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SYNTHESIS, STRUCTURE, AND BIOLOGICAL ACTIVITY OF NOVEL BISPIDINE DERIVATIVES

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ABSTRACT

Objective: Derivatives of 3,7-diazabicyclo[3.3.1]nonan-9-one attract considerable attention from pharmacists for the treatment of a wide range of diseases. According to this interest, the novel derivatives of 3-cyclopropanmethyl-7-alkoxyalkyl-3,7-diazabicyclo[3.3.1]nonan-9-one with isopropoxypropyl and ethoxypropyl substituents in the position 7 had been synthesized to study their biological activity and toxicity. The practical significance of the work is in the accumulation and development of scientific representations about diazabicyclic compounds, methods for their synthesis, structure, and properties, which can subsequently be used in a targeted design and identification of even more complex systems, as well as in the development of further research in the field of 3,7-diazabicyclo[3.3.1]nonanes. For this purpose, complexes of the synthesized compounds with β-cyclodextrin are obtained and their biological activity is investigated at the Department of Pharmacology of S.D. Asfendiyarov Kazakh National Medical University with the aid of the pharmacological tests.

Methods: An experimental study of local anesthetic activity on the models of infiltration, conduction anesthesia, and acute toxicity of synthesized molecules was carried out using primary screening methods.

Results: As a result of pharmacological screening, it has been found that the compounds exhibit local anesthetic activity and low toxicity and was recommended for in-depth study of their pharmacological properties.

Conclusion: It turned out that a nature of the N-alkoxyalkyl radical does not affect the toxicity of cyclopropanmethyl- substituted bispidines. In the series of O-benzoyloximes of bispidinones, the isopropoxypropyl- substituted analog is 1.3 times less toxic than ethoxypropyl- one.

Keywords: Bispidine, Synthesis, Structure, Activity, Anesthetics, Acute toxicity.

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INTRODUCTION

An analysis of current trends in the medical use of drugs indicates the ongoing gradual replacement of obsolete drugs with more effective and safe drugs of novel generations. Therefore, research on the search for novel potentially biologically active substances is relevant.

The aim of research is the synthesis of novel potentially pharmacologically active derivatives of 3-cyclopropanmethyl-7-alkoxyalkyl-3,7-diazabicyclo[3.3.1]nonan-9-one, as well as the establishment of the structure and evaluation of their biological activity.

The interest of chemists in the study of heteroanalogs of bicyclo[3.3.1] nonane is due to the unique properties of these compounds, which makes them valuable from a theoretical and practical point of view [1-10]. It is also known that methylenecyclopropane residue is a valuable structural unit of bioactive compounds, particularly, opiate antagonists [11,12].

1-(3-Isopropoxypropyl- and 3-ethoxypropyl-)-4-oxopiperidine (1, 2) as starting substrates was used to obtain the target bispidines (3-14) according to Fig. 1.

The reaction products were obtained with high yields as viscous oils. Monitoring of the progress of the reaction was carried out by TLC on alumina. The structure of bispidinone derivatives (3-10) was determined by nuclear magnetic resonance (NMR) and infrared (IR) spectroscopies.

To study the biological properties of the novel derivatives of 3,7-diazabicyclo[3.3.1]nonan-9-ones, their complexes with β-cyclodextrin had been synthesized (11-14).

METHODS

Experimental chemical part

NMR spectra of the studied compounds were recorded on a JEOL JNM-ECA400 spectrometer with an operating frequency on carbon nuclei of 100.53 MHz in $CDCl_3$ with hexamethyldisiloxane as internal standard. Elemental analysis data were consistent with calculated values. IR spectra were recorded on a Nicolet 5700 instrument between KBr plates. Column chromatography and thin-layer chromatography were carried out on alumina (Al_2O_3) of the third degree of activity, R_f of the compounds is provided for this type of plate. The spots were developed in iodine vapors.

3-Cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1] nonan-9-one (3)

In a three-necked flask equipped with a stirrer, reflux condenser, and dropping funnel, 60 ml of methanol was deoxygenated under a stream of nitrogen. After 30 min, a mixture of 4.6 g (0.059 mol) of cyclopropanemethylamine, 7.2 g (0.46 mol) of paraform, 3.1 ml of concentrated hydrochloric acid, and 4.5 ml of glacial acetic acid was added and stirred for 15 min in the atmosphere of nitrogen. A solution of 11.7 g (0.059 mol) of 1-(3-isopropoxypropyl)piperidin-4-one (1) and 4.5 ml of glacial acetic acid in 15 ml of methanol was added

dropwise. After 10 h of heating the reaction mixture at 60–65°C, a second equivalent of paraform was added and held for another 12 h at the same temperature. The solvent was evaporated, the residue was dissolved in 30 ml of water. The extraction of neutral products was carried out with diethyl ether. The aqueous layer was alkalinized to pH 12 and the organic part was extracted with chloroform, dried over $MgSO_4$. The solvent was evaporated, the resulting product was purified on a column of aluminum oxide, benzene:dioxane 5:1. 9.3 g (73.6%) of 3-cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1] nonan-9-one (3) was obtained in the form of a light yellow oil with R_f=0.23 (Al_2O_3, benzene:isopropanol = 6:1).

Found, %: C 69.41, H 10.24, N 9.55. $C_{17}H_{30}N_2O_2$.

Calculated, %: C 69.38, H 10.20, N 9.52.

IR spectrum, cm^{-1}: 1734 ($\nu_{C=O}$), 1122 (ν_{C-O-C}).

^{13}C-NMR spectrum, δ, ppm ($CDCl_3$): 46.6 ($C_{1,5}$), 58.4 ($C_{6,8}$), 58.8 ($C_{2,4}$), 214.4 (C_9), 57.9 (C_{10}), 31.7 (C_{11}), 64.8 (C_{12}), 67.7 (C_{13}), 28.2 (C_{14}). 63.1 (C_{16}), 7.7 (C_{17}), 4.0 (C_{18}).

3-Cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (4)

In a three-necked flask equipped with a stirrer, reflux condenser, and dropping funnel, 80 ml of methanol was deoxygenated under a stream of nitrogen. After 30 min, a mixture of 5.2 g (0.070 mol) of cyclopropanemethylamine, 8.7 g (0.140 mol) of paraform, 3.8 ml of concentrated hydrochloric acid, and 5.6 ml of glacial acetic acid was added and stirred for 15 min under nitrogen atmosphere. A solution of 13.4 g (0.070 mol) of 1-(3-ethoxypropyl)piperidin-4-one (2) and 6.2 ml of glacial acetic acid in 11 ml of methanol was added dropwise. After heating the reaction mixture for 10 h at 60–65°C, the second equivalent of paraform is added and the mixture was held for another 12 h at the same temperature. Throughout the reaction, the reaction mixture is purged with a stream of nitrogen. The solvent was evaporated, the residue was dissolved in 113 ml of water. The extraction of neutral products was carried out with diethyl ether. The aqueous layer was alkalinized to pH 12 and the organic part was extracted with chloroform, dried over $MgSO_4$. The solvent was evaporated, the resulting product was purified on a column of aluminum oxide, benzene:dioxane = 5:1. 16.1 g (79%) of 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyc-lo[3.3.1]nonan-9-one (4) was obtained in the form of a light yellow oil with R_f=0.50 (Al_2O_3, benzene:isopropanol = 6:1).

Found, %: C 68.38, H 10.12, N 9.92. $C_{16}H_{28}N_2O_2$.

Calculated, %: C 68.57, H 10.00, N 10.00.

IR spectrum, cm^{-1}: 1736 ($\nu_{C=O}$), 1118 (ν_{C-O-C}).

^{13}C-NMR spectrum, δ, ppm ($CDCl_3$): 46.6 ($C_{1,5}$), 58.2 ($C_{6,8}$), 58.7 ($C_{2,4}$), 211.1 (C_9), 58.4 (C_{10}), 27.6 (C_{11}), 66.2 (C_{12}), 68.6 (C_{13}), 15.2 (C_{14}), 67.1 (C_{15}), 9.1 (C_{16}), 4.0 (C_{17}).

3-Cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1] nonan (5)

A mixture of 2.0 g (0.0068 mol) of 3-cyclopropanmethyl-7-(3-isopropoxy-propyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (3) and 1.09 g (0.034 mol) of hydrazine hydrate (99% solution) in 20 ml of triethylene glycol at 60°C was added 4.7 g (0.084 mol) of KOH. The reaction mixture was heated to 150°C and stirred at this temperature for 4 h. At a temperature of 190–200°C, water and excess hydrazine were removed by evaporation. After cooling, 33 ml of distilled water was added, extracted with diethyl ether, and dried over anhydrous $MgSO_4$. The solvent was evaporated, the obtained product was purified by column chromatography on Al_2O_3, benzene:dioxane 5:1. 2.1 g (42%) of 3-cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1] nonane (5) is obtained as a pale yellow oil with R_f=0.23 (Al_2O_3, benzene:isopropanol = 7:1).

Found, %: C 72.89, H 11.41, N 10.05. $C_{17}H_{32}N_2O$

Calculated, %: C 72.85, H 11.43, N 10.00.

IR spectrum, cm^{-1}: 1112 (ν_{C-O-C}).

^{13}C-NMR spectrum, δ, ppm ($CDCl_3$): 29.8 ($C_{1,5}$), 58.4 ($C_{6,8}$), 58,8 ($C_{2,4}$), 31,4 (C_9), 57.7 (C_{10}), 28.1 (C_{11}), 66.5 (C_{12}), 73.3 (C_{13}), 22.3 (C_{14}), 67.0 (C_{16}), 8.6 (C_{17}), 4.1 (C_{18}).

3-Cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo[3.3.1] nonan (6)

A mixture of 5.0 g (0.018 mol) 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (4) and 2.88 g (0.090 mol) hydrazine

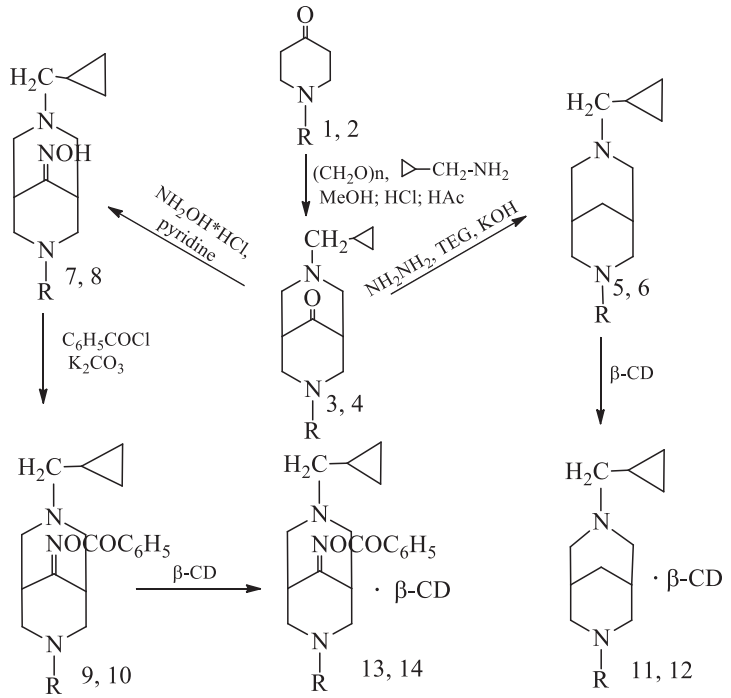


Fig. 1: Synthetic routes to the target bispidines

hydrate (99% solution) in 52.5 ml of triethylene glycol at 60°C add 12.5 g (0.22 mol) of KOH. The reaction mixture was heated to 150°C and stirred at this temperature for 4 h. At a temperature of 190–200°C, water and excess hydrazine were removed by evaporation. After cooling, 64 ml of distilled water was added, extracted with diethyl ether, and dried over anhydrous $MgSO_4$. The solvent was evaporated, the obtained product was purified by column chromatography on Al_2O_3, benzene:dioxane 5:1. 2.1 g (42% of theory) of 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo[3.3.1]nonane (6) was obtained in the form of a light yellow oil with R_f=0.34 (Al_2O_3, benzene:isopropanol = 7:1).

Found, %: C 72.32, H 11.12, N 10.65. $C_{16}H_{30}N_2O$.

Calculated, %: C 72.18; H 11.27; N 10.52.

IR spectrum, ν, cm^{-1}: 1111 ($\nu_{C\text{-}O\text{-}C}$).

^{13}C-NMR spectrum, δ, ppm ($CDCl_3$): 30.6 ($C_{1,5}$), 58.6 ($C_{6,8}$), 58.0 ($C_{2,4}$), 31.8 (C_9), 57.4 (C_{10}), 27.9 (C_{11}), 66.1 (C_{12}), 69.6 (C_{13}), 15.2 (C_{14}), 67.2 (C_{15}), 54.1 (C_{17}), 9.5 (C_{16}), 4.2 (C_{17}).

Oxime of 3-cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (7)

3.0 g (0.0102 mol) of 3-cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (3) in 60 ml of ethyl alcohol and 1.23 g (0.0153 mol) of pyridine were placed in a three-necked flask equipped with a mechanical stirrer, reflux condenser with a calcium chloride tube, and dropping funnel. While stirring, 1.84 g (0.0265 mol) of hydroxylamine hydrochloride was added. The reaction mixture was heated at 110–120°C for 20 h, the solvent was evaporated, and the residue was dissolved in 15 ml of water, alkalized with NaOH to pH 12, extracted with chloroform, and dried over anhydrous $MgSO_4$. The solvent was evaporated, the residue was purified by column chromatography on Al_2O_3, benzene:dioxane =5:1. 1.83 g (59%) of oxime of 3-cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (7) was obtained in the form of light yellow oils with R_f=0.016 (Al_2O_3, benzene:isopropanol = 20:1).

Found, %: C 66.05, H 10.65, N 13.63. $C_{17}H_{31}N_3O_2$.

Calculated, %: C 66.02, H 10.67, N 13.59.

IR spectrum, cm^{-1}: 1668 ($\nu_{C=N}$), 3074 ($\nu_{O\text{-}H}$).

^{13}C-NMR spectrum, δ, ppm ($CDCl_3$): 38.1 (C_1), 32.0 (C_5), 59.5; 59.7 ($C_{2,4}$), 57.9, 58.0 ($C_{6,8}$), 162.0 (C_9), 57.6 (C_{10}), 31.7 (C_{11}), 64.8 (C_{12}), 67.7 (C_{13}), 28.2 (C_{14}), 63,1 (C_{15}), 7.7 (C_{16}), 4.0 (C_{17}).

Oxime of 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (8)

6.0 g (0.0155 mol) of 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (4) in 160 ml of ethyl alcohol and 3 ml (0.0375 mol) of pyridine were placed in a three-necked flask equipped with a mechanical stirrer, reflux condenser with a calcium chloride tube, and dropping funnel. With stirring, 4.5 g (0.065 mol) of hydroxylamine hydrochloride was added. The reaction mixture was heated at 110–120°C for 20 h, the solvent was evaporated, and the residue was dissolved in 10 ml of water, alkalized with NaOH to pH 12, extracted with chloroform, and dried over anhydrous MgSO4. The solvent was evaporated, the residue was purified by column chromatography on Al_2O_3, benzene:isopropanol-20:1. 5.4 g (85%) of oxime of 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (8) was obtained in the form of light yellow oils with Rf=0.06 (Al_2O_3, benzene:isopropanol = 20:1).

Found, %: C 65.22, H 9.89, N 14.36. $C_{16}H_{29}N_3O_2$.

Calculated, %: C 65.08, H 9.83, N 14.24.

IR spectrum, cm^{-1}: 1643 ($\nu_{C=N}$), 3291 ($\nu_{O\text{-}H}$).

^{13}C-NMR spectrum, δ, ppm ($CDCl_3$): 37.0 (C_1), 32.5 (C_5), 58.5, 58.3 ($C_{2,4}$), 58.5, 58.4 ($C_{6,8}$), 165.8 (C_9), 58.7 (C_{10}), 27.3 (C_{11}), 69.1 (C_{12}), 70.8 (C_{13}), 27.3 (C_{14}), 54.0 (C_{16}), 8.4 (C_{17}), 4.0 (C_{18}).

O-Benzoyloxime of 3-cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (9)

1.5 g (0.0048 mol) of oxime of 3-cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (7) was mixed with 15 ml of absolute benzene and a mixture of 2 ml of absolute benzene and 0.6 ml (0.0048 mol) of benzoyl chloride was added dropwise. Reaction took place at room temperature. At the end, 10 ml of distilled water was added to the reaction mixture and neutralized with potash to pH 10–11, the product was extracted with chloroform, the combined extracts were dried over anhydrous $MgSO_4$. The solvent was evaporated and residue was distilled *in vacuo*. 1.9 g (91 % of theory) of O-benzoyloxime of 3-cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one was obtained in the form of a yellow oil (9), R_f=0.77 (Al_2O_3, benzene:isopropanol = 7:1).

Found, %: C 69.64, H 8.24, N 10,20. $C_{24}H_{35}N_3O_3$.

Calculated, %: C 69.73, H 8.48, N 10.17.

IR spectrum, cm^{-1}: 1745 ($\nu_{C=O}$), 1641 ($\nu_{C=N}$).

^{13}C-NMR spectrum, δ, ppm ($CDCl_3$): 38.7, 34.6 ($C_{1,5}$), 57.6, 59.3, 59.6, 59.7 ($C_{2,4,6,8}$), 161.2 (C_9), 57.8 (C_{10}), 28.1 (C_{11}), 64.8 (C_{12}), 70.8 (C_{13}), 28.2 (C_{14}), 63,1 (C_{15}), 7.9 (C_{16}), 4.0 (C_{17}), 171.4 (C_{18}), 129.2 (C_{19}), 129.7 (C_{20}), 128.4 (C_{21}), 133.4 (C_{22}).

O-Benzoyloxime of 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (10)

4.0 g (0.01356 mol) of oxime of 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one (8) was mixed with 40 ml of absolute benzene and a mixture of 12 ml of absolute benzene and 1.9 ml (0.01356 mol) of benzoyl chloride was added dropwise. Reaction took place at room temperature. At the end, 15 ml of distilled water was added to the reaction mixture and neutralized with potash to pH 10–11, the product is extracted with chloroform, the combined extracts were dried over anhydrous $MgSO_4$. The solvent was evaporated and residue was distilled *in vacuo*. 2.4 g (46% of theory) of O-benzoyloxime of 3-cyclopropanmethyl-7-(3-ethoxy-propyl)-3,7-diazabicyclo[3.3.1]nonan-9-one was obtained in the form of a yellow oil (10), R_f=0.30 (Al_2O_3, benzene:isopropanol = 7:1).

Found, %: C 69.12, H 8.34, N 10.65. $C_{23}H_{33}N_3O_3$.

Calculated, %: C 69.17, H 8.27, N 10.52.

IR spectrum, cm^{-1}: 3300.4 ($\nu_{O\text{-}H}$), 1675.4 ($\nu_{C=N}$).

^{13}C-NMR spectrum, δ, ppm ($CDCl_3$): 37.2, 33.6 ($C_{1,5}$), 57.0, 57.4, 57.9, 58.4 ($C_{2,4,6,8}$), 164.4 (C_9), 58.7 (C_{10}), 27.4 (C_{11}), 67.1 (C_{12}), 68.9 (C_{13}), 31.9 (C_{14}), 62.1 (C_{15}), 8.4 (C_{16}), 3.9 (C_{17}), 171.6 (C_{18}), 128.4 (C_{19}), 129.7 (C_{20}), 128.5 (C_{21}), 133.2 (C_{22}).

Complex of 3-cyclopropanmethyl-7-(3-isopropoxy-propyl)-3,7-diazabicyclo[3.3.1]nonane with β-cyclodextrin (11)

Hot solutions of 0.9 g (0.0033 mol) of 3-cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1]nonane (5) in 25 ml of ethyl alcohol and 3.7 g (0.0033 mol) of β-cyclodextrin in 35 ml of distilled water were mixed together. The mixture was placed in a drying cupboard, ethanol and water were evaporated at 50–55°C to produce 4.5 g of compound (11).

Found, %: C 50.14, H 7.15, N 1.93. $C_{59}H_{102}N_2O_{36}$.

Calculated, %: C 50.07, H 7.21, N 1.98.

Complex of 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo[3.3.1]nonane with β-cyclodextrin (12)

Hot solutions of 1.7 g (0.0064 mol) of 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabi-cyclo[3.3.1]nonane (6) in 25 ml of ethyl alcohol and 7.2 g (0.0064 mol) of β-cyclodextrin in 45 ml of distilled water were mixed together. The mixture was placed in a drying cupboard, ethanol and water were evaporated at 50–55°C to produce 8.7 g of compound (12).

Found, %: C 59.61, H 7.26, N 1.93. $C_{58}H_{100}N_2O_{36}$.

Calculated, %: C 59.71, H 7.14, N 2.00.

Complex of O-benzoyloxime of 3-cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabi-cyclo[3.3.1]nonan-9-one with β-cyclodextrin (13)

Hot solutions of 1.9 g (0.004 mol) of O-benzoyloxime of 3-cyclopropanmethyl-7-(3-isopropoxypro-pyl)-3,7-diazabicyclo[3.3.1] nonan-9-one (9) in 25 ml of ethyl alcohol and 4.8 g (0.004 mol) of β-cyclodextrin in 30 ml of distilled water were mixed together. The mixture was placed in a drying cupboard, ethanol and water were evaporated at 50–55°C to produce 5.4 g of compound (13).

Found, %: C 51.16, H 6.69, N 2.66. $C_{66}H_{105}N_3O_{38}$.
Calculated, %: C 51.20, H 6.79, N 2.71.

Complex of O-benzoyloxime of 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo-[3.3.1]nonan-9-one with β-cyclodextrin (14)

Hot solutions of 1.4 g (0.00366 mol) of O-benzoyloxime of 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo[3.3.1] nonan-9-one (10) in 20 ml of ethyl alcohol and 4.15 g (0.00366 mol) of β-cyclodextrin in 30 ml of distilled water were mixed together. The mixture was placed in a drying cupboard; ethanol and water were evaporated at 50–55°C to produce 4.2 g of compound (14).

Found, %: C 50.85, H 6.78, N 2.77. $C_{65}H_{103}N_3O_{38}$.
Calculated, %: C 50.88, H 6.72, N 2.74.

Experimental biological part

An experimental study of local anesthetic activity on the models of infiltration, conduction anesthesia, and acute toxicity of the synthesized molecules was carried out using primary screening methods in six animals in each series at the Department of Pharmacology of S.D. Asfendiyarov Kazakh National Medical University in accordance with the "Rules for the use of experimental animals" (European Convention No. 123, 1986, Helsinki Declaration 2000) and the State Standard of the Republic of Kazakhstan "Good Laboratory Practice" (the main provisions of ST RK 1613-2006) and the "Rules of Good Laboratory Practice of the Customs Union" (Attachment to the Decision of the Commission of the Customs Union, No. 564, dated March 2, 2011). Besides, the team has a positive opinion from the LEC (local expert commission) of S.D. Asfendiyarov Kazakh National Medical University (Minutes No. 7 (58) dated September 12, 2017) on the compliance of work with experimental animals.

The anesthetic activity of the synthesized compounds was compared with those of widely used anesthetics – trimecaine, lidocaine, and novocaine.

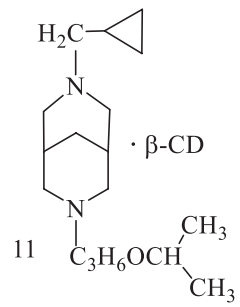
The evaluation of infiltration anesthesia was studied on guinea pigs according to the Bulbring-Wade method. The following values were determined: The depth of anesthesia (anesthesia index), the duration of deep anesthesia, and the total duration of the anesthetic effect.

The conduction anesthesia was examined through the modified tail flick method on rats. It allows to measure the duration of deep anesthesia and the total duration of the anesthetic effect of the compound.

The study of acute toxicity was carried out on mice of the same species, sex, age, and weighing 18–22 g. Acute toxicity (LD_{50}) was determined by a single subcutaneous injection of aqueous solutions of the compounds.

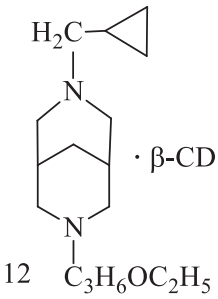
The local anesthetic effect and acute toxicity of novel derivatives of 3,7-diazabicyclo[3.3.1] nonan-9-one in the form of complexes with β-cyclodextrin were studied under laboratory codes LA 1-4:

LA-1



Complex of 3-cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1]nonane with β-cyclodextrin

LA-2



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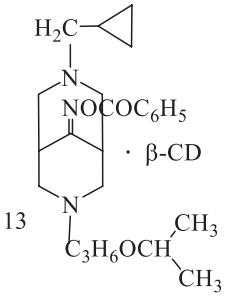
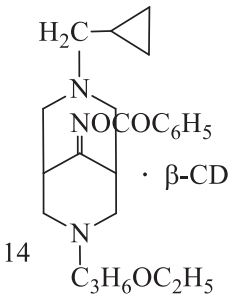
LA-3



Complex of O-benzoyloxime of 3-cyclopropanmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one with β-cyclodextrin

LA-4



Complex of O-benzoyloxime of 3-cyclopropanmethyl-7-(3-ethoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one with β-cyclodextrin

RESULTS AND DISCUSSION

The infiltration anesthesia

It turned out that on the model of infiltration anesthesia in a bispidines family, the most active is LA-2, the anesthesia index of which is 35.60±0.19 (Table 1). In addition, it caused a longer complete anesthesia of 34.00±1.26 min, while the total duration of action was 43.00±1.84 min. Replacing the ethoxypropyl radical in the LA-2 compound with an isopropoxypropyl- one led to a three-fold decrease in activity.

Among the two of O-benzoyloximes of 3,7-diazabicyclo[3.3.1]nonan-9-ones, LA-3 with cyclopropanmethyl and isopropoxypropyl radicals at nitrogen atoms was active, its anesthesia index was 35.60±1.18, and the duration of deep anesthesia was equal to 34.80±0.98 min. The total duration of action was 43.10±1.17 min. Replacing the isopropoxypropyl radical with a ethoxypropyl- one led to a decrease in activity (LA-4) (anesthesia index was 29.16±1.45; the duration of deep anesthesia was 16.00±2.90 min; and the total analgesic effect lasted (29.16±2.06 min). However, it should be noted that it was weaker than comparison preparations (не знаю, говорят ли так, я предлагаю язамену) for the reference compounds.

Conduction anesthesia

It should be noted that bispidines and O-benzoyloximes on the model of conduction anesthesia did not show any significant effect. However, LA-1 can be distinguished, which in terms of total duration of action is comparable to novocaine and inferior to trimecaine and lidocaine.

In the group of O-benzoyloximes, according to the duration of total anesthesia, LA-4 exceeded trimecaine and novocaine (Table 2).

Acute toxicity

The acute toxicity of bispidine derivatives is presented in Table 3.

Table 1: Infiltration anesthetic activity and duration of action of 0.25% solutions of LA-1-4 and comparison preparations

Preparation	0.25%					
	Anesthesia index, M±м		Duration of deep anesthesia, min		Total duration of action, min	
LA-1	28.50±0.76	p_1<0.050 p_2<0.001 p_3<0.020	11.30 ± 2.00	p_1<0.010 p_2>0.050 p_3>0.050	30.80± 0.90	p_1<0.010 p_2>0.050 p_3>0.050
LA-2	35.60±0.19	p_1<0.050 p_2<0.001 p_3<0.001	34.00±1.26	p_1<0.001 p_2<0.001 p_3<0.001	43.00±1.84	p_1<0.050 p_2<0.001 p_3<0.001
LA-3	35.60±1.18	p_1<0.005 p_2<0.001 p_3<0.001	34.80±0.98	p_1<0.001p_2<0.001p_3>0.001	43.10±1.17	p_1<0.05 p_2<0.001 p_3>0.001
LA-4	29.16±1.45	p_1>0.050 p_2<0.002 p_3<0.050	16.00±2.90	p_1>0.050 p_2>0.050 p_3<0.050	29.16±2.06	p_1<0.002 p_2>0.050 p_3>0.050
Trimecaine	32.10±1.50		20.00±1.70		38.30±1.05	
Lidocaine	23.10±0.90		14.20±0.80		30.80±0.80	
Novocaine	25.00±1.00		10.00±1.20		29.10±1.50	

p_1 – correlation coefficient compared with trimecaine, p_2 – compared with lidocaine, p_3 - compared with novocaine

Table 2: Conduction anesthetic activity and duration of action of 1% solutions of the preparations

Preparation	1%			
	Duration of deep anesthesia, min		Total duration of action, min	
LA-1	25.00±3.17	p_1<0.020 p_2<0.050 p_3>0.050	45.0±1.2	p_1<0.050 p_2<0.020 p_3>0.050
LA-2	14.16±3.76	p_1<0.001 p_2<0.020 p_3>0.050	22.5±3.3	p_1<0.020 p_2<0.002 p_3>0.050
LA-3	10.80±2.72	p_1<0.001 p_2<0.020 p_3>0.002	32.8±2.4	p_1>0.050 p_2<0.010 p_3>0.050
LA-4	35.00±7.40	p_1>0.050 p_2>0.050 p_3>0.050	67.5±6.4	p_1>0.050 p_2>0.050 p_3>0.050
Trimecaine	47.30±8.40		56.9±12.8	
Lidocaine	65.00±18.40		90.0±18.4	
Novocaine	35.20±7.10		42.3±13.6	

p_1 – correlation coefficient compared with trimecaine, p_2 – compared with lidocaine, p_3 – compared with novocaine

Table 3: Acute toxicity of the preparations

Preparation	LD_{50} (mg/kg)	P – reliability of results
LA-1	505.0	p_1<0.001, p_2<0.001, p_3>0.050, p_4<0.001
LA-2	525.0	p_1<0.001, p_2<0.001, p_3>0.050, p_4<0.001
LA-3	825.0	p_1<0.001, p_2<0.001, p_3<0.001, p_4<0.001
LA-4	633.0	p_1<0.001, p_2<0.001, p_3<0.001, p_4<0.001
Trimecaine	378.2±19.4	
Lidocaine	248.6±18.4	
Novocaine	480.0±9.8	
Dikain	41.5±1.9	

p_1 – correlation coefficient compared with trimecaine, p_2 – compared with lidocaine, p_3 – compared with novocaine, p_4 – compared with dikain

As follows from the data of Table 3, derivatives of 3,7-diazabicyclo[3.3.1] nonan-9-one are low toxic compounds. The most harmless is LA-3 (complex of O-benzoyloxy-3-cycloproylmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-one with β-CD), where the LD_{50} is 825.0 mg/kg.

It turned out that a nature of the N-alkoxyalkyl radical does not affect the toxicity of cyclopropanmethyl- substituted bispidines (LA-1 and LA-2). In the series of O-benzoyloximes of bispidinones, the isopropoxypropyl-substituted analog is 1.3 times less toxic than ethoxypropyl- one.

CONCLUSION

In this research, derivatives of 3,7-diazabicyclo[3.3.1]nonan-9-ones were synthesized and the biological properties were studied for their complexes with β-cyclodextrin.

Thus, as a result of structural modifications of bicyclic piperidines, novel low toxic substances possessing a local anesthetic effect had been obtained, the activity of which depends on the nature of the N-alkoxyalkyl- and substituents at the position 9 of the 3,7-diazabicyclo[3.3.1]nonane ring. The most harmless is the complex of O-benzoyloxy-3-cycloproylmethyl-7-(3-isopropoxypropyl)-3,7-diazabicyclo[3.3.1]nonan-9-ketoxime with β-CD, where the LD_{50} is 825 mg/kg. The compound was recommended for in-depth study of its pharmacological properties.

Moreover, the results of research will be used in a targeted design and identification of even more complex systems on the base of 3,7-diazabicyclo[3.3.1]nonan-9-ones with different biological activity.

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AUTHORS' CONTRIBUTIONS

All the authors have contributed equally.

CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

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