EXPERIMENTAL BIOLOGICAL

The antimicrobial activity was determined by the method of double serial dilution in Khottinger bouillon at pH 7.2 against the test microbes St. aureus 209P, E. coli M-17, Pr. vulgaris 39, Ps. aeruginosa 165, and C. albicans 42.

The antiphagal activity of the materials was determined in the phage-bacteria systems against DNK (T_6)- and RNK(MS-2)-containing phages. The indicator cultures were E. coli B and E. coli Hfre, respectively. The antiphagal activity was studied by the agar gel method to determine the percent activation by the method of [1]. The materials were dissolved in DMSO and diluted with sterile distilled water to the desired concentration.

LITERATURE CITED

1. D. M. Gol'dfarb, Bacteriophages [in Russian], Moscow (1961), p. 125.
2. FRG Patent 3242519. Ref. Zh. Khim., No. 3-0-453P (1985).
3. Japanese Patent 59-31761. Ref. Zh. Khim., No. 7-0-466P (1985).
4. V. G. Kharchenko, S. N. Chalaya, O. V. Litvinov, et al., Zh. Org. Khim., No. 6, 1208-1211 (1984).
5. V. G. Kharchenko, S. N. Chalaya, and O. V. Litvinov, Khim. Geterotsikl. Soedin., No. 3, 352-354 (1985).

SYNTHESIS AND ANTIMICROBIAL ACTIVITY OF BIS[5-INDOLYL]METHANES AND BIS[5-INDOLYL] OXIDES

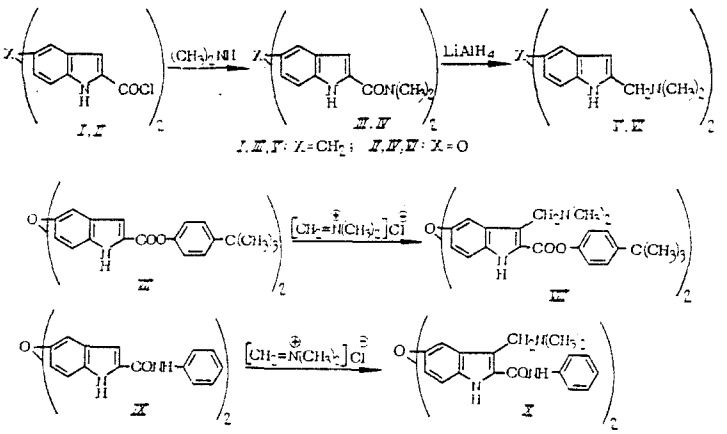
Sh. A. Samsoniya, Z. Sh. Lomtatidze, N. N. Ovsyannikova, and N. N. Suvorov

UDC 615.281/282:547.759.1].012.1

We have reported previously [3-5] the curariform activity of some bisindolyl quaternary ammonia compounds obtained by us.

Continuing this study, we examined the antimicrobial activity of some bis(5-indolyl)-methanes [1] and bis(5-indolyl) oxides [2].

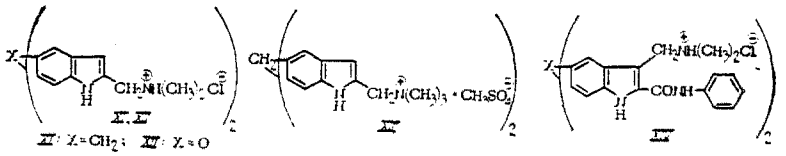
For this purpose, we have synthesized the novel bisisogramine 2,2'-bis(dimethylamino-methyl)bis(5-indolyl) oxide (VI) and the bisisogramine (V) previously obtained by us [1], as follows:



Aminolysis of the diacid chlorides of 2,2'-dicarboxybis(5-indolyl)methane (I) and the corresponding oxide (II) was carried out in an aqueous dioxane solution of dimethylamine, subsequent reduction of the amides (III) and (IV) being carried out with lithium alumino-hydride [4]. Also synthesized were the biogramines (VIII) and (X), by reacting 2,2'-di(p-tert-butylphenoxycarbonyl)bis(5-indolyl) oxide (VII) and 2,2'-di(phenylaminocarbonyl)bis(5-

Tbilisi University. Translated from Khimiko-farmatsevticheskii Zhurnal, Vol. 21, No. 7, pp. 827-829, July, 1987. Original article submitted October 15, 1985.

indolyl) oxide (IX) with the Mannich reagent $[CH_2N(CH_3)_2]Cl$. The structures of the products were confirmed by IR, UV, and PMR spectroscopy. For the examination of their antimicrobial properties, compounds (V), (VI), and (X) were converted into their dihydrochlorides (XI), (XII), and (XIV), and (VI) into its bismethylsulfate (XIII).



EXPERIMENTAL CHEMICAL

The progress of the reactions was followed and the purity of the products established by TLC on Silufol UV-254 plates (Czech SSR). IR spectra were obtained on a UR-20 instrument (East Germany) in KBr disks, and UV spectra on a Specord spectrophotometer in THF. PMR spectra were recorded on a Varian CFT-20 spectrometer (USA), internal standard TMS. The precision of measurement of the chemical shifts was ±0.2 ppm, and of the coupling constants ±0.1 Hz.

2,2'-Di(dimethylaminocarbonyl)bis(5-indolyl)methane (III). Obtained as described in [4]. Yield of (III) 67%, mp 315-317°C (lit. mp 315-317°C).

2,2'-Di(dimethylaminocarbonyl)bis(5-indolyl) Oxide (IV). Obtained as for (III) [4], from 0.75 g (2 mmole) of the diacid chloride (II). Yield of (IV) 0.72 g (92%), mp 337-338°C, R_f 0.41 (benzene-acetone, 1:2). IR spectrum, ν, cm^{-1}: 3400, 3280 (NH), 1610 (C=O). UV spectrum, λ_{max}, nm ((log ε): 227 (4.39), 301 (4.32). Found, %: C 67.44; H 5.28; N 14.73. $C_{22}H_{22}N_4O_3$. Calculated, %: C 67.69; H 5.64; N 14.35.

2,2'-Bis(dimethylaminomethyl)bis(5-indolyl)methane (V). Obtained by the method described in [4]. Yield of (V) 95%, mp 101-102°C (lit. mp 101-102°C).

2,2'-Bis(dimethylaminomethyl)bis(5-indolyl) Oxide (VI). Obtained as for (V) [4], by reducing 0.38 g (10 mmole) of the amide (IV) with $LiAlH_4$. Yield of (VI) 0.25 g (35%), mp 170-171°C, R_f 0.48 (isopropanol 33%—aqueous ammonia, 5:1). IR spectrum, ν, cm^{-1}: 3410 (NH), 1470, 1440, 1420 (aliphatic CH). UV spectrum, λ_{max}, nm (log ε): 227 (4.72), 279 (4.03). Found, %: C 72.55; H 7.64; N 15.50. $C_{22}H_{26}N_4O$. Calculated, %: C 72.82; H 7.18; N 15.46.

2,2'-Di(p-tert-butylphenoxycarbonyl)-3,3-di(dimethylaminomethyl)bis(5-indolyl) Oxide (VIII). To a solution of 0.6 g (1 mmole) of (VII) in 10 ml of dry DMF was added 0.47 g (5 mmole) of N,N-dimethylmethyleneimmonium chloride. The mixture was stirred for 1 h at room temperature, kept for 2 h at 60-70°C, cooled, poured into 20 ml of water, and basified with NaOH to pH 10.0. The colorless crystals which separated were filtered off, washed with water to pH 7.0, and dried in vacuo over KOH. Yield of (VIII) 0.63 g (88%), mp 205-207°C, R_f 0.73 (isopropanol—NH_4OH, 100:1). IR spectrum, ν, cm^{-1}: 3420, 3360 pl (NH), 1740 (C=O). UV spectrum, λ_{max}, nm (log ε):232 (4.66), 307 (4.65). Found, %: C 73.73; H 7.47; N 8.19. $C_{44}H_{50}N_4O_5$. Calculated, %: C 73.94; H 7.00; N 7.84.

2,2'-Di(phenylaminocarbonyl)-3,3'-di(dimethylaminomethyl)bis(5-indolyl) Oxide (X). Obtained as for (VIII), from 0.49 g (1 mmole) of (IX). Yield of (X) 0.54 g (90%), mp 243-245°C. R_f 0.71 (isopropanol—NH_4OH, 500:1). IR spectrum, ν, cm^{-1}: 3430, 3280 (NH), 1660, 1630 (C=O), 1570, 1530 (amide II). UV spectrum, λ_{max}, nm (log ε):227 (4.73), 320 (4.85). Found, %: C 72.36; H 6.36; N 14.18. $C_{36}H_{36}N_6O_3$. Calculated, %: C 72.00; H 6.00; N 14.00.

2,2'-Di(dimethylaminomethyl)bis(5-indolyl)methane Dihydrochloride (XI). Obtained as described in [4]. Yield of (XI) 93%, mp 224-225°C (lit. mp, 224-225°C).

2,2'-Di(dimethylaminomethyl)bis(5-indolyl) Oxide Dihydrochloride (XII). Obtained as for (XI) [4], from (VI). Yield of (XII) 0.39 g (90%), mp 134-136°C (decomp.). IR spectrum, ν, cm^{-1}: 3400, 3250 (NH), 2650, 2470 (NH). Found, %: C 60.49; H 6.19; Cl 16.22; N 12.69. $C_{22}H_{28}Cl_2N_4O$. Calculated, %: C 60.69; H 6.44; Cl 16.32; N 12.87.

2,2-Di(dimethylaminomethyl)bis(5-indolyl)methane Bismethylsulfate (XIII). Obtained as described in [4]. Yield of (XIII) 94%, mp 196-198°C (lit. mp 196-198°C).

2,2'-Di(phenylaminocarbonyl)-3,3-di(dimethylaminomethyl)bis(5-indolyl) Oxide Dihydrochloride (XIV). Obtained as for (XI) [4], from (X). Yield of (XIV) 0.59 g (88%), mp 206-

207°C (decomp.). IR spectrum, ν, cm^{-1}: 3430 (NH), 2700 ($\overset{+}{N}H$), 1670 pl., 1640 (C=O), 1570 (amide II). Found, %: C 63.97; H 5.59; Cl 10.12; N 12.11. $C_{36}H_{38}Cl_2N_6O_3$. Calculated, %: C 64.19; H 5.64; Cl 10.55; N 12.48.

EXPERIMENTAL BIOLOGICAL

Compounds (VIII) and (XI-XIV) were examined for antimicrobial activity.

The test organisms were the following strains of phytopathogenic bacteria (the cultures were obtained from the All-Union Research Institute for Chemical Plant Protection): Bacterium tumefaciens (canker of grapevine), Xanthomonas campestris, Pectobacterium aroideae (causes bacteriosis in white-headed cabbage, and rot), and the actinomycetes (cultures obtained from the Institute of Microbiology, Academy of Sciences of the USSR), Actinomycetes streptomycini, Actinomyces lavendulae, Streptomyces spp., Nocardiopsis spp., and the fungus Aspergillus niger.

The test microorganisms were cultured on the following solid nutrient media: for the phytopathogenic bacteria, Brucholter's medium (potato broth 1 liter, pentane 5 g, Na_2HPO_4 2 g, glucose 6 g, NaCl 2 g, sodium citrate 1 g, asparagine 1 g, agar 20 g, distilled water 1 liter), and for the actinomycetes, synthetic medium No. 1 (KNO_3 1 g, K_2PO_4 0.5 g, $MgSO_4$ 0.5 g, NaCl 0.5 g, $FeSO_4$ trace, $CaCO_3$ 1 g, starch 20 g, agar 20 g, and tap water 1 liter); the fungus was cultured on wort agar (wort 0.5 liter, agar 20 g, tap water 0.5 liter).

The antimicrobial activity of the compounds was measured by the method of holes in the thickness of agar. The agarized media were poured into Petri dishes, and inoculated with the test organisms (density of inoculation, 2-2.5 million cells/ml). The inoculum was obtained in the case of bacteria and the fungus after incubation for 24 h, and of the actinomycetes after 56 h. Holes of diameter 8 mm were cut in the thickness of the agar plates (five holes each). In some of the holes were placed a solution of the test compound in concentrations of 1 and 0.1 g/liter and in others the solvent used for these compounds (ethanol) as control.

The Petri dishes were placed in a thermostat at 28°C. The results were assessed after incubation for 24 h for the phytopathogenic bacteria and the fungus, and after 96 h for the actinomycetes.

The results showed that the test compounds possess inhibitory properties. Comparisons of the suppression zones of the test compounds showed (XI) and (XIII) to be more active than (VIII) and (XIV), while (XII) showed no antimicrobial actvity. It is noteworthy that treatment with (VIII) and (XIV) gave different sizes of zones of growth suppression of the actinomycetes. Suppression of the growth of the phytopathogenic bacteria was observed at all concentrations of the test compounds.

The experimental findings showed that (VIII), (XII), and (XIV) were without effect on the growth of Aspergillus niger.

Examination of the results leads to the conclusion that (VIII) and (XIV) suppress the growth of Bact. tumefaciens, Pectobact. aroideae, Xanthomonas campestris, Actinomyces streptomycini and A. lavendulae.

The relatively high activity of (XI) is evidently due to the presence therein of the group $CH_3SO_4^-$.

LITERATURE CITED

1. Sh. A. Samsoniya, I. Sh. Chikvaidze, N. N. Suvorov, and I. M. Gverdtsiteli, Soobshch. Akad. Nauk GSSR, 91, No. 3, 609-612 (1978).
2. Sh. A. Samsoniya, D. M. Tanidze, and N. N. Suvorov, Khim. Geterotsikl. Soedin., No. 1, 57-61 (1981).
3. Sh. A. Samsoniya, B. A. Medvedev, D. O. Kadzhrishvili, et al., Khim.-farm. Zh., No. 11, 55-58 (1982).
4. Sh. A. Samsoniya, B. A. Medvedev, S. V. Dolidze, et al., ibid., No. 1, 29-31 (1984).
5. I. Sh. Chikvaidze, B. A. Medvedev, Sh. A. Samsoniya, et al., ibid., No. 5, 36-39 (1980).