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Bis-Quaternary Derivatives of 3-Azabicyclo [3. 2. 1] octanes *

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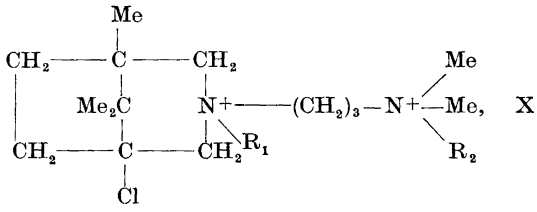
Some N-substituted chlorinated camphidine derivatives containing substituted ammonium groups in the side chain were found to have strong and prolonged ganglion blocking and antihypertensive effects. The structural requirements for this were studied. A peculiar discrepancy in potency towards different ganglion containing biological preparations is reported.

Several years ago evidence was presented in a series of papers from this laboratory that non-symmetric bis-quaternary ammonium salts are generally more potent ganglionic blocking agents than the corresponding symmetric analogues.[1,2] In pursuance of this theme a study was undertaken of bis-quaternary ammonium salts with a greater degree of non-symmetry, *e.g.* compounds in which one nitrogen was part of a rather bulky heterocyclic system while the other was substituted with aliphatic groups. At about the same time other groups independently found similar compounds with very strong ganglion blocking activity.[3–5]

The work to be reported here deals with the preparation and properties of bis-quaternaries in which one nitrogen is part of a chlorine substituted camphidine ring as an example of a large, non-planar heterocyclic system, together with the bis-tertiary precursor (III) and some mono-quaternary salts. (Mono-quaternization is supposed to take place at the terminal N-atom, see below).

The compounds are represented by the formula



* Terpene Amines I.

Table 1. Salts of the N-(γ-trimethylammoniumpropyl)-5-chloro-methylcamphidinium ion.

Salt	M.p. °C	Recryst. solvent	Yield %	Formula	Carbon %		Hydrogen %		Nitrogen %		Chlorine %	
					calc.	found	calc.	found	calc.	found	calc.	found
Hydrogen sulfate (VII)	242 – 244	EtOH-ether	50	$C_{17}H_{37}ClN_2O_8S_2$	41.1	41.6	7.5	7.7	5.6	5.4	7.1	7.3
Tartrate (VIII)	168 – 171	iPrOH-water	38	$C_{25}H_{45}ClN_2O_{12}$	49.9	50.0	7.5	7.8	5.9	6.0	4.7	4.9
Fumarate (IX)	96 – 113	EtOH-ether	32	$C_{25}H_{41}ClN_2O_8$	56.3	56.7	7.8	7.5	5.3	4.8	6.7	6.5
Maleate (X)	175 – 177	EtOH-ether	22	$C_{25}H_{41}ClN_2O_8$	56.3	56.3	8.0	7.8	5.3	5.3	6.7	6.8
Phosphate (XI)	223 – 224	MeOH – Me_2CO	34	$C_{17}H_{39}ClN_2O_8P_2$	41.0	41.0	7.9	7.9	5.8	5.6	7.1	7.4
Bromide (XII)	209 – 210	EtOH-ether	52	$C_{17}H_{35}Br_2ClN_2$	44.1	44.0	7.6	7.7	6.1	6.1	7.7	7.4

Bis-quaternary salts. (See Experimental Part and Table 1). $R_1 = R_2 =$ Me. Compounds IV, V, VIII–XII.

Mono-quaternary salts. (See Table 2). $R_1 =$ H. $R_2 =$ Me, propyl, amyl or lauryl. Compounds XIII–XV, XXI, XXII. In compounds XVI–XVIII there is no salt formation at the ring N-atom.

Similar compounds without halogen in the ring have been reported.[6] The substance corresponding to IV (but without Cl) was included as a pharmacological reference and is designated as XXIV.

The bis-quaternary salts of the chlorocamphidine are strong ganglion blocking agents. In spite of their ionic character they are readily absorbed from the gastrointestinal tract and are capable of lowering the blood pressure in normotensive cats as well as in renal hypertensive rats independent of the route of administration.

Experiments on the anaesthetized cat suggested that the hypotensive effect was not entirely due to interference with peripheral synaptic transmission, but that some other mechanism might be involved. It was therefore speculated that the bis-tertiary precursor or a mono-quaternary amine might retain a hypotensive effect without influencing synaptic transmission. However, the experimental evidence failed to support this hypothesis.

N-(γ-Dimethylaminopropyl)-5-chlorocamphidine di-methiodide (IV) was prepared from 5-chlorocamphorimide (I) by the following sequence of reactions. (I was obtained from *d*-camphoric acid according to the method described by Scheiber and Knothe [7]).

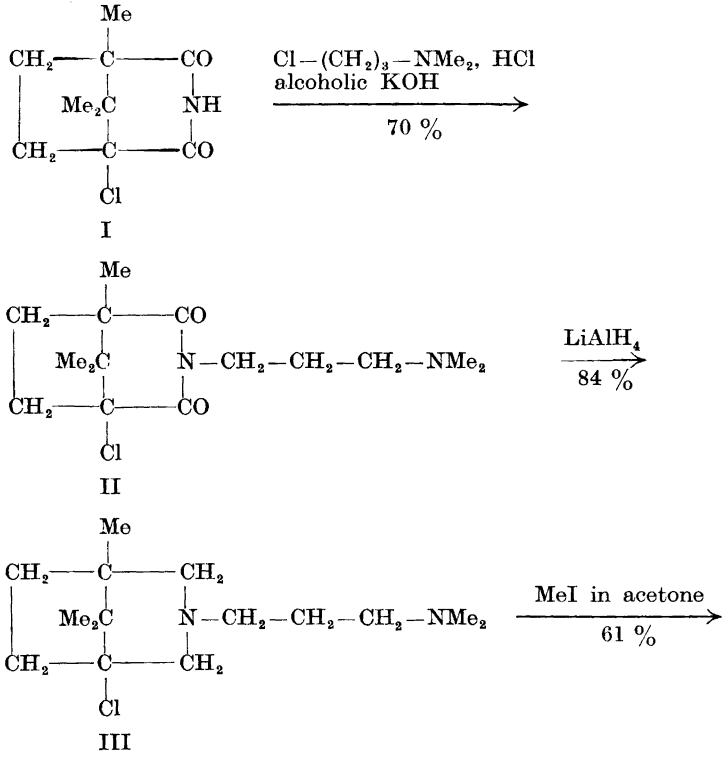
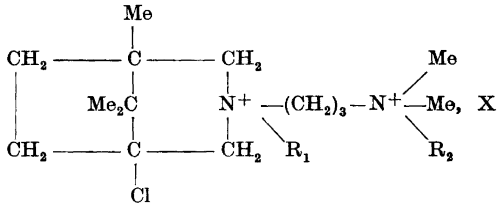
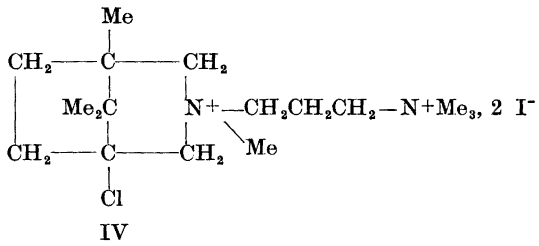


Table 2. Salts of mono-quaternary compounds



Compound	R_1	R_2	X	Method	M.p.	Recryst. solvent	Yield	Formula	Carbon %		Hydrogen %		Nitrogen %	
									calc.	found	calc.	found	calc.	found
XIII	H	Me	2 I^-	*a*	208–212	MeOH	34[b]	$C_{16}H_{33}ClI_2N_2$	35.3	36.0	6.1	6.3	5.2	5.0
XIV	H	Me	2 Br^-	*c*	234–235	EtOH	65[b]	$C_{16}H_{33}Br_2ClN_2$	42.8	42.8	7.4	7.5	6.3	6.3
XV	H	Me	COO^- $2(CHOH)_2$ COOH	*d*	no definite m.p.	water-iPrOH	76[e]	$C_{24}H_{43}ClN_2O_{12}$, $3H_2O$	45.0	45.4	7.7	7.8	4.4	4.6
XVI	Mono-tertiary amino	Me	Br^-	*f,g*	233–235[h]	Me/CO-ether	71[b], 79[e]	$C_{16}H_{32}BrClN_2$	52.2	52.2	8.8	9.0	7.6	7.6
XVII	—»—	Me	I^-	*i,j,k*	236–237[l]	Me_2CO	81[m], 73[n] 60[e]	$C_{16}H_{32}ClIN_2$[r]	46.3	45.0	7.8	7.8	6.8	6.7
XVIII	—»—	n-Pr	I^-	*o*	192–194	Me_2CO	56[b]	$C_{18}H_{36}N_2ClI$[s]	48.8	48.9	8.2	8.3	6.3	6.5
XXI	H	n-amyl	2 I^-	*p*	214–216	EtOH	26[b]	$C_{20}H_{41}N_2ClI_2$	40.1	40.5	6.9	7.0	4.7	4.7
XXII	H	lauryl	2 I^-	*q*	219–220	EtOH	22[b]	$C_{27}H_{55}N_2ClI_2$	46.5	46.9	8.0	8.1	4.0	4.0

a) III was treated as described for the preparation of IV using acetone which had not been dried. *b*) From III. *c*) As method *a*) using MeBr instead of MeI. *d*) From XIV in alcoholic KOH by adding an aqueous solution of tartaric acid. *e*) From XIV. *f*) From III dissolved in acetone and MeBr at room temperature. *g*) From XIV and an equimolar amount of alcoholic KOH. *h*) No depression of m.p. of XVI from methods *f*) and *g*). *i*) From XIII and an equimolar amount of aqueous $NaHCO_3$. *j*) From XVI and an alcoholic solution of NaI. *k*) XIV was dissolved in alcoholic KOH (1/2 eq.), KBr was filtered off and the filtration treated with an alcoholic solution of NaI. *l*) No depression of m.p. of XVII from methods *i*, *j* and *k*. *m*) From XIII. *n*) From XVI. *o*) From III dissolved in acetone and *n*-PrI at room temperature. *p*) III dissolved in acetone and amyl iodide were kept at 120° for 16 h. The oily reaction product (XIX) was dissolved in acetone and MeI and kept at 120° for 17 h. *q*) As method *p*) using lauryl iodide instead of amyl iodide. The oily reaction product (XX) was treated as described under *p*). *r*) Found: I 30.8. Calc.: I 30 6 *s*) Found: I 29.2. Calc.: I 28.7.



IV

During the attempted bis-quaternization of III with methyl iodide a mono-quaternary product (in the form of the hydroiodide) (XIII) was formed in varying amounts together with IV. After removal of mono-quaternary product (see Experimental Part) pure IV (which was obtained in a 61 % yield) was converted into the corresponding sulfate (V) with silver sulfate. A number of other bis-quaternary salts were prepared from IV by the reaction of solutions of the corresponding free base, N-(γ-dimethylaminopropyl)-5-chloro-camphidine di-methohydroxide (VI) with the appropriate acids. The following salts were prepared: hydrogen sulfate (VII), tartrate (VIII), fumarate (IX), maleate (X), phosphate (XI) and bromide (XII) (see Table 1).

As mentioned above the hydroiodide of a mono-quaternary product (XIII) was formed during attempted bis-quaternization of III with methyl iodide. When the acetone used as a solvent was dried over "Sikkon" (Fluka), XIII was formed in a yield of 11 %. In a separate experiment using ordinary acetone, which had not been dried, attempted bis-quaternization gave 34 % of XIII.

A similar experiment using methyl bromide gave 65 % of the hydrobromide of a mono-quaternary product (XIV). XIV was converted into the corresponding tartrate (XV).

A mono-quaternary product is actually formed when methyl bromide is added to a cold solution of III in acetone, whereby a mono-methobromide (XVI) is formed in a 71 % yield. It was shown that XIV could be converted into XVI.

A mono-methiodide (XVII) was prepared in three different ways, *viz.* XIII → XVII (81 %), XVI → XVII (73 %), and XIV → XVII (60 %).

Probably mono-quaternization easily takes place at the terminal N-atom whereas more drastic conditions (120°, autoclave) are required for the quaternization of the N-atom of the camphidine ring. In this connection it is of interest to note that it was impossible to quaternize some mono-quaternary compounds prepared from III and higher alkyl iodides (*viz.* propyl (XVIII), amyl (XIX) and lauryl (XX)) with methyl iodide.

No bis-quaternary compounds were isolated, but certain amounts of the hydroiodides of the mono-quaternary compounds were formed. XVIII was isolated as a crystalline product, XIX and XX were obtained as oils. Attempted quaternization of XIX and XX with methyl iodide gave the hydroiodides (XXI and XXII) of XIX and XX (see Table 2).

EXPERIMENTAL *

N-(γ-Dimethylaminopropyl)-5-chlorocamphorimide (II). A mixture of 5-chlorocamphorimide [7] (600 g), 85 % potassium hydroxide (367 g) and ethanol (5.8 l) was heated

* All melting points are uncorrected. Microanalyses were carried out by Dr. W. Kirsten and his staff, Institute of Medical Chemistry, Uppsala, Sweden.

under reflux with stirring and a solution of γ-dimethylaminopropyl chloride hydrochloride (439 g) in ethanol (1.2 l) was added during 2 min with vigorous stirring. After heating under reflux for 6 h the mixture was left standing overnight, the potassium chloride removed by filtration and washed twice with ethanol. The filtrate was evaporated in a vacuum, the residue dissolved in benzene (1.6 l) and extracted with 3 N sodium hydroxide (4 × 250 ml) and water (3 × 200 ml). Drying and evaporation in a vacuum gave 581 g (70 %) of crude II, which was used directly in the next step. (Crude II decomposed at distillation and resisted attempts to crystallization).

N-(γ-Dimethylaminopropyl)-5-chlorocamphidine (III). A solution of II (1150 g) in anhydrous benzene (1.84 l) was added during 6 h with stirring to a mixture of lithium aluminium hydride (334 g) in anhydrous ether (7.3 l) and a further 2.6 l of anhydrous ether was added. After standing overnight at room temperature the mixture was heated under reflux for 14 h. After cooling, water (415 ml) was gradually added followed by 15 % sodium hydroxide (415 ml) and finally water (770 ml). After stirring for 1 h magnesium sulfate was added, the residue removed by filtration and washed 6 times with ether. After drying with magnesium sulfate and evaporation the residue was distilled. Yield of III 880 g (84 %); b.p.$_{0.2}$ 91–95°, n_D^{25} 1.4889. (Found: N 10.2. Calc. for $C_{15}H_{29}ClN_2$: N 10.3).

The dihydrochloride (XXIII) was prepared in the usual way from a solution of III in ether and gaseous hydrogen chloride. After crystallization from ethanol-ether the m.p. was 295–296°. (Found: Cl 20.6. Calc. for $C_{15}H_{31}Cl_3N_2$: Cl 20.6 (ionic)).

N-(γ-Dimethylaminopropyl)-5-chlorocamphidine di-methiodide (IV). Methyl iodide (410 ml) was gradually added with stirring to a solution of III (450 g) in acetone (dried over "Sikkon" Fluka) (2.3 l), the temperature being kept at 35–40° by cooling. The mixture was placed in an autoclave and kept at 120° for 17 h. After cooling the precipitate was removed by filtration, washed with cold acetone and dried. The yield was 735 g (80 %) of crude IV. Potentiometric titration with 0.1 N sodium hydroxide showed that the crude product contained 10.8 % of the hydroiodide of III mono-methoiodide (XIII).

Crude IV (735 g) was dissolved in hot water (700 ml) and a solution of sodium hydrogen carbonate (20 g) in hot water (200 ml) was added. After filtration the filtrate was evaporated in a vacuum. The residue was dissolved in boiling ethanol (3.12 l). After filtration the filtrate was cooled, the precipitate removed by filtration, washed twice with cold ethanol and dried. Potentiometric titration with 0.1 N sodium hydroxide showed that the by-product had been removed. Yield of IV 557 g (62 %); m.p. 221–222°. (Found: C 36.7; H 6.3; Cl 6.5; N 5.0; I 45.5. Calc. for $C_{17}H_{35}ClI_2N_2$: C 36.6; H 6.3; Cl 6.4; N 5.0; I 45.6).

N-(γ-Dimethylaminopropyl)-5-chlorocamphidine di-methosulfate (V). A mixture of IV (600 g), silver sulfate (336 g) and water (6 l) was heated under reflux with stirring for 2 h. After standing overnight at room temperature the precipitate was removed by filtration and the filtrate treated with gaseous hydrogen sulfide. The precipitate was removed by filtration and the filtrate evaporated in a vacuum. The residue was pulverized, washed 4 times with acetone and dried. Yield of V 473 g (97 %); m.p. 176–177°. (Found: C 45.9; H 9.4; N 6.3; Cl 7.5; S 7.1; H_2O (Karl Fischer) 12.5. Calc. for $C_{17}H_{35}ClN_2O_4S$, $3H_2O$: C 45.1; H 9.1; N 6.2; Cl 7.8; S 7.1; H_2O 11.9).

After drying for 4 h. at 70° in a vacuum over phosphorus pentoxide a monohydrate is obtained. (Found: C 49.2; H 9.3; N 6.5; Cl 8.3. Calc. for $C_{17}H_{35}ClN_2O_4S$, H_2O: C 49.0; H 8.9; N 6.7; Cl 8.5).

Solution of N-(γ-dimethylaminopropyl)-5-chlorocamphidine dimethohydroxide (VI). Silver oxide (prepared from silver nitrate (3.05 g) in water (25 ml) and 1 N sodium hydroxide (18 ml)) was suspended in water (125 ml) and a solution of IV (5.0 g) in water (25 ml) was added. The mixture was shaken for 4 h at room temperature and the precipitate removed by filtration and washed.

Dimetho salts. From solutions of VI the following salts were prepared: hydrogen sulfate (VII), tartrate (VIII), furmarate (IX), maleate (X), phosphate (XI) and bromide (XII). Only one detailed description of a preparation (*viz.* of VII) will be given since the remaining preparations were carried out essentially in a similar way. The salts are listed in Table 1.

A mixture of a solution (50 ml) containing VI (0.604 g) and 0.558 N sulfuric acid (12.84 ml) was evaporated to dryness in a vacuum. The residue was crystallized three times from ethanol-ether. Yield of VII 0.45 g (50 %).

Table 3. Some pharmacological properties of 3-azabicyclo [3.2.1] octane derivates.

Compound	Anticholinergic effect[a] % of atropine	Ganglion blocking effect		BP action of 0.5-1 mg/kg. Change in % of initial pressure[d]	Mydriasis factor, mice of 0.1 mg/kg i.p.[e]	LD 50 i.p.[f] mg/kg Mice
		P[b] % of C_6	S[c] % of C_6			
IV, V, XI	0.01	60	100	25—40 (decrease)	261	250
XIII	1	100	0	0		
XIV	0.01	160		0		
XXII	0.002	0		0		
XXIV	0.002	240	100	25—40 (decrease)	121	218

a) Determined on isolated guinea pig ileum suspended in Tyrode solution and stimulated with carbachol. *b*) Parasympathetic ganglion blocking action determined on isolated guinea pig ileum suspended in Tyrode solution and stimulated with DMPP (dimethylphenylpiperazinium iodide), a specific stimulator of the peripheral synapses. Technique according to Fakstorp and Pedersen [8]. Blockade expressed in % of that elicited by hexamethonium (C_6). (*c*) Sympathetic ganglion blockade determined on superior cervical ganglion of the anesthetized cat intermittantly stimulated with a square wave electrical stimulus provoking a contraction of the nictitating membrane. For details of technique see [8]. Results expressed in percent of action of hexamethonium. *d*) Action on blood pressure measured manometrically in chloralose anesthetized cat cannulated in one carotid artery. *e*) Determined according to Edge [9]. Mydriasis factor is the area formed by a regression line, abscissa and ordinate in a coordinate system in which the abscissa shows time in hours after treatment, the ordinate indicates increase in pupil diameter in arbitrary units. Other ganglion blocking agents were included as standards. Only chlorisondamine (Ecolid ®) was active at the dosage level shown in the table, having a factor of 191. Pentapyrrolidinium (Ansolysen ®), mecamylamine (Inversine ®), oxapentammonium (Oxaditon ®) and hexamethonium (Vegolysen ®) were inactive at this level, but active at higher dosage. *f*) Mean lethal dose 24 h after intraperitoneal injection, mice. Calculated according to Kärber [10].

PHARMACOLOGY *

The trimethylammoniumpropyl chloromethyl camphidinium ion was used in various tests in the form of three salts, the iodide (IV), the sulfate (V) and the phosphate (XI). The bis-tertiary precursor was tested in the form of a bis-hydrochloride (XXIII). Compound XIV is a mono-quaternary amine used in the form of a bromide-hydrobromide, while XXII correspondingly was in the form of iodide-hydroiodide.

The acute experiments concerning the influence of the various compounds on the two parts of the autonomic nervous system and on circulation are summarized in Table 3, while the results obtained in a chronic experiment with hypertensive rats are shown in Table 4.

Ganglion blocking action of IV, V, XI upon the sympathetic superior cervical ganglion is of the same order of magnitude as that shown by hexamethonium, although of much longer duration. However, upon the parasympathetic ganglia of the ileum the chlorinated compounds (IV, V, XI) are considerably less active than the chlorine free substance (XXIV), indicating a desirable specificity of action.

This observation is, however, not corroborated by the action on the pupil. In the absence of anticholinergic effects (only XXIII has any such action) the observed mydriasis can be ascribed to blockade of peripheral parasympathetic synapses. Accordingly in

* In part presented at the Joint Meeting of the British and Scandinavian Pharmacological Societies, Copenhagen, July 26—29, 1960.

Table 4. Influence of two 3-azabicyclo [3.2.1] octane derivatives upon blood pressure of the unanaesthetized renal hypertensive rat.

Compound	Dose mg/kg/day [a]	Pre-drug period		Drug period		Post-drug period	
		Days	BP[b]	Days	BP	Days	BP
IV	3.5	15	171	14	154	8	173
XXIV	3.1	14	173	14	153	20	163

a) Compounds mixed in the feed. Each compound studied in groups of 4 animals made hypertensive by unilateral clamping of renal artery according to Wilson & Byrom [11]. *b*) Blood pressure in mm Hg, average of daily observations during period shown in preceding column.

this technique the chlorinated compound IV shows very strong parasympathetic blocking properties indeed, stronger than the chlorine free XXIV. No explanation can be offered for the discrepancy between potency values obtained in the two techniques.

Blood pressure of the anesthetized cat is affected by all the bis-quaternary compounds. The action is slow and sustained, and persists even after the synaptic transmission and the carotid occlusion reflex, which are depressed or blocked soon after the drug is given, are restored. This may indicate a component of action on blood pressure distinct from that which interrupts synaptic transmission.

The lowering of blood pressure is observed after giving the drugs intravenously (0.5 mg/kg) or by stomach tube (2 mg/kg).

The mono-quaternary compounds (XIV and XXII) and the bis-tertiary amine (XXIII) are without effect upon blood pressure.

A lowering of blood pressure during oral application to the hypertensive rat is observed with the chlorine-containing as well as with the chlorine-free bis-quaternary compound.

REFERENCES

1. Fakstorp, J., Christiansen, J. and Pedersen, J. G. A. *Acta Chem. Scand.* **7** (1953) 134.
2. Fakstorp, J., Pedersen, J. G. A., Poulsen, E. and Schilling, M. *Acta Pharmacol. Toxicol.* **13** (1957) 52.
3. Plummer, A. J., Trapold, J. H., Schneider, J. A., Maxwell, R. A. and Earl, A. E. *J. Pharmacol. Exptl. Therap.* **115** (1955) 172.
4. Archer, W. L., Cavallito, C. J. and Gray, A. P. *J. Am. Chem. Soc.* **78** (1956) 1227.
5. Lape, H. E., Fort, D. J. and Hoppe, J. O. *J. Pharmacol. Exptl. Therap.* **116** (1956) 462.
6. O'Malley, W. E., Haemmerli, G. W., Rice, L. M. and Geschickter, C. F. *J. Am. Pharm. Assoc. Sci. Ed.* **47** (1958) 263.
7. Scheiber, J. and Knothe, M. *Ber.* **45** (1912) 1551.
8. Fakstorp, J. and Pedersen, J. G. A. *Acta Pharmacol. Toxicol.* **10** (1954) 7.
9. Edge, N. D. *Brit. J. Pharmacol.* **8** (1953) 10.
10. Kärber, G. *Arch. exptl. Pathol. Pharmakol.* **162** (1931) 480.
11. Wilson, C. and Byrom, F. B. *Lancet* **1939-I** 136.

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