Phenanthroline derivatives with potential practical applications

Ph.D. Thesis

Summary

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With the completion of this stage in my life, I would like to say a few words of thanks to those who have guided me and have given me full support during the completion of this PhD thesis. First, I would like to thank my PhD supervisor, Prof. dr. Ionel Mangalagiu for his permanent guidance, support and encouragement throughout the preparation period and drafting of this doctoral thesis.

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The work is accompanied by 290 bibliographical references. The summary includes a review of results obtained during the PhD research stage, general conclusions and selective bibliography. The experimental results are the subject of five scientific articles published in journals with impact factor.
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Introduction

Research undertaken in the field of heterocyclic chemistry has developed tremendously in the last half of the 20th century, due to the particular implications of heterocycles in natural processes, which gives them a great potential for practical applications. Heterocycles, being an integral part of such as life molecules, are ubiquitous in biological systems, and are crucial players in many biological functions due to their ability to form various noncovalent interactions with ions and neutral species. Among them, phenanthroline derivatives attracted attention in the last years especially due to their biological effects, as well as materials science applications, crystal engineering, their unique π-electrons delocalization and complexation properties. Phenanthrolines polycyclic skeletons are also present in sterols, sex hormones, cardiac glycosides, bile acids and morphine alkaloids.

Compared to 1,10-phenanthroline, which has been widely studied for both synthesis and application purposes (especially complexation properties), 1,7-phenanthroline and 4,7-phenanthrolines received much less interest due to difficulties in their synthesis. Therefore, the aim of this thesis is the synthesis of new derivatives containing a 1,7- and 4,7-phenanthroline skeleton and the study of their biological, electrical and fluorescence properties. Continuing the tradition of the Organic Chemistry Department of “Alexandru Ioan Cuza” University of Iasi in the field of cycloimmonium ylides chemistry, we will use, as key intermediates in the synthesis of the target compounds, ylides derived from monoquaternary salts of 1,7 and 4,7-phenanthrolinium. Thus, the in situ generated ylides will be used as 1,3-dipoles in 3+2 cycloaddition reactions to different dipolarophiles, in order to obtain new fused heterocyclic compounds.

All syntheses also include the optimization of chemical reactions and product purification, as well as the spectral and physicochemical characterization of the obtained compounds. In the cases where we were able to obtain monocrystals suitable for X ray diffraction (XRD), the compounds were also characterized using XRD.

Based on the above considerations, the objectives of this thesis are the following:

- O1. Synthesis of the precursor heterocycles: 1,7- and 4,7-phenanthroline;
- O2. Synthesis of new monoquaternary and diquaternary salts derived from 1,7-phenanthroline;
• O3. Synthesis of new monoquaternary and diquaternary salts derived from 4,7-phenanthroline;
• O4. Synthesis of pyrrolo[1,2-i][1,7]phenanthroline derivatives;
• O6. Synthesis of imidazo[1,2-i][1,7]phenanthroline derivatives;
• O8. Evaluation of anticancer / antimycobacterial activity of selected compounds and performing structure-activity relationship (SAR) studies;
• O9. Investigation of the electrical properties of thin films of selected new compounds;
• O10. Study the fluorescence properties of selected new compounds;
• O11. To study the practical applicability of newly synthesized compounds (we consider the possibility of using some of the synthesized compounds with interesting properties as semiconductors, fluorescent markers or drugs).

During our study, due to the unexpected outcomes of some reactions, which brought up some new ideas, we expanded our goal, adding several new objectives:
• O12. Study of the reaction of other cycloimmonium ylides and ethyl cyanoformate.
• O13. Study of some 3+2 cycloaddition reactions using 1,10-phenanthrolinium ylides.

The thesis is structured in two parts:

Part I. Theoretical considerations (Chapter I)

Part II. The original results containing: results and discussion, the experimental part, conclusions and references. (Chapters II and III)
II.2. Synthesis of new monoquaternary salts of 1,7- and 4,7-phenanthrolin-ium

II.2.2. Synthesis of new monoquaternary salts of 4,7-phenanthrolin-ium

In order to accomplish our objectives, we first synthesized the phenanthrolin-ium monoquaternary salts 5a-h. The reaction pathway adopted for the synthesis of phenanthroline derivatives is straightforward and efficient, involving an N-alkylation reaction of 1,7-phenanthroline (Scheme II.4).

Our initial goal was to obtain both the phenanthrolin-ium monoquaternary salts (5a-h) and diquaternary salts (6a-h). Regardless of the conditions employed in terms of molar ratio (1,7-phenanthroline: reactive halide; 1:2.2; 1:5 or 1:7) and reaction conditions (solvents, temperature), only phenanthrolin-ium mono salts, 5a-h were obtained.

![Scheme II.4. Reaction pathway to obtain the 4,7-phenanthrolin-ium salts](image)

II.3. Synthesis of new fused indolizines with 1,7- and 4,7-phenanthroline skeleton using 3+2 cycloaddition of N-ylides corresponding to salts 3a-h and 5a-h to symmetrical and unsymmetrical substituted activated alkynes

II.3.1. 3+2 Cycloaddition reactions to symmetrically substituted reactive alkynes

II.3.1.1. 3+2 Dipolar cycloaddition reactions of 1,7-phenanthrolin-ium N-ylides to symmetrically substituted reactive alkynes

1,7-Phenanthrolin-ium ylides generated in situ from the corresponding cycloimmonium salts (3a-h), in basic medium (Et\(_3\)N), react as 1,3-dipoles with dimethylacetylene dicarboxylate (DMAD) as dipolarophile via 3+2 dipolar cycloaddition reactions.

The reactions took place in dichloromethane at room temperature and gave moderate yields of pyrrolo[1,2-i][1,7]phenanthroline derivatives 9a-h (Scheme II.5). The reaction of ylide 2h and DMAD gave a mixture consisting of 8,9-dihydropyrrolo[1,2-i][1,7]phenanthroline...
derivative 10h and aromatized compound 9h, while from the reaction of ylide 2g only the 8,9-dihydropyrrolo[1,2-i][1,7]phenanthroline derivative resulted. In all reactions we assume that at first, unisolable 9,15-dihydropyrrolo[1,2-i][1,7]phenanthroline intermediates 8a-h are formed, and then an oxidative aromatization takes place, yielding policyclic indolizine compounds 9a-h. The formation of dihydroderivative 10h from 8h occurs probably due to the excess of triethylamine, which induces a stereo- and regioselective prototropic rearrangement, leading to the thermodynamically more stable product 10h.

Scheme II.5. Synthesis of pyrrolo[1,2-i][1,7]phenanthroline derivatives by 3+2 dipolar cycloaddition of in situ generated ylides with DMAD

Figure II.10. X-ray crystal structure of compound 9a with thermal ellipsoids at 50% probability level
II.3.2. 3+2 Cycloaddition reactions to unsymmetrically substituted reactive alkynes

II.3.2.1. 3+2 Dipolar cycloaddition reactions of 1,7-phenanthroline N-ylides to unsymmetrically substituted reactive alkynes

The next goal was to study the cycloaddition reaction of 1,7-phenanthroline salts to ethyl propiolate (EP). Thus, dipolar cycloaddition of the ylides derived from salts 3a-h in dichloromethane at room temperature to EP gave moderate yields of pyrrolo[1,2-i][1,7]phenanthroline derivatives 24a-h (scheme II.10). Because a single regioisomer was obtained, we consider these reactions to be highly regioselective in accordance with the electronic and steric effects of both dipole and dipolarophiles. In the case of salt 3h, EP has an interesting behavior, having both 1,3-dipolarophile and N-amidation roles. Thus, from the reaction mixture, we isolated cycloadducts 24g and 24g’ having the amide group mono- and disubstituted, respectively.
II.3.2.2. 3+2 Dipolar cycloaddition reactions of 4,7-phenantrolinium monoquaternary salts to unsymmetrical substituted reactive alkynes

The cycloaddition reactions of the in situ generated ylides from the 4,7-phenantrolinium monoquaternary salts to EP were carried out in similar conditions with the ones described for 1,7-phenanthroline ylides. As expected, from the reactions of ylides 11 with EP, only a single regioisomer was isolated, in accordance with the electronic effects in both reactants. The reaction of ylide 11b and EP gave a mixture consisting of 8,9-dihydropyrrolo[2,1-c][4,7]phenanthroline derivative 27b and fully aromatized cycloadduct 26b (Scheme II.11). In the case of salt 5f, EP played both dipolarophile and N-amidation roles, as we only isolated cycloadduct 28f from the reaction mixture, which has the amide group N-monoalcoholated with a molecule of EP.

Scheme II.11. Synthesis of fused pyrrolo derivatives 26, 27 and 28

Figure II.36. X-ray molecular structure of compound 27b with thermal ellipsoids at 50% probability level.
II.3.3. Cycloaddition reactions to symmetrically substituted alkenes

II.3.3.1. 3+2 Dipolar cycloaddition reactions of 1,7-phenanthrolinium monoquaternary N-ylides with symmetrically reactive alkenes

For this objective, salts 3 were deprotonated under triethylamine treatment, and the \textit{in situ} resonance stabilized N-ylides 7 were reacted with N-ethylmaleimide (NEtMI) or N-phenylmaleimide (NPhMI) to give tetrahydropyrrolo[1,2-i][1,7]phenanthroline derivatives 31. In the case of salt 3g we obtained 10-ethyl-9,11-dioxo-10,11-dihydro-9H-pyrido[3',4':3,4]pyrrolo[1,2-i][1,7]phenanthroline-12-carboxamide 32g as a byproduct (Scheme II.13) probably through an oxidative dehydrogenation that occurred under air work up.

The cycloaddition reactions with N-ethyl or N-phenylmaleimide occur with high stereoselectivity, only a single stereoisomer (31) being obtained.

\begin{align*}
\text{Scheme II.13. The reaction pathway of the \textit{in situ} generated ylides from 1,7-phenanthrolin-7-iium salts 3 and activated symmetrical alkenes}
\end{align*}

\begin{align*}
\text{Figure II.42. X-ray molecular structure of compound 31a. Thermal ellipsoids are drawn at 50\% probability level}
\end{align*}
II.3.4. Cycloadducts synthesis by reactions with unsymmetrically substituted reactive alkenes

II.3.4.1. 3+2 Dipolar cycloaddition reactions of 1,7-phenanthroline monoquaternary N-ylides to acrylonitrile

Having the experience of the reaction with NEtMI, we expected some instability for the compounds obtained through this reaction. Due to the different tendency to stabilize after oxidation, the structures of the final compounds (using similar conditions – in dichloromethane, at room temperature for 24h) were different depending on the starting ylide used. Thus, a single compound 39e (with tetrahydropyrrolo structure) was obtained in the case of the ylide generated from the salt 3e, a single compound 40g (with dihydropyrrolo structure) was obtained in the case of the ylide generated from salt 3g, while for the ylide generated from the salt 3b, a mixture of tetrahydro and dihydroderivatives (39b and 40b) was obtained (Scheme II.17).

Scheme II.17. The cycloaddition reaction pathway of the in situ generated ylides from 1,7-phenanthroline-7-ium salts 3 to acrylonitrile

Figure II.53. X-ray molecular structure of compound 39e. Thermal ellipsoids are drawn at 50% probability level
II.4. Study of the reactions of ethyl cyanoformate with cycloimmonium salts and their ylides

In our first attempt to synthesize a new imidazophenanthroline skeleton, we used 1,10-phenanthrolin-1ium halides in basic medium (trimethylamine in methylene chloride) for the generation of the corresponding ylides in order to react with ethyl cyanoformate in a 3+2 cycloaddition reaction. Instead the expected cycloadducts 46, we obtained 4-cyano substituted 1,10-phenanthrolines 47 (Scheme II.20). Interestingly, in case of the reaction of salt 46c, the final product 47c contained a methyl ester group instead the initial amide group, this replacement probably occurring during the column chromatography, when we used CHCl₃/MeOH system as eluent.

Scheme II.20. Synthesis of 4-cyano-substituted 1,10-phenanthrolines 47

Figure II.68. X-ray crystal structure of compound 47a with thermal ellipsoids at 50% probability level.

From the reactions of 4,7-phenanthrolin-4ium salts 5 under similar conditions (Scheme II.22), we isolated both γ-cyano-substituted 1,4-dihydro-4,7-phenanthrolines 49, and 3-oxo-3,4-dihydro-4,7-phenanthrolines 50 (Scheme II.22).
Scheme II.22. Synthesis of 1-cyano-substituted 4,7-phenanthrolines 49 and 50

We then decided to extend the study to reactions of other cycloimmonium salts under similar conditions. Thus, quinolinium salts 51 underwent similar γ-cyanation and α-oxidation yielding compounds 52a-c and other byproducts (53, 54 and 55) suggesting instability of the in situ ylides generated from the salts 51 under the reaction conditions (Scheme II.23).

Scheme II.23. Synthesis of 4-cyano substituted quinolines 52

Interestingly, isoquinolinium salts 56 having no available γ-position for cyanation yielded 3+2 cycloadducts 57a-c with imidazo[2,1-a]isoquinoline structure together with few decomposition coproducts (58, 59, 60) (Scheme II.24).

Scheme II.24. Synthesis of imidazoisoquinolines 57

Finally, we investigated phthalazinium bromides 61a-c, also without an unsubstituted position γ to nitrogen, in a similar reaction. As expected, we obtained the imidazo[2,1-a]phthalazines 62 as product of 3+2 cycloaddition. As secondary products in the same reaction we isolated compounds 63 with 8,8a,16,16a-tetrahydropyrazino-[2,1-a;4,5-a']-
diphthalazine structure (Scheme II.25) by dimerization via the [3+3] cycloaddition of the corresponding ylides.

![Diagram](image)

**Scheme II.25.** Synthesis of imidazophthalazines 62 and 8,8a,16,16a-tetrahydropyrazino-[2,1-a;4,5-a’]-diphthalazines 63

### II.5. Design and evaluation of biological activity of selected new synthesized compounds

#### II.5.1. Design and evaluation of anticancer activity of the selected new derivatives

In our synthesis plan, the target compounds (new indolizines with phenanthroline skeleton) were designed to have potential anticancer activity (Scheme II.29). In order to investigate the anticancer activity of the newly synthesized compounds, we submitted selected compounds to the National Cancer Institute to be tested on their 60 human tumour cell line. Fourteen compounds (Scheme II.30) were accepted and tested for anticancer activity under the Developmental Therapeutics Program (DTP), at a single dose (10⁻⁵M) cell assay. This assay was performed in a 60 human tumor cell line panel, representing leukemia, melanoma and cancers of the lung, colon, brain, breast, ovary, kidney and prostate, in accordance with the protocol of the NCI. The results are expressed in terms of ‘percentage growth inhibition’ (PGI), and represent growth relative to the no-drug control, and relative to the zero time number of cells, Table II.1. This allows detection of both growth inhibition (values between 0 and 100) and lethality (values less than 0).

The results from Table II.1 indicate that:

- indolizines with 1,7-phenanthroline skeleton, 9b and 13a, exhibit a significant antitumor growth inhibitory activity (around 50%) against Breast Cancer (MCF7 and T-47D).
- pyrrolo [2,1-c][4,7]phenanthroline derivatives 26d and 26g exhibit a significant antitumor growth inhibitory activity (around 50%) against renal cancer (UO-31) and breast cancer (MCF7), respectively.
derivatives 47a and 48e exhibit a significant antitumor growth inhibitory activity (around 25, 45%) against ovarian cancer (SK-OV-3). Compounds 48e, 57c and 62b exhibit a significant antitumor growth inhibitory activity (around 40%) against prostate cancer (PC-3) and Non-Small Cell Lung Cancer (NCI-H522) (around 35%). We may also notice a weaker antitumor growth inhibitory activity (around 35%) of 62b, 50b, 57c and 48e against Leukemia CCRF-CEM, 48e and 57c against renal cancer A498 and 48e and 63b against Breast Cancer (T-47D and MDA-MB-231/ATCC)

Scheme II.29. Design in the class of (aza) indolizines derivatives with phenanthroline
Scheme II.30. Structure of evaluated compounds
Table II.1. Percentage growth inhibition (PGI, lM) data of compounds 9b, 13b, 13d, 24a, 26d, 26g, 47a, 48e, 48g, 50b, 57c, 62a, 62b and 62c against an NCI 60 human tumour cell lines (selection)

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<td>4.82</td>
<td>3.14</td>
<td>0</td>
<td>12.18</td>
<td>10.99</td>
<td>17.56</td>
<td>12.26</td>
<td>21.32</td>
<td>4.6</td>
<td>14.27</td>
<td>8.17</td>
</tr>
<tr>
<td><strong>Breast cancer</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>MCF7</td>
<td>13.85</td>
<td>46.97</td>
<td>7.41</td>
<td>46.97</td>
<td>6.9</td>
<td>4.36</td>
<td>5.38</td>
<td>14.19</td>
<td>22.02</td>
<td>6.33</td>
<td>3.25</td>
<td>19.24</td>
<td>17.92</td>
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</tr>
<tr>
<td>MDA-MB-231/ATCC</td>
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<td>0</td>
<td>0.94</td>
<td>0</td>
<td>5.27</td>
<td>4.06</td>
<td>9.29</td>
<td>4.46</td>
<td>28.37</td>
<td>8.98</td>
<td>16.54</td>
<td>18.29</td>
<td>29.57</td>
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</tr>
<tr>
<td>T-47D</td>
<td><strong>45.62</strong></td>
<td>11.25</td>
<td>18.91</td>
<td>11.25</td>
<td>13.99</td>
<td>29.24</td>
<td>10.34</td>
<td>9.1</td>
<td>18.87</td>
<td>2.06</td>
<td>19.81</td>
<td>14.08</td>
<td><strong>40.69</strong></td>
<td>0.34</td>
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</tbody>
</table>
II.5.2. Design and evaluation of antimycobacterial activity of the selected new derivatives

The evaluation of the antimycobacterial activities of the compounds were performed at the Center of Tuberculosis Antimicrobial Acquisition and Coordinating Facility (TAACF) of the Southern Research Institute.

First we submitted for testing compounds 3 and 5 (Scheme II.32). The obtained results are listed in Table II.2 and illustrate that compounds 3e and 3f, from the tested 1,7-phenanthrolinium monoquaternary salts 3a-h, and compound 5d from the tested 4,7-phenanthrolinium monoquaternary salts 5a-d,f had the most pronounced activity against *M. tuberculosis* H37Rv under aerobic conditions.

**Scheme II.32.** Structure of tested compound 3 and 5

By comparison, we basically observe only a minor influence of the heterocycle on antimycobacterial activity.

**Table II.2.** Antimycobacterial activity of phenanthrolinium salts 3a-h and 5a-d,f against *M. tuberculosis* H37Rv.

<table>
<thead>
<tr>
<th>Compound</th>
<th>IC_{50} (μM)</th>
<th>IC_{90} (μM)</th>
<th>MIC (μM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>3a</td>
<td>100</td>
<td>&gt;200</td>
<td>&gt;200</td>
</tr>
<tr>
<td>3b</td>
<td>&gt;200</td>
<td>&gt;200</td>
<td>&gt;200</td>
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<tr>
<td>3c</td>
<td>180</td>
<td>&gt;200</td>
<td>&gt;200</td>
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<tr>
<td>3d</td>
<td>120</td>
<td>&gt;200</td>
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<tr>
<td>3e</td>
<td>88</td>
<td>&gt;200</td>
<td>&gt;200</td>
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<tr>
<td>3f</td>
<td>88</td>
<td>&gt;200</td>
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<td>3g</td>
<td>&gt;200</td>
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<tr>
<td>3h</td>
<td>&gt;200</td>
<td>&gt;200</td>
<td>&gt;200</td>
</tr>
<tr>
<td>5a</td>
<td>110</td>
<td>&gt;200</td>
<td>&gt;200</td>
</tr>
<tr>
<td>5b</td>
<td>&gt;200</td>
<td>&gt;200</td>
<td>&gt;200</td>
</tr>
<tr>
<td>5c</td>
<td>&gt;200</td>
<td>&gt;200</td>
<td>&gt;200</td>
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<tr>
<td>5d</td>
<td>83</td>
<td>&gt;200</td>
<td>&gt;200</td>
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<tr>
<td>5f</td>
<td>&gt;200</td>
<td>&gt;200</td>
<td>&gt;200</td>
</tr>
<tr>
<td>Rifampicin</td>
<td>0.0036</td>
<td>0.0061</td>
<td>0.0055</td>
</tr>
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</table>
II.6. Electric properties of selected compounds

This study was made in collaboration with the group of prof. Liviu Leontie from the Physics Department of Alexandru Ioan Cuza University of Iasi.

The structural investigation by X-Ray diffraction showed that the synthesized organic compounds (Scheme II.35) in thin films have a prevailing polycrystalline structure (Fig. II.84). The structures of the samples were found to depend on both compound nature and film thickness.

The d.c. electric conductivity of the synthesized compounds was investigated using thin-film samples deposited through the spin-coating technique, at room temperature, onto glass substrates.

We have experimentally established that, after two series of successive heating/cooling runs, the film structure becomes stable and the temperature dependences of the electric conductivity become reversible. This behavior indicates stabilization of a thin-film solid-state structure within the investigated temperature range, ΔT. In the higher temperature range, an intrinsic conduction mechanism is present in the organic films under study (Fig. II.87).
The recently synthesized organic compounds (SMC series) in thin films behave as typical n-type polycrystalline semiconductors. The electron transfer in the investigated compounds is strongly influenced by their specific molecular structures, enabling formation of extended conjugation systems, as well as by their packing capacity.

![Figure II.87. Temperature dependence of electric conductivity for the heat-treated samples (SMC series)](image)

II.7. Fluorescence properties of selected compounds

This study was realized in collaboration with the group of Dr. Anton Airinei from the Institute of Macromolecular Chemistry “Petru Poni” Iasi.

Taking in consideration the fluorescent properties of indolizines derivatives we decided to study fluorescence and the relationship between optical properties and structure (the effect of the substituents and conjugation) of several pyrrolo[2,1-c][4,7]phenanthroline and pyrrolo[1,2-i][1,7]phenanthrolines (Scheme II.32), compounds with an extended π electrons conjugation.

![Scheme II.32. The structure of the investigated compounds](image)

1. \( R_1 = \text{CN; } R_2 = \text{H; } R_3 = \text{COOEt; } \\
2. \( R_1 = \text{COOMe; } R_2 = \text{H; } R_3 = \text{COOEt; } \\
3. \( R_1 = \text{COClH}_2\text{OME}(p); R_2 = \text{H; } R_3 = \text{COOEt; } \\
4. \( R_1 = \text{COClH}_2\text{Cl}(p); R_2 = \text{H; } R_3 = \text{COOEt; } \\
5. \( R_1 = \text{CN; } R_2 = \text{COOMe; } R_3 = \text{COOMe; } \\
6. \( R_1 = \text{COOMe; } R_2 = \text{COOMe; } R_3 = \text{COOMe; } \\
7. \( R_1 = \text{COClH}_2\text{OME}(p); R_2 = \text{COOMe; } R_3 = \text{COOMe; } \\
8. \( R_1 = \text{COOMe; } R_2 = \text{H; } R_3 = \text{COOEt; } \\
9. \( R_1 = \text{COClH}_2\text{OMe}(p); R_2 = \text{H; } R_3 = \text{COOEt; } \\
10. \( R_1 = \text{COClH}_2\text{Cl}(p); R_2 = \text{H; } R_3 = \text{COOEt; } \\
11. \( R_1 = \text{COClH}_2\text{Br}(p); R_2 = \text{H; } R_3 = \text{COOEt; } \\
12. \( R_1 = \text{COClH}_2\text{Br}(p); R_2 = \text{COOMe; } R_3 = \text{COOMe; } \\

The electronic absorption spectra of phenanthroline derivatives exhibit three main absorption bands located in the following domains: 350-410 nm (α band), 295-325 nm (p band) and 250-280 nm (β band) (Figure II.94, 96). All electronic absorption spectra exhibit the fine structure specific to the phenanthrene spectrum. The substituents in the pyrrole ring exert an important influence on the position of the absorption and emission bands of phenanthroline derivatives. It can be seen that the introduction of CN, COOMe and COOEt groups in the positions 8, 10 of the pyrrole moiety results in a blue shift of the absorption and emission bands of phenanthroline derivatives (1, 2, 8).

Figure II.94. Electronic absorption spectra in dichloromethane of disubstituted 4,7-phenanthroline derivatives:
1- compound 1; 2- compound 2; 3- compound 3; 4- compound 4

Figure II.96. UV-Vis absorption spectra in dichloromethane solution of 1,7-phenanthroline derivatives: 1-
compound 13; 2- compound 8; 3- compound 9; 4- compound 10; 5- compound 11; 6- compound 12
General conclusions

According with the objectives of this PhD thesis, the following main results were achieved:

1. The synthesis and structural characterization of new classes of 1,7-phenanthrolin-7-ium and 4,7-phenanthrolin-4-ium monoquaternary halides. Compounds were prepared by using a straight forward and efficient method of synthesis.

2. New fused pyrrolo-phenanthrolines, dihydropyrrolo-phenanthrolines and tetrahydropyrrolo-phenanthrolines were synthesized. The strategy adopted for each synthesis involves a typical Huisgen [3+2] dipolar cycloaddition of [1,7]phenanthrolin-7-ium ylides (generated \textit{in situ} from the corresponding salts) to activated symmetrical and unsymmetrical substituted alkynes or alkenes.

3. The cycloaddition reactions to symmetrically substituted dipolarophiles (N-ethyl- or N-phenylmaleimide) are highly stereoselective (a single isomer being obtained), whereas the cycloadditions to nonsymmetrically substituted dipolarophiles (acrylonitrile and ethyl propiolate) are highly regioselective (a single regioisomer being formed), under charge control.

4. The formation of aromatized and partially aromatized (dihydro-) fused polycyclic indolizines (in some cases), could be explained by an oxidative dehydrogenation of the initially obtained tetrahydro- fused intermediary, a process that leads to thermodynamically more stable compounds.

5. A particular behavior was identified for cycloaddition reactions with phenanthrolinium salts in which R= NH$_2$. The amide group was proven to be reactive enough to suffer subsequent enamidation steps with the alkynes used as dipolarophiles in the 3+2 cycloaddition reactions.

6. Reactions of cycloimonium salts with ethyl cyanoformate in basic conditions undergo through two different pathways depending of the availability of the \(\gamma\)-position of the heterocycle for a nucleophilic cyanation. Thus, in the case of 1,10-phenanthrolin-1-ium salts, 1,7-phenanthrolin-7-ium salts, 4,7-phenanthrolin-4-ium salts and quinolinium salts, ethyl cyanoformate acts as a source of cyanide ion, leading selectively to the corresponding \(\gamma\)-cyano substituted compounds. In case of isoquinolinium and phthalazinium ylides having bridged \(\gamma\)-position, ethyl cyanoformate acts as a dipolarophile in a 3+2 dipolar cycloaddition, leading to fused imidazo[2,1-\(a\)]isoquinoline and imidazo[2,1-\(a\)]phthalazine derivatives, respectively. From the reaction mixtures of phthalazinium ylides with ethyl cyanoformate we isolated 8,8a,16,16a-tetrahydropyrazino[2,1-\(a\):5,4-\(a\)]dipthalazine dimers as byproducts.
7. The structures of the newly synthesized compounds were confirmed by elemental and spectral analysis: IR, $^1$H-NMR, $^{13}$C-NMR and XRD. The X-ray diffraction (single-crystal) studies unambiguously demonstrated the structures of compounds (including those of stereo- and regioisomers) and brought valuable information concerning lattice structure, interesting intermolecular C-H···N and C-H···O hydrogen bonds, as well as π-π stacking interactions.

8. Fourteen new compounds were evaluated by NCI for anticancer activity. The anticancer screening against a NCI 60 human tumor cell line panel revealed that the indolizines with a phenanthroline skeleton, 9b and 24a, exhibit a selective and significant antitumor activity (around 50% growth inhibition) against Breast Cancer (MCF7 and T-47D). A slightly moderate antitumor activity (around 25% growth inhibition) was observed against Leukemia (CCRF-CEM, MOLT-4, RPMI-8226, SR), Non-Small Cell Lung Cancer (HOP-62, NCI-H522), Renal Cancer (ACHN, UO-31) and Breast Cancer (MDA-MB-468). The X-ray analysis of the indolizines with a phenanthroline skeleton demonstrate a flat coplanar structure which, corroborated with their anticancer activity, allow us to suggest that an interaction with DNA (via an intercalation mechanism) would be reasonable. Pyrrolo[2,1-c][4,7]phenanthroline derivatives 26d and 26g exhibit a significant antitumor activity (around 50% growth inhibition) against renal cancer (UO-31) and breast cancer (MCF7), respectively. We could also observe a weak antitumor activity (around 25% growth inhibition) of 26d against CNS Cancer (SF-SNB-19), 13b against Melanoma (M14) and 26d against Breast Cancer (T-47D). Derivatives 47a and 48e exhibit a significant antitumor growth inhibitory activity (around 25 and 45% growth inhibition, respectively) against ovarian cancer (SK-OV-3). Compounds 48e, 57c and 62b exhibit a significant antitumor activity (around 40% growth inhibition) against prostate cancer (PC-3) and Non-Small Cell Lung Cancer (NCI-H522) (around 35% growth inhibition). We may also notice a weaker antitumor activity (around 35% growth inhibition) of 62b, 50b, 57c and 48e against Leukemia (CCRF-CEM), 48e and 57c against renal cancer (A498) and 48e and 62b against Breast Cancer (T-47D and MDA-MB-231/ATCC).

9. Forty-three new compounds were evaluated for in vitro antimycobacterial activity against *M. tuberculosis* H37Rv (grown under aerobic conditions), as a part of the TAACF TB screening program under the direction of the US National Institute of Health, the NIAID division. The tested compounds with phenanthroline skeleton had a good solubility in the microbiological medium (between 10 and 200 μM), which is quite promising from a pharmacological point of view. Two out of the eight tested 1,7-phenanthroline
monoquaternary salts had activity against *M. tuberculosis* H37Rv under aerobic conditions (3e, 3f), while one of the five tested 4,7-phenanthrolinium monoquaternary salts (5f) also had activity against *M. tuberculosis* H37Rv.

Unfortunately, only compound 31a of all tested pyrrolophenanthroline compounds had activity against *M. tuberculosis* H37Rv under aerobic conditions.

10. Five monoquaternary salts and six pyrrolo cycloadducts were tested for their electrical properties. The recently synthesized organic compounds, (SMC and LL compounds) in thin films, behave as typical n-type polycrystalline semiconductors. The electron transfer in the investigated compounds is strongly influenced by their specific molecular structures, enabling the formation of extended conjugation systems, as well as their packing capacity. In the higher temperature range ($T>T_c$), the d. c. electric conductivity of examined compounds can be described in terms of the band gap representation model. In the lower temperature domain (298 K<$T<T_c$) the Mott’s variable-range hopping model can be conveniently used.

11. Twelve phenanthroline derivatives were tested for their fluorescent properties. The electronic absorption spectra of phenanthroline derivatives exhibit three main absorption bands located in the following domains: 350-410 nm ($\alpha$ band), 295-325 nm (p band) and 250-280 nm ($\beta$ band). The substituents in the pyrrole ring exert an important influence on the position of the absorption and emission bands of phenanthroline derivatives. The fluorescence spectra of 1,7-phenanthroline derivatives displayed an emission band in the 440-450 nm region, while the emission range of 4,7-phenanthrolines was found to be from 420 to 480 nm depending on the nature of the substituent at the pyrrole moiety. The strongest emission was observed for compounds with a CN group at position 8 in the 4,7-phenanthroline series, while in the 1,7-phenanthroline series compound 8 had the highest fluorescence. The presence of chlorine or bromine in the phenanthroline derivatives practically quenches the fluorescence emission. The fluorescence quantum yields of pyrrolo-phenanthroline derivatives are dramatically dependent on the position of the substituent at the pyrrole ring.

12. 100 New compounds belonging to different classes of heterocycles were synthesized and fully characterized.

13. The results presented in this thesis were disseminated in five ISI articles and 13 international and national symposiums or conferences.
References (selection)

The scientific activity and dissemination of results

Publications


Oral presentations

• C. Al Matarneh, R. Danac, L. Leontie, C. Doroftei, I. Mangalagiu, Synthesis and electron transport properties of new pyrrolo [1,2-i][1,7]-phenanthroline derivatives in thin films, 8ème Colloque Franco-Roumain de Chimie Appliquée (COFrRoCA), Montpellier, Franta, September 2014.

• Maria Cristina Al-Matarneh, Ionel Mangalagiu, Ramona Danac, Synthesis and structure of new fused polycyclic indolizines, the XXXIII\textsuperscript{th} National Chemistry Conference Calimanesti, Valcea, October 2014.

• M.C. Al-Matarneh, R. Danac, I. Mangalagiu, Synthesis and antimycobacterial properties of new derivatives of 1,7-phenanthroline, 3\textsuperscript{rd} French-Romanian Colloquium on Medicinal Chemistry, Iasi, October 2014.

Posters


• L. Leontie, C. Doroftei, C. Al Matarneh, R. Danac, Electron transport properties of new pyrrolo [1,2-i][1,7]-phenanthroline derivatives in thin films, MSCMP 2014 - 7th International Conference on Materials Science and Condensed Matter Physics, Chisinau, September 2014.

• M. C. Al-Matarneh, A. Chirciu, M. Apostu, R. Danac, Synthesis and structure of new 1,10-phenantrolene derivatives, „University Days” Iasi, October-November 2014.

• **M. C. Al-Matarneh**, R. Danac, M. O. Apostu, I. Mangalagiu, Synthesis and structure of new pyrrolo[1,2-a][1,10]phenanthroline derivatives and α-monosubstituted 1,10-phenanthrolines, 19th Romanian International Conference on Chemistry and Chemical Engineering (RICCCE), Sibiu, Romania, September 2015.


• **Cristina Maria Al Matarneh**, Ramona Danac, Mircea O. Apostu, Ionel I. Mangalagiu, Reactions of ethyl cyanoformate with cycloimmonium salts and their ylides: a direct pathway to fused or substituted azaheterocycles, The XXXIV\textsuperscript{th} National Chemistry conference, Calimanesti, Valcea, October 2016.