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SYNTHESIS, CHARACTERIZATION AND ANTIMICROBIAL STUDIES OF 6-PHENOTHIAZINYL-1,8-NAFTHOYLENE-1′,2′-BENZIMIDAZOLE-1-ONE

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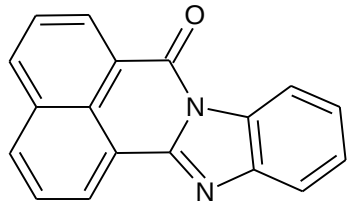
Abstract: *The subject of the present work is the synthesis of isomeric luminescent compounds of orange-red color, containing a phenothiazine ring and a 1,8-naphthoylene-1',2'-benzimidazole group. The IR and NMR spectra of the individual isomers, their separation from the reaction mixture and their antimicrobial activity have been studied.*

Keywords*: phenothiazine, synthesis, antimicrobial activity.*

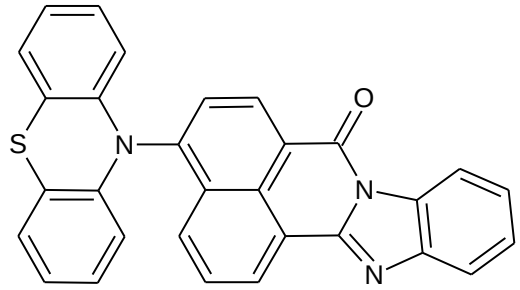
INTRODUCTION

1,8-Naphthoylene-1′,2′-benzimidazole (I) and many of its substituted derivatives (in the naphthalene and benzene nuclei) are effective luminophores in the yellow and yellow-green regions (Krasovitskiy B. M., Levchenko N. F. & Makarenko Yu. I., 1966; Krasovitskiy B. M., Shevchenko Э. A. & Pereyaslova D. G., 1965; Shevchenko Э. A., Distanov V. B. & Bogdanova L. I., 1984; Krasovitskiy B. M., Shevchenko Э. A. & Distanov V. B., 1983).

By combining in one molecule the 1,8-naphthoylene-1′,2′-benzimidazole group with that of phenothiazine, we obtained orange-red luminophores (II).

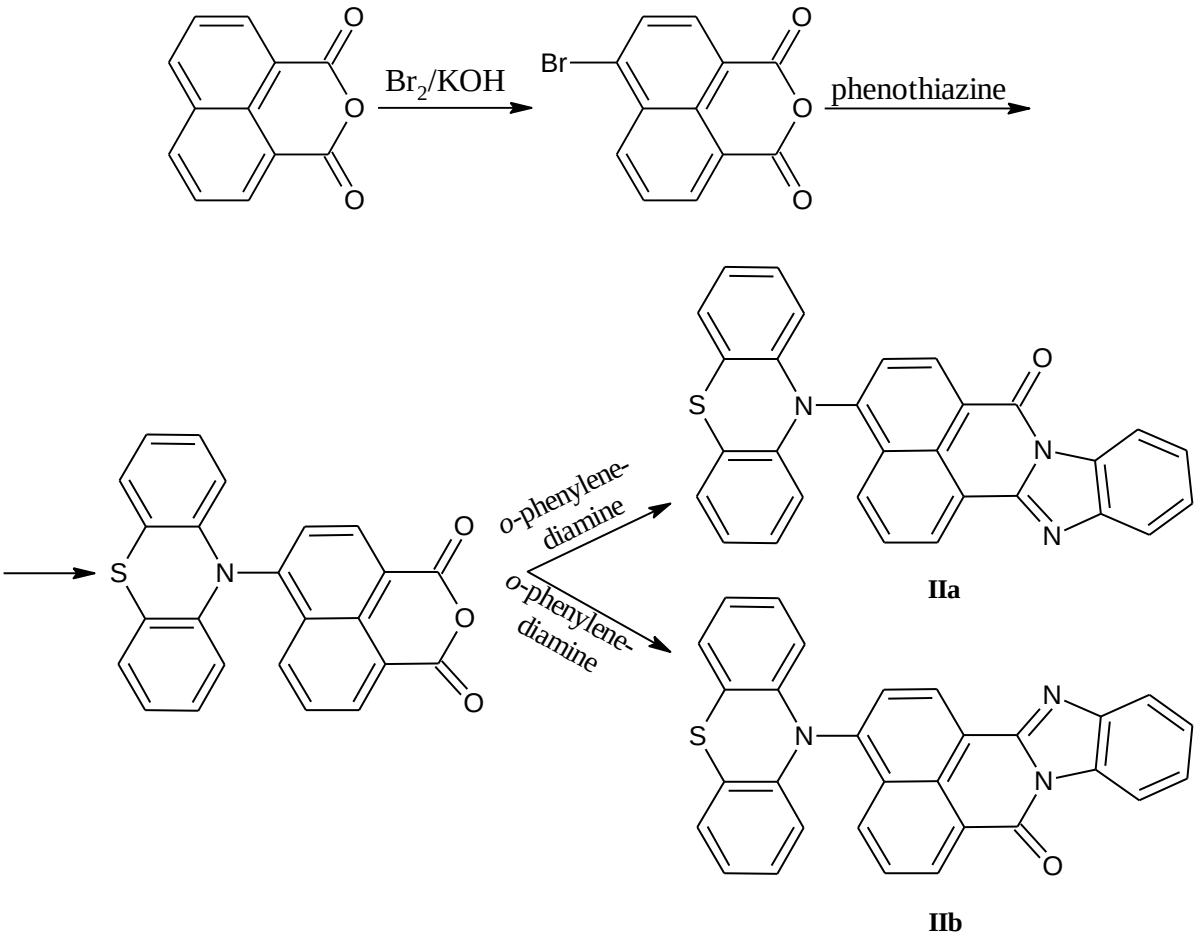


I



II

The scheme of synthesis of luminophores (II) can be presented as follows:



Two isomeric products IIa and IIb were obtained.

Phenothiazines, nitrogen- and sulfur-containing tricyclic compounds have been known for more than a hundred years. The phenothiazine nucleus (10*H*-dibenzo-1,4-thiazine) had been proven by Bernthsen in 1883. Since then, more than 5000 phenothiazine derivatives have been synthesized. This class of organic compounds could be extremely important due to its significant biological and chemical properties. Phenothiazines demonstrate good antimicrobial, antiprion, anthelmintic and insecticidal activities (Pluta, K, et al., 2011, Wainwright, M., et al., 2012).

EXPOSITION

Materials and methods

All used chemicals were purchased from Merck and Sigma-Aldrich. The melting points were determined by a SMP-10 digital melting point apparatus. The IR spectra were taken on Perkin-Elmer FTIR-1600 spectrometer in KBr discs. The NMR spectra were obtained on Bruker Avance III HD (500.13 MHz for ^{1}H and 125 MHz for ^{13}C NMR) spectrometer. The chemical shifts are given in parts per million (δ) relative to tetramethylsilane as internal standard for spectra in $CDCl_3$ solutions.

Synthesis of 6-(10*H*-phenothiazine-10-yl)-1*H*,3*H*-benzo[*de*]-isochromen-1,3-dione derivative of 4-aminophenylacetic acid

6-Phenothiazinyl-1,8-naphthalene anhydride (2.76 g/0.007 mol) and 4-aminophenylacetic acid (1.2 g/0.008 mol) in 20 mL of glacial acetic acid are refluxed for 4 hours. After cooling, the mixture is poured into 200 mL of cold water and allowed to stay for 24 hours. This is followed by filtration and recrystallization with absolute ethanol.

Condensation of 6-phenothiazinyl-1,8-naphthalic anhydride with *o*-phenylenediamine

34.2 g of 6-phenothiazinyl-1,8-naphthalic anhydride, 23.1 g of *o*-phenylenediamine and 350 mL of 1% sulfuric acid are refluxed for 8 hours. The precipitate is then filtered off, washed with water and dried.

Separation of isomers

40 g of the isomeric mixture is dissolved in 400 mL of ethanol and 80 mL of 15% potassium hydroxide and stirred while heating in a water bath for 6 hours. The mixture is cooled down to room temperature, and the undissolved residue IIa is filtered and washed with 150 mL of cold ethanol and 300 mL of water, then dried and recrystallized from glacial acetic acid.

The filtrate is diluted with four times the amount of water. The precipitate from IIb is filtered off, washed with 150-200 ml of water and dried. The compound is eluted with benzene on a packed column Al_2O_3.

Antimicrobial activity of isomers

The antimicrobial activity of the obtained isomers was determined by the method of diffusion in agar and test microorganisms: Gram-positive bacteria *Staphylococcus aureus* ATCC 6538, *Bacillus subtilis* ATCC 6633, Gram-negative bacteria *Escherichia coli* ATCC 8739, *Pseudomonas aeruginosa* ATCC 9027 and *Salmonella abony* NTCC 6017, yeast *Candida albicans* ATCC 10231, *Saccharomyces cerevisiae* ATCC 2601, molds *Fusarium moniliforme* and *Aspergillus brasiliensis* ATCC 16404. A 1% solution in solvent dimethyl sulfoxide (DMSO) was prepared from the isomers. The experiments were performed on nutrient medium Tryptic soy agar (Merck) - for bacteria, and Sabouraud dextrose agar (Merck) for yeast and molds. Agar media were melted in a Koch apparatus. They were cooled down to 50 - 48°C and inoculated with 1% of the pre-prepared suspensions of the test micro-organisms. 20 mL of inoculated media were poured into sterile petri dishes (∅ = 90 mm). The agar was left to solidify. A cork borer was used to punch holes (∅ = 8 mm) in the agar. 50µl of the pre-prepared solutions were added dropwise to each hole and, after 30 minutes of pre-infusion at room temperature, the petri dishes were placed in a thermostat at 37°C for 24 hours for the bacteria; at 28°C for 48 h for yeast and for 72 h for mold fungi (Balouiri M., Sadiki M. & Koraichi Ibnsouda S., 2016). After cultivation, the diameters of the zones of growth inhibition were measured in mm, as: up to 15 mm the microbial culture was weakly sensitive; from 15 to 25 mm - sensitive and over 25 mm - highly sensitive. The experiments were performed in parallel with the control sample of the solvent, taking into account its action as well. The data on antimicrobial activity were arithmetic average of three measurements.

RESULTS AND DISCUSSION

Physicochemical characteristics and spectral data of the isomer IIa

-Yield: 15 g (37%);
-M. p.: 213-214°C;
-IR (KBr, cm^{-1}): 1762 (C=O);
-^{1}H NMR ($CDCl_3$, δ, ppm): 6.79-8.39 (m, 13H, CH);
-^{13}C NMR ($CDCl_3$, δ, ppm): 166.5 (C=O).

Physicochemical characteristics and spectral data of the isomer IIb

-Yield: 10.5g (26%);
-M. p.: 186-187°C;
-IR (KBr, cm^{-1}): 1762 (C=O);
-^{1}H NMR ($CDCl_3$, δ, ppm): 6.71-8.23 (m, 13H, CH);
-^{13}C NMR ($CDCl_3$, δ, ppm): 163.8 (C=O).

Antimicrobial action of the isomers IIa and IIb

The studied isomers do not reveal any antibacterial activity against Gram-positive bacteria *S. aureus* and *B. subtilis* and Gram-negative bacteria *E. coli*, *P. aeruginosa* and *S. abony*. The studied moulds ***A**. brasiliensis* and *F. moniliforme* and yeast *S. cerevisiae и C. albicans* are not sensitive to compounds IIa and IIb.

CONCLUSION

Isomeric orange-red luminescent compounds containing a phenothiazine ring and a 1,8-naphthoylene-1′,2′-benzimidazole group have been synthesized, and the resulting compounds identified. The synthesized isomers do not show any antimicrobial activity.

Acknowledgments

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