetic or chloropropionic acid chlorides) and then reaction of the obtained chloroalkylamide **7-9** with 4-hydroxypiperidine [15, 16]. Heating thiazoles **10-12** with hexamethyldisilazane leads to the formation of the trimethylsilyl ethers **13-15**.



7, 10, 13 $R^1 = R^2$ = H, *n* = 2; **8, 11, 14** R^1 = Ph, R^2 = H, *n* = 1; **9, 12, 15** R^1 = Ph, $R^2 = n\text{-}C_{14}H_{29}$, *n* = 1

The organosilicon salt **16** was obtained by the quaternization of 2-aminothiazole with 3-(trimethylsilyl)propyl iodide.



The neurotropic activity of the synthesized compounds was studied in a series of tests. Results of the study of the neurotropic properties and the acute toxicity are given in Tables 1 and 2.

TABLE 1. Neurotropic Activity of Hydroxyl-containing Thiazoles and Their Trimethylsilyl Ethers

Com-pound	M ± m, % control (100%)							
	Test							
	amphetamine hypothermia °C (30 min)	amphetamine hyperactivity (30 min)	hypoxic hypoxia	hexenal narcosis	ethanol narcosis	corazole spasms (clonic/tonic)	retrograde amnesia*[2]	instruction, sec*[3]
1	-0.2	16*	120*	16.9	117*	96/117*	100*	143.4* ± 7.8
2	-2.6*	17*	171*	94	256*	156*/137*	100*	140.8* ± 9.3
3	-2.4*	39*	152*	87	172*	146*/129*	60	95.2 ± 20.7
4	-2.0*	85	122*	87*	113	166*/140*	40*	85.8 ± 18.2
5	-0.8*	83*	146*	97	185*	128*/130*	40	51.5 ± 38.4
6	0.4	30*	141*	105	172*	145*/128*	100*	147.8* ± 7.1
10	-2.6*	81*	152*	121*	167*	146*/109	80*	137.0* ± 8.6
11	-3.4*	65*	137*	102	131*	137*/142*	20	59.8 ± 1.4
12	0.7	48*	178*	89*	238*	144*/184*	80*	128.2* ± 14.2
13	-0.4	137*	155*	150*	108	119/116	0	33.8 ± 21.9
14	-2.8	48*	162*	119	139*	140*/144*	40	87.4 ± 22.8
15	-1.2	99	170*	110	156*	149*/180*	80*	127.8* ± 8.7
16	-0.8	42*	124*	131*	89	149*/130*	100*	161.4* ± 1.8

* $P < 0.05$.

*[2] Control, %: 33.3.

*[3] Control, sec: 58.1±10.9.

TABLE 2. The Effect of Hydroxyl-containing Thiazoles and Their Trimethylsilyl Ethers on the Tone of the Skeletal Musculature and Movement Coordination

Compound	LD_{50}, mg/kg	ED_{50}, mg/kg					
		Test					
		rotating rod	tubes	rectal temperature	analgesia	narcosis	pulling on a beam
1	>1000	>250	>250	>250	>250	>250	>250
2	>250						
3	>1000	355 (249-461)	137 (50-262)	224 (144-285)	129 (61-202)	>250	346 (120-662)
4	>1000	>250	>250	65 (37-100)	51 (29-79)	>250	>250
5	>400	69 (24-130)	274 (99-524)	45 (31-60)	355 (249-461)	>250	274 (99-524)
6	>250	258 (108-357)	>250	141 (92-209)	>250	>250	>250
10	815 (567-1110)	87	>250	172 (73-332)	69 (24-130)	>250	>250
13	>250	>250	>250	258 (168-357)	>250	>250	>250
11	>1000	89 (63-120)	69 (24-130)	41 (27-55)	205 (146-288)	>250	87 (30-165)
14	>250	>250	>224 (144-285)	45 (31-60)	204 (144-285)	>250	>250
12	>500	>250	>250	69 (24-130)	141 (68-209)	>250	>250
15	>1000	69 (24-130)	>250	41 (27-55)	51 (36-69)	>250	>250
16	51.5 (36.2-69.2)	28.2 (18.3-37.2)	28.2 (18.3-37.2)	28.2 (18.3-37.2)	25.8 (16.8-35.7)	>25	28.2 (18.3-37.2)

All the compounds investigated possess antihypoxic properties and prolong the life of mice under conditions of hypoxia by 20-78%. The silylated and unsilylated compounds in the majority of cases display antihypoxic activity of the same order, in individual cases the activity grew by 25-51% (see **1** and **2**, **11** and **14**) on introducing the trimethylsilyl group.

The most active antihypoxic agents were the piperidine-containing thiazoles **12** and **15**, prolonging the life of mice by 78 and 70% respectively.

The action of the synthesized compounds at a dose of 5 mg/kg on the prolongation of hexenal narcosis varied. The piperidine-containing thiazole **10** prolonged narcosis by 21%, and its trimethylsilyl ether **13** by 50%. The prolongation of narcosis was reduced insignificantly under the influence of trimethylsilyl ether **4** and the piperidine-containing thiazole **12**.

In the case of ethanol narcosis the action of the synthesized compounds was more marked. They almost all prolonged the action of ethanol narcosis (by 17-156%), while the silylated and unsilylated compounds displayed activity of the same order (117-238 and 139-256% respectively). The most reliable prolongation of narcosis (by 129%) on introducing the trimethylsilyl substituent was noted in the case of 5-(β-hydroxyethyl)-4-methylthiazole (**1**) and its trimethylsilyl ether **2**.

Almost all of the compounds studied possess an antispasmic action on corazole spasms (clonic and tonic). They increased the threshold of corazole spasms by 28-66% in the tonic and 17-84% in the clonic phase. In several cases in this test the trimethylsilyl ethers were stronger anticonvulsants (see **1** and **2**, **3** and **4**, **5** and **6**, **11** and **14**). The strongest anticonvulsive action was possessed by the piperidine-containing thiazole **12** and its trimethylsilyl ether **15** (133/184 and 149/180% respectively).

The substances investigated did not display protective properties in the maximal electroshock test.

The majority of the compounds investigated act as amphetamine antagonists, reducing locomotor activity caused by the administration of amphetamine by 17-84%.

The greatest effect on the interaction with amphetamine was observed for compound **1** and its trimethylsilyl ether **2** which significantly reduced locomotor activity (by 85-84%) for 30 min. The most reliable strengthening of the antagonistic properties in relation to amphetamine on introducing a trimethylsilyl group (by 53%) was noted on comparing 4-hydroxymethyl-2-phenylthiazole **5** and its silyl ether **6**.

Study of the influence of the substances investigated on memory processes showed that for a series of compounds retrograde amnesia was prevented (100%), and also the latent period of instruction was increased. The most active on these processes proved to be trimethyl ether **6**, which at a dose of 5 mg/kg completely prevented retrograde amnesia.

All of the compounds had almost no effect on the tone of the skeletal musculature and movement coordination (Table 2).

Hypothermic action, like the analgesic action, was also expressed extremely weakly in the compounds studied.

According to the results of investigating the neurotropic activity of 2-amino-3-(γ-trimethylsilyl)propylthiazolium iodide (**16**), this organosilicon salt shows neurotropic action in several tests of the same order as the remaining compounds. Its greatest effect might be expected on memory processes.

The cytotoxic ptoperties (*in vitro*) of the compounds synthesized were also studied in relation to two lines of tumor cells, HT-1080 (human lung fibrosarcoma) and MG-22A (mouse hepatoma) (see Table 3).

A large proportion of the compounds studied, mainly the piperidine-containing derivatives **10-12**, **13-15**, possess low cytotoxicity in relation to cell line HT-1080 and moderate in relation to MG-22A. Compounds **1-4** were not cytotoxic, and the cytotoxic effect of compounds **5** and **6** was low. The strongest cytotoxic action on tumor cells (MG-22A) was possessed by the piperidine derivative of thiazole **11** and its trimethylsilyl ether **14**. The introduction of the trimethylsilyl group was clearly followed by a strengthening of the cytotoxic effect in both tests. The organosilicon salt **16** displayed the greatest activity in relation to HT-1080 cells among all the compounds studied.

TABLE 3. Cytotoxic Activity *in vitro* TD_{50} and NO of Hydroxyl- and Silicon-containing Thiazoles*

Compound	Cell line					
	HT 1080			MG 22-A		
	TD_{50}, μg/ml		NO, %	TD_{50}, μg/ml		NO, %
	CV	MTT	CV	CV	MTT	CV
1	No activity		4	No activity		4
2	No activity		5	No activity		9
3	No activity		3	No activity		6
4	No activity		5	No activity		6
5	76	75	9	100	No activity	6
6	No activity		4	69	80	10
10	48	48	200	35	27	200
11	59	84	27	15	24	300
12	34	61	350	8	57	350
13	44	56	250	44	45	200
14	51	65	40	5.3	12	150
15	77	76	250	37	59	200
16	22	14	275	46	23	550

* TD_{50} is the concentration causing 50% death of cells; CV is staining with crystal violet; MTT is staining with 3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyltetrazolium bromide.

The greatest level of NO generation was detected for the hydroxypiperidine-containing thiazoles **11** (up to 300% on line MG-22A) and **12** (up to 350% on both lines), and also for the organosilicon salt **16**, particularly on line MG-22A (up to 550%).

It was established from the investigations carried out that all the substances synthesized possess a sedative action. The greatest effect was noted in the tests of hypoxic hypoxia, ethanol narcosis, corazole spasm, and in tests reflecting the effect of substances on memory processes. The compounds studied are amphetamine antagonists. The greatest depriming type of activity was displayed by (hydroxyethyl)-thiazole **1** and its trimethylsilyl ether **2**, and also by the trimethylsilyl ether of 4-hydroxymethyl-2-phenylthiazole (**6**).

In several cases an increase of activity was observed on introducing the trimethylsilyl substituent into the molecule of thiazole derivative, although on the whole the trimethylsilyl ethers of thiazoles and their unsilylated precursors displayed psychotropic activity of the same order. The greatest cytotoxic effect on MG-22A cells was observed in the case of trimethylsilyl ether **14**.

EXPERIMENTAL

The ^{1}H NMR spectra were taken on a Varian Mercury 200 (200 MHz) spectrometer in $CDCl_3$, internal standard for the unsilylated compounds was hexamethyldisiloxane (HMDS), the organosilicon compounds were measured relative to the signal of the solvent (7.25 ppm). Elemental analyses were carried out with a Carlo Erba 1108 analyzer. The GLC analysis was effected on a Chrom-5 (Czech Republic) chromatograph with a flame ionization detector. A glass column (1.2 m × 3 mm) was used packed with 5% OV-17 on Chromosorb W-HP (80-100 mesh) as carrier.

4-Methyl-5-(β-trimethylsilyloxyethyl)thiazole (2). A mixture of 5-(β-hydroxyethyl)-4-methylthiazole **1** (715 mg, 5 mmol) and hexamethyldisilazane (7 ml) was heated with stirring for several hours. The progress of the reaction was followed by GLC. At the end of the reaction the excess of hexamethyldisilazane was removed

in vacuum on a rotary evaporator. The residue was purified on a chromatographic column (support was Acros silica gel 0.060-0.0200 mm, pore diameter 6 nm, eluent benzene). Compound **2** (570 mg, 53%) was obtained. ^{1}H NMR spectrum, δ, ppm: 8.56 (1H, d, 2-H); 3.73 (2H, t, CH_2O); 2.96 (2H, t, CH_2C); 2.39 (3H, s, CH_3); 0.08 [9H, s, $Si(CH_3)_3$]. Found, %: C 49.97; H 7.90; N 6.42; S 14.55. $C_9H_{17}NOSSi$. Calculated, %: C 50.16; H 7.95; N 6.50; S 14.88.

2-Amino-4-(trimethylsilyloxymethyl)thiazole (4) was obtained by the procedure described above from 2-amino-4-hydroxymethylthiazole **3** (650 mg, 5 mmol) in 54% yield (545 mg); mp 74-75°C. ^{1}H NMR spectrum. δ, ppm: 6.38 (1H, s, 5-H); 4.98 (2H, br s, NH_2); 4.55 (2H, s, CH_2); 0.15 [9H, s, $Si(CH_3)_3$]. Found. %: C 41.87; H 7.03; N 13.99; S 15.86. $C_7H_{14}N_2OSSi$. Calculated, %: C 41.56; H 6.97; N 13.84; S 15.85.

2-Phenyl-4-trimethylsilyloxymethylthiazole (6) was obtained analogously from 4-hydroxymethyl-2-phenylthiazole **5** (764 mg, 4 mmol) in 49% yield (515 mg); mp 74-75°C. ^{1}H NMR spectrum, δ, ppm: 7.86-7.97 and 7.36-7.46 (5H, m, C_6H_5); 7.17 (1H, d, 5-H); 4.85 (2H, d, CH_2); 0.19 [9H, s, $Si(CH_3)_3$]. Found, %: C 60.03; H 6.38; N 5.22; S 12.52. $C_{13}H_{17}NOSSi$. Calculated, %: C 59.27; H 6.50; N 5.32; S 12.17.

N-(2-Thiazolyl)-3-chloropropionamide (7), N-(4-Phenyl-2-thiazolyl)-2-chloroacetamide (8), and N-(4-Phenyl-5-tetradecyl-2-thiazolyl)-2-chloroacetamide (9) were obtained by the procedure given in [15].

N-(2-Thiazolyl)-3-(4-hydroxypiperidino)propionamide (10), N-(4-Phenyl-2-thiazolyl)-2-(4-hydroxypiperidino)acetamide (11), and N-(4-Phenyl-5-tetradecyl-2-thiazolyl)-2-(4-hydroxypiperidino)-acetamide (12) were synthesized by the procedure given in [15,16].

N-(2-Thiazolyl)-3-(4-trimethylsilyloxypiperidino)propionamide (13) was obtained by the procedure given above from compound **10** (765 mg, 3 mmol) in 49% yield (480 mg); mp 87°C. ^{1}H NMR spectrum, δ, ppm: 12.80 (1H, s, NH); 7.42 (1H, d, 4-H); 6.91 (1H, d, 5-H); 3.80 (1H, m, $CH_{cycl}O$); 2.85, 2.38, and 1.81 (8H, m + t + t, CH_{2cycl}); 2.71 and 2.58 (4H, t + t, CCH_2N + $COCH_2C$); 0.12 [9H, s, $Si(CH_3)_3$]. Found, %: C 51.62; H 7.73; N 12.85; S 9.79. $C_{14}H_{25}N_3O_2SSi$. Calculated, %: C 51.34; H 7.69; N 12.83; S 9.79.

N-(4-Phenyl-2-thiazolyl)-2-(4-trimethylsilyloxypiperidino)acetamide (14) was obtained by an analogous method from compound **11** (792 mg, 2.5 mmol) in 51% yield (492 mg); mp 115-118°C. ^{1}H NMR spectrum, δ, ppm: 7.27-7.90 (5H, m, C_6H_5); 7.15 (1H, s, 5-H); 3.75 (1H, m, $CH_{cycl}O$); 3.23 (2H, s, $COCH_2N$); 2.82, 2.42, and 1.75 (4H + 2H + 2H, m + m + m, (CH_{2cycl}); 0.12, [9H, m, $Si(CH_3)_2$]. Found, %: C 58.71; H 7.08; N 10.39; S 8.09. $C_{19}H_{27}N_3O_2SSi$. Calculated, %: C 58.58; H 6.99; N 10.79; S 8.22.

N-(5-Tetradecyl-4-phenyl-2-thiazolyl)-2-(4-trimethylsilyloxypiperidino)acetamide (15) was obtained analogously from compound **12** (770 mg, 1.5 mmol) in 45% yield (395 mg); mp 77-80°C. ^{1}H NMR spectrum, δ, ppm: 7.28-7.60 (5H, m, C_6H_5); 3.76 (1H, m, $CH_{cycl}OSi$); 3.21 (2H, d, $COCH_2N$); 2.85, 2.39, and 1.92 (4H + 2H +2H, m + m + m, CH_{2cycl}); 1.65 and 1.23 (4H + 22H, m + m, CH_2); 0.90 (3H, s, CH_3); 0.12 [9H, s, $Si(CH_3)_3$]. Found, %: C 67.68; H 9.39; N 7.19; S 5.40. $C_{33}H_{55}N_3O_2SSi$. Calculated, %: C 67.64; H 9.46; N 7.17, S 5.47.

2-Amino-3-(γ-trimethylsilylpropyl)thiazolium Iodide (16). A mixture of 2-aminothiazole (325 mg, 3.25 mmol), 3-iodopropyltrimethylsilane (784 mg, 3.24 mmol), and acetonitrile (2 ml) was heated at ~45°C for 10 h. The solution obtained was filtered, and evaporated to an oil. The oil was triturated with absolute ether until a bright yellow solid was obtained. The yield of compound **16** was 642 mg (58%), mp 66-69°C. ^{1}H NMR spectrum, δ, ppm: 9.07 (2H, s, NH_2); 6.85 (1H, d, 4-H); 6.77 (1H, d, 5-H); 4.30 (2H, t, N^+CH_2); 1.79 (2H, m, CCH_2C); 0.60 (2H, m, CCH_2Si); -0.02 [9H, s, $Si(CH_3)_3$]. Found, %: C 31.18; H 5.39; N 8.19; S 9.30. $C_9H_{19}IN_2SSi$. Calculated, %: C 31.58; H 5.59; N 8.18; S 9.37.

Biological Section. Neurotropic activity was studied in BALB/c strain mice and random-bred male rats. An oil solution of the substance being studied was introduced intraperitoneally 30 min before beginning the test.

The action of a substance on the central nervous system was assessed by 1) the effect on movement coordination and muscular tone (tests were "rotating rod", "tubes", "pulling on a beam"), 2) body temperature, 3) analgesic effect (test "hot plate"), 4) antispasmic activity (tests were maximal electroshock and corazole

spasm), 5) duration of hexenal and ethanol narcosis, 6) lifespan under conditions of hypoxic hypoxia, 7) locomotor activity and body temperature under joint action of amphetamine, 8) unavoidable stress situation and influence on processes of memory and retrograde amnesia.

The experimental data were processed statistically. Mean values of LD_{50} and ED_{50} were found from 12-20 observations using the express method of [17]. Assessment of the significance of differences between mean values (M + m) was carried out by the Student criteria. Differences were considered significant at a probability level of $P \leq 0.05$.

Investigation of cytotoxicity on tumor cell lines HT-1080 and MG-22A were carried out in 96-well panels [18,19]. Optical density in the biological tests was determined with a Tetertek Multiscan MCC/340 horizontal spectrophotometer.

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