SYNTHESIS OF AMINOMETHOXY DERIVATIVES OF CYCLOHEXANOL AND STUDYING THEM AS BACTERICIDE INHIBITORS

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Based on cyclohexanol, cyclic amines (piperidine, morpholine, hexamethyleneimine), and formaldehyd, new Mannich bases have been synthesized. The reaction was carried out at a temperature of 78–80°C for 4–5 h in a benzene solution at an equimolar ratio of the starting components. The yield of compounds was 69–75%. The physicochemical data of the synthesized compounds were determined. The composition and structure of the target products were confirmed by elemental analysis, IR, 1H and 13C NMR spectroscopy, and mass spectrometry. Their effect on the vital activity of sulfate-reducing bacteria of the "Desulfovibrio desulfuricans" type at three concentrations (25; 50; 100 mg/l) was studied. The obtained compounds showed high bactericidal properties, 1% solutions of these compounds in isopropl alcohol at a concentration of 100 mg/l showed 100% bactericidal effect. A 1% solution of the compound with a fragment of morpholine already at a concentration of 50 mg/l exhibits a 100% bactericidal effect. Compounds with a fragment of piperidine and hexamethyleneimine at a concentration of 50 mg/l showed 96.2 and 98% bactericidal effects, respectively. And at a concentration of 25 mg/l, the bactericidal effect of all three compounds was 16.4%, 36.4%, and 58.2%, respectively. The results obtained show that the synthesized compounds showed more bactericidal properties compared to the industrially used bactericide-inhibitors AMДОР ИК-7 and AMДОР ИК-10 taken as a standard. Established, that these aminomethoxy derivatives of cyclohexanol affect bacteria at very low concentrations, they can be proposed as effective inhibitors against sulfate-reducing bacteria.

Keywords: cyclohexanol, secondary amines, Mannich bases, sulfate-reducing bacteria, inhibitor-bactericides, biocorrosion.

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Introduction

A significant part of the corrosion damage of oilfield equipment is due to the vital activity of a number of microorganisms and, first of all, sulfate-reducing bacteria (SRB) [1, 2]. Sulfate-reducing bacteria, a group of anaerobic prokaryotes, are widely distributed in various anoxic habitats such as lakes, swamps, rice fields, oil fields, underground pipelines, and some industrial wastewater. Oil companies are showing an increased interest in inhibitors, which, along with slowing down hydrogen sulfide and carbon dioxide corrosion of metal equipment, have a bactericidal effect on SRBs [3, 4].

Organic compounds containing various functional groups and heteroatoms, such as sulfur and nitrogen, along with biological activity, are characterized by antimicrobial activity [5]. Among the biologically active compounds of the cyclohexane series, their amine-containing derivatives stand out, the value of which is associated with the presence of two pharmacophore fragments in their molecules – a nitrogen-containing group and a cyclohexane ring [6, 7].

One of the convenient methods for the synthesis of new generations of functionally substituted aminomethoxy derivatives is the three-component Mannich reaction, since the use of various starting compounds with an active hydrogen atom, as well as amine and aldehyde components, makes it possible to obtain a wide range of polyfunctional derivatives of this class of compounds [8–11]. Mannich bases with a cyclohexane fragment, which have a wide range of properties, also exhibit bactericidal properties [12–16], but their effect on the
growth of sulfate-reducing bacteria (SRBs) has been little studied [17].

**Discussion of the results**

This paper presents the results of the synthesis and study of the properties of amidomethoxy derivatives of cyclohexanol. For this purpose, the Mannich condensation reaction of cyclohexanol (I), formaldehyde (II), and cyclic amines [piperidine (III), morpholine (IV), hexamethyleneimine (V)] was carried out. The reaction proceeds according to the following scheme:

\[
\text{I} + \text{CH}_2=\text{O} + \text{HN} \rightarrow \text{VI-VIII}
\]

X = CH\(_2\) (III, VI), O (IV, VII), CH\(_2\)-CH\(_2\) (V, VIII).

The reaction was carried out at a temperature of 78–80°C for 4–5 h in a benzene solution at an equimolar ratio of the starting components. The yield of the compounds was 69–75%, with the maximum yield obtained using hexamethyleneimine. Target compounds are liquids with a characteristic odor, insoluble in water, and readily soluble in organic solvents. The composition and structure of compounds VI–VIII were established based on elemental analysis, IR, \(^1\)H, and \(^13\)C NMR spectroscopy, and mass spectroscopy.

Thus, in the IR spectra of all synthesized cyclohexanol derivatives (VI–VIII), there is no absorption band in the region of 3334 cm\(^{-1}\) (Figure 1), which is characteristic of the hydroxyl group, and absorption bands are observed in the regions of 1232 and 1024 cm\(^{-1}\), which are related to stretching vibrations R\(_3\)N groups, absorption bands in the regions of 1150, 1134 and 1070 cm\(^{-1}\) refer to the stretching vibrations of the simple ether bond (C–O–C). The \(^1\)H NMR spectra of compounds VI–VIII fully confirm the structure of the synthesized compounds. The protons of the methyl group in the position of the amine fragment give a signal at approximately \(\delta = 0.86\) ppm. in the form of a triplet (Figure 2).

![Fig. 1. IR spectrum of morpholinomethoxycyclohexane (VII).](image-url)
The protons of the methylene groups give signals in the ranges δ = 1.2–1.86 ppm in the form of a multiplet, and the protons of the CH₂–N–CH₂ group, in the regions δ = 2.4–2.78 ppm. The CHO proton of the cyclohexane ring gives signals approximately in the region of δ = 4.07 ppm, in the form of a doublet of doublets. The protons of the OCH₂N group give signals approximately in the regions δ = 4.4 ppm in the form of a doublet of doublets (J = 2.5 Hz, J = 6.5 Hz). The 13C NMR spectra of compounds VI–VIII also confirm the structure of the synthesized compounds (Figure 3).

The bactericidal-inhibiting properties according to OST 39-234-89 were studied at three concentrations (25; 50; 100 mg/l), 1143 strains of Desulfovibrio desulfuricans were used as CBR, the nutrient medium was Postgate B, pH 7.0–7.5, the duration of incubation was thermostat at 30–32°C – 15 days.

Fig. 2. ¹H NMR spectrum of hexamethyleneiminomethoxycyclohexane (VIII).

Fig. 3. ¹³C NMR spectrum of piperidinomethoxycyclohexane (VI).
For comparison, as standards in the same dilutions (25; 50; 100 mg/l), bactericide-inhibitors used in industry were studied: AMDOR IK-7 (10% solution of higher amines \((\text{C}_{10}^{}-\text{C}_{16}^{})\) in an aprotic solvent) and AMDOR IK-10 (a mixture of imidazolines and amidoamines obtained by the interaction of polyethylenopolyamine and oleic acid).

**Experimental part**

The IR spectra of the synthesized compounds were recorded on a BRUKER instrument (ALPHA IR FURYE) in the range of 4000–400 cm\(^{-1}\). \(^1\)H and \(^{13}\)C NMR spectra were recorded on a BRUKER AM-300 spectrometer at a frequency of 300 MHz in 
CDCl\(_3\) solvent, HMDS as the internal standard. Mass spectra were obtained on a VG-7070E mass spectrometer (ionizing voltage 70 eV). The purity of the reaction products was determined from the boiling point, elemental analysis, and gas-liquid chromatography. Elemental analysis was carried out on a CARLOERBA instrument, model EA 1108. GLC analysis was carried out on an LKhM-8 MD chromatograph (Russia), a steel column (300×3 mm) with 5% PEGS on Dinochrome P, carrier gas helium (40 cm\(^3\)/min), detector – katharometer, column temperature 150\(0^\circ\)C, evaporator temperature – 230\(0^\circ\)C. The refractive index \((n_{d}^{20})\) was measured with an ABBEMAT 350/500 refractometer; the relative density \((d_{4}^{20})\) was determined with a pycnometer [18].

Cyclohexanol was used as the starting compound, boiling point 161\(0^\circ\)C. Secondary amines (chemically pure) were distilled before use. Paraform, a formaldehyde polymerization product that depolymerizes to formaldehyde when heated during the reaction, was used in the form of a reactive powder. Benzene was taken as a solvent, which was purified and dried according to a known method [19]. Ammonia water, which is a medical preparation, was used in the form of a 10% solution.

The synthesized compounds (VI–VIII) were studied as bactericide inhibitors against SRB according to the procedure [20]. For this purpose, 1% solutions of compounds VI–VIII in isopropanol was prepared. The bactericidal effect of the reagents is studied mainly by observing for 15 days and calculating the amount of \(\text{H}_2\text{S}\) formed at the end of the experiment. The formation of \(\text{H}_2\text{S}\) is determined by iodometric titration. For comparison, 2 samples without reagent were taken: control 1 and control 2. Control 1 – only Postgate B nutrient medium, control 2 – nutrient medium and SBR cultures. The results of the study are presented in the Table.

<table>
<thead>
<tr>
<th>Reagent</th>
<th>Concentration, c, mg/l</th>
<th>Number of bacteria (number of cells/ml)</th>
<th>Amount of (\text{H}_2\text{S}), mg/l</th>
<th>Bactericidal effect, Z-%</th>
</tr>
</thead>
<tbody>
<tr>
<td>VI</td>
<td>25</td>
<td>10(^{7})</td>
<td>213</td>
<td>16.4</td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>10(^{6})</td>
<td>9.6</td>
<td>96.2</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>VII</td>
<td>25</td>
<td>10(^{6})</td>
<td>162</td>
<td>36.4</td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>–</td>
<td>–</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>–</td>
<td>–</td>
<td>100</td>
</tr>
<tr>
<td>VIII</td>
<td>25</td>
<td>10(^{6})</td>
<td>105</td>
<td>52.7</td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>10(^{6})</td>
<td>8.4</td>
<td>98</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>–</td>
<td>–</td>
<td>100</td>
</tr>
<tr>
<td>АМДОР-ИК-7 (etalon)</td>
<td>25</td>
<td>stimul.</td>
<td>stimul.</td>
<td>stimul.</td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>10(^{6})</td>
<td>88.8</td>
<td>60</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>10(^{6})</td>
<td>55.5</td>
<td>75</td>
</tr>
<tr>
<td>АМДОР-ИК-10 (etalon)</td>
<td>25</td>
<td>stimul.</td>
<td>stimul.</td>
<td>stimul.</td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>10(^{6})</td>
<td>77.7</td>
<td>65</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>10(^{6})</td>
<td>44.4</td>
<td>80</td>
</tr>
<tr>
<td>Control 1</td>
<td>–</td>
<td>–</td>
<td>14.0</td>
<td>–</td>
</tr>
<tr>
<td>Control 2</td>
<td>10(^{8})</td>
<td></td>
<td>222.0</td>
<td>–</td>
</tr>
</tbody>
</table>
SYNTHESIS OF AMINOMETHOXY DERIVATIVES OF CYCLOHEXANOL.....

The table shows that all three samples showed high bactericidal properties. Moreover, 1% solutions of compounds (VI–VIII) at a concentration of 100 mg/l showed a 100% bactericidal effect. A 1% solution of compound (VII) already at a concentration of 50 mg/l exhibits a 100% bactericidal effect. Compounds (VI and VIII) at a concentration of 50 mg/l showed 96.2 and 98% bactericidal effects, respectively. And at a concentration of 25 mg/l, the bactericidal effect of compounds (VI–VIII) was 16.4%, 36.4%, and 58.2%, respectively.

Synthesis of aminomethoxy derivatives of cyclohexanol (VI–VIII). General synthesis method. To a mixture of 0.1 mol of formaldehyde (II) and 0.1 mol of cyclohexanol (I) in benzene, add 0.1 mol of secondary amine (III) and 0.1 mol of benzaldehyde (II) and 0.1 mol of cyclic amines (VIII). General synthesis of cyclohexanol, formaldehyde and cyclic amines was 16.4%, 36.4%, and 58.2%, respectively.

Piperidinomethoxy cyclohexane (VI) was obtained from 10 g (0.1 mol) of formaldehyde, and 8.5 g (0.1 mol) of piperidine (III). The yield of the compound was 14.5 g (73%), b.p. 96–99°C (3 mm Hg), n D 1.4774, d 4 0.9423 g/sm 3. IR spectrum, ν, cm -1: 776, 835 (C–H, CH 2 mathematical vibrations), 1036, 1071, 1130, 1154 (C–O–C), 1023, 1232 (C–N), 1318, 1348, 1366, 1412, 1448, 2852, 2927 (δ C–H, CH, CH 2, CH 3). 1 H NMR spectrum, δ, ppm: 1.15–1.25 m (16H, 8CH 2), 2.5–2.63 m (4H, CH 2–N–CH 2), 4.05 d.d (1H, OCH), 4.20 d (2H, OCH 2N, J 10.5 Hz). 13 C NMR spectrum, δ, ppm: 24.15, 24.18, 25.47, 25.85, 32.52, 35.55, 50.60, 53.03, 69.89, 74.91 (2C), 88.31. Mass spectrum, m/z (I rel, %): 198(10) [M+H] +, 197(9) [M]+, 180(8) [M–OH] +, 165(11) [M–H 2O–CH 2] +, 114(100) [M–C 3H 5N]+, 96(13) [M–C 3H 5N–H 20] +, 81(5), 70(20), 42(6). Found, %: C 74.03; H 11.04; No. 6.94. C 12 H 23 NO. Calculated, %: C 73.04; H 11.75; N 7.10. M 197.18.

Morpholinomethoxy cyclohexane (VII) was obtained from 10 g (0.1 mol) of cyclohexanol, 3.0 g (0.1 mol) of formaldehyde, and 8.7 g (0.1 mol) of morpholine (IV). The yield of the compound was 13.8 g (69%), b.p. 112°C (6 mm Hg), n D 1.479, d 4 1.0061 g/sm 3. IR spectrum, ν, cm -1: 864, 902 (C–H, CH 2 mathematical vibrations), 1016, 1070, 1150 (C–O–C), 1016, 1232 (C–N), 1257, 1281 (C–O–C cycle), 1313, 1360, 1411, 1450, 2851, 2927 (δ C–H, CH, CH 2, CH 3). 1 H NMR spectrum, δ, ppm: 1.2–1.66 m (10H, 5CH 2), 2.45–2.66 m (4H, CH 2–N–CH 2), 3.55–3.80 m (4H, CH 2OCH 2 cycle), 4.01 d. d (1H, OCH), 4.7 d.d (2H, OCH 2N, J 2.5, J 6.4 Hz). 13 C NMR spectrum, δ, ppm: 24.15, 26.4, 28.50, 30.50, 32.3, 52.4, 55.6 (2C), 66.7, 76.0, 84.10. Mass-spectrum, m/z (I rel, %): 200(8) [M+H] +, 199(7) [M]+, 182(13) [M–OH] +, 168(20) [M–OH–CH 2]+, 131(21) [M–C 3H 5N]+, 100(10) [M–C 3H 5NO]+, 74(100) [M–C 3H 5O–H 20]+, 95(25), 75(54). Found, %: C 67.03; H 10.14; N 7.34. C 11 H 21 NO 2 Calculated, %: C 66.29; H 10.62; N 7.03. M 199.16.

Hexamethyleneaminomethoxy cyclohexane (VIII) was obtained from 10 g (0.1 mol) of cyclohexanol, 3.0 g (0.1 mol) of formaldehyde, and 9.9 g (0.1 mol) of hexamethylenimine (V). The yield of the compound was 15.9 g (75%), b.p. 109°C (2 mm Hg), n D 1.485, d 4 0.9478 g/sm 3. IR spectrum, ν, cm -1: 887, 944, 984 (C–H, CH 2 mathematical vibrations), 1035, 1070, 1134 (C–O–C), 1023, 1183, 1236 (C–N), 1341, 1357, 1411, 1451, 2850, 2920 (δ C–H, CH, CH 2, CH 3). 1 H NMR spectrum, δ, ppm: 1.15–1.78 (1H, OCH), 2.63–2.78 m (4H, CH 2–N–CH 2), 4.14 d.d (1H, OCH), 4.7 d.d (2H, OCH 2N, J 2.5, J 6.4 Hz). 13 C NMR spectrum, δ, ppm: 24.4, 27.32, 27.54, 28.20, 29.75, 29.24, 29.45, 32.1, 52.41, 53.32 (2C), 76.0, 83.17. Mass-spectrum, m/z (I rel, %): 211(11) [M]+, 194(12) [M–OH] +, 180(21) [M–OH–CH 2]+, 128(17) [M–C 3H 5N]+, 113(51) [M–C 3H 6O]+, 99(31) [C 6H 11O]+, 92(100) [C 5H 9O]+, 74(27) [M–C 3H 5O–H 20]+, 81(6), 42(6). Found, %: C 73.03; H 12.14; N 7.04. C 13 H 25 NO. Calculated, %: C 73.88; H 11.92; No. 6.63. M 211.19.

Conclusions

Three-component Mannich reaction of cyclohexanol, formaldehyde and cyclic amines
was used to obtain aminomethoxy derivatives of cyclohexanol. The reaction was carried out at a temperature of 78–80°C for 4–5 h in a benzene solution at an equimolar ratio of the starting components. The yield of compounds was 69–75%. Their physicochemical properties were determined, and the composition and structure of the obtained compounds were confirmed using the data of elemental analysis, IR, 1H, 13C NMR spectroscopy and mass spectrometry.

The effect of the synthesized compounds on SRBs of the "Desulfovibrio desulfuricans" type was tested at three concentrations (25, 50, 100 mg/l). Based on the amount of hydrogen sulfide formed, the bactericidal effect of the presented samples was calculated. It has been established that 1% solutions of aminomethoxy derivatives of cyclohexanol exhibit high bactericidal activity, and they act on bacteria at very low concentrations compared to the industrially used bactericide-inhibitors AMDOR IK-7 and AMDOR IK-10 taken as a standard.

Acknowledgments

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TSIKLOHEKSANOLIN AMINOMETOKSI TÖRƏMƏLƏRİNİN SİNTEZI VƏ INHİBİTOR-BAKTERİSID XASSƏLƏRİNİN ÖVRƏNILMƏSİ


Tsiiklohexsanol, tsiiklik aminlar (piperidin, morfolin, heksametilenimin) və formaldehid asasında yəni Mannix aşılarını sintez olunmuşdur. Reaksiya 78–80°C temperaturada 4–5 saat müddətində, ildən maddələrin evkəmololar nəzərdən belə bənzo mehulunda aparilmışdır. Birlişmələrin əhlətə 69–75%-dir. Sintezi olunmuş birləşmələrin fiziki-kimiyevi gostəriciləri tayin edilmişdir. Məqsədli mehulların tərkib və quruğuluş element analizi, İQ, 1H və 13C NMR spektroskopiyası, eləcə də külə spektrometriyə səsərli ilə təsdiqlənmişdir. Onların üç qatılıqda (25; 50; 100 ml/q) "Desulfovibrio desulfuricans" təpili sulfatredaksiyaedici baktəriyalara çox fəaliyyətə təsir yoxlanılmışdır. Alınmış birləşmələr yüksək baktərisid xassı göstərməmişdir, bu birləşmələrin izopropil spirtində 1%-li mehulları 100 ml/q qatılıqda 100% baktərisid təsir göstərməmişdir. Morfolin frəqmentli birlişmin 1%-li mehulları 50 ml/q qatılıqda 100% baktərisid təsir göstərmişdir. Piperidin və heksametilenimin frəqmentli salxayən birləşmələr 50 ml/q qatılıqda məvəfiq olaq 96.2 və 98% baktərisid təsir göstərmişdir. 25 ml/q qatılıqda isə hər üç birləşminin baktəriyalar təsir məvəfiq olaq 16.4%, 36.4% və 58.2% təskil etmişdir. Alınmış mətbəçələr göstərdir ki, sintez edilmiş birləşmələr sonayədə istifadə olunan standart olaraq qabul edilən AMİDOR İK-7 və AMİDOR İK-10 baktərisid inhibitornun məxsusiyətdə daha çox baktərisid xüsusiyyətlər göstərmişdir. Tsiiklohexsanolun aminometoksi törəmlərinin çox aşağı qatılıqlarda baktəriyalara təsirini nazarə alaraq, onlar sulfatredaksiyaedici baktəriyalara qarşı effektli inhibitör kimi təkflif edilə bilər.

Açar sözər: tsiiklohexsanol, ikili aminlar, Mannix aşıları, sulfatredaksiyaedici baktəriyalar, inhibitor-baktərisidik, biokorroziya.

SYNTHESIS OF AMINOMETHOXY DERIVATIVES OF CYCLOHEXANOL.....

СИНТЕЗ АМИНОМЕТОКСИПРОИЗВОДНЫХ ЦИКЛОГЕКСАНОЛА И ИЗУЧЕНИЕ ИХ В КАЧЕСТВЕ ИНГИБИТОР-БАКТЕРИЦИДОВ

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На основе циклогексанола, циклических аминов (пиперидин, морфолин, гексаметилимин) и формальдегида синтезированы новые основания Манина. Реакция проводилась при температуре 78–80°C в течение 4–5 ч, в растворе бензола при эквивалентном соотношении исходных компонентов. Выход соединений составил 69–75%. Определены физико-химические данные синтезированных соединений. Состав и строение целевых продуктов подтверждены методами элементного анализа, ИК, 1H и 13C ЯМР спектроскопии, а также масс-спектрометрии. Исследовано влияние их на жизнедеятельность сульфатвосстанавливающих бактерий типа “Desulfovibrio desulfuricans” в трех концентрациях (25; 50; 100 мг/л). Полученные соединения проявили высокие бактерицидные свойства, 1%-ые растворы этих соединений в изопропиловом спирте при концентрации 100 мг/л проявили 100%-ный бактерицидный эффект. 1%-ный раствор соединения с фрагментом морфолина уже при концентрации 50 мг/л проявляет 100%-ный бактерицидный эффект. Соединения с фрагментом пиперидина и гексаметилиминами при концентрации 50 мг/л проявили 96.2 и 98%-ный бактерицидный эффект, соответственно. А при концентрации 25 мг/л бактерицидный эффект всех трех соединений составил 16.4%, 36.4% и 58.2%, соответственно. По полученным результатам видно, что синтезированные соединения проявили более высокие бактерицидные свойства по сравнению с ветвистыми в качестве эталона промышленно используемыми бактерицид-ингибиторами AMİDOR İK-7 и AMİDOR İK-10. Учитывая то, что указанные аминометокси-производные циклогексанол влияют на бактерии при очень низких концентрациях, их можно предложить в качестве эффективных ингибиторов против сульфатвосстанавливающих бактерий.

Ключевые слова: циклогексанол, вторичные аммины, основания Манина, сульфатвосстанавливающие бактерии, ингибитор-бактерицид, биокоррозия.