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Praca doktorska:

Oligofunkcjonalizacja klasterów boru jako bloków budulcowych nowych materiałów i związków bioaktywnych.

Doctoral thesis:

Oligofunctionalization of boron clusters as building blocks for new materials and bioactive molecules.

- Promotor/Supervisor
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DOCTORAL DISSERTATION ABSTRACT

The objective of the research described in this doctoral thesis was to create chemical basis for the preparation of new types of nanoparticles designed as therapeutic nucleic acid carriers, being one of the tasks of the Symfonia 3 grant project. These aims were also the basis for the Individual Research Plan at the BioMedChem Doctoral School. The planned aim was achieved as a result of several years of research and allowed the development of useful in-practice methods of oligofunctionalization of both *ortho*-carborane as well as its complex with a metal, metallocarborane containing cobalt cation, and their use in the construction of new types of nanoparticles.

The structure of boron clusters is stabilized by a three-dimensional network of delocalized covalent bonds in which boron (and carbon) atoms are coordinated with at least three, and sometimes even five or six, boron atoms, creating extremely stable, "non-classical" molecular systems. The unique properties of carboranes, including aromaticity in three dimensions cause each substitution at a boron or carbon atom change of the distribution of electron densities throughout the whole molecular cluster, and therefore change the reactivity of all other atoms in the molecule. The reactivity change of boron and carbon atoms is the result of the overlap of many factors. In the literature introductory part of the thesis, I attempted to catalog these factors and use them to explain the preferential formation of derivatives of disubstituted carboranes and metallacarboranes compared to their monosubstituted derivatives. This is a problem that often complicates the oligofunctionalization of carboranes and metallacarboranes in the desired form which I also encountered in my research.

In the experimental part of my thesis, I described the results of research on the oligofunctionalization of 1,2-dicarba-*closo*-dodecaborane on boron atoms B_9 and B_{12} and carbon atoms C_1 and C_2, as well as on the oligonufunctionalization of a metallacarborane containing cobalt, bis(1,2-dicarba-*closo*-dodecaborane)-commo-3,3’-cobaltate (III) (COSAN) on boron atoms B_8 and B_8' and carbon atoms C_1, C_2, C_1' or C_2'. Due to the practical goal of my research, which was to obtain functionalized derivatives of boron clusters allowing the synthesis of building blocks of nanoparticles, composites of boron clusters and antisense DNA-oligonucleotides, they had to comply with the requirement of attaching two substituents with a trityl protection group and one substituent with a free hydroxyl group to the cluster. The practical goal depended on parallel basic research broadening knowledge about the chemistry of boron clusters, methods of their oligofunctionalization by attaching, if possible, in a

controlled manner, one, two, three, or four substituents to boron and carbon atoms, as well as expanding knowledge about the stereochemistry of these complex systems.

In the section on oligofunctionalization of 1,2-dicarba-*closo*-dodecaborane, I modified and optimized the previously described method of synthesis of 9,12-bis(3-O-trityloxyprop-1-yl)-1,2-dicarba-*closo*-dodecaborane, the key compound for further synthetic steps and I designed the method of attaching hydroxyalkyl substituent. Thus synthesized trisubstituted 1,2-dicarba-*closo*-dodecaborane was successfully used to obtain functional nanoparticles of second generation, composites of boron clusters, and DNA-oligomers. Using ^{11}B-NMR and ^{1}H-NMR spectroscopy I analyzed the influence of incorporation of subsequent substituents into boron cluster structure on its properties change related to the changing of electron density distribution in cluster and stereochemistry of obtained derivates.

In the section on functionalization of metallacarborane bis(1,2-dicarba-*closo*-dodecaborane)-commo-3,3'-cobaltate (III) (COSAN) I designed: 1) synthesis method of COSAN derivates, disubstituted on boron atoms B_8 and $B_{8'}$ with alkylhydroxy groups with trityl or *tert*-butyldimethylsilyl protections, 2) synthesis method of COSAN derivates functionalized on B_8 and $B_{8'}$ boron atoms and simultaneously mono- or difunctionalized on carbon atoms C_1, $C_{1'}$, C_2, $C_{2'}$, 3) synthesis method of cyclic thiophosphate esters of 8,8'-dihydroxy COSAN with hampered rotation of carboranyl ligands, alkylated on sulfur atom with linear or branched substitutents, 4) method of oligofunctionalization with alkylhydroxy groups on carbon atoms C_1, $C_{1'}$, C_2 or $C_{2'}$ of COSAN derviates with hampered rotation of carboranyl ligands mentioned in 3, 5) method of synthesis of 8-(5-hydroxy-3-oxa-pentoxy)-[1,1'-di(2-O-trityloxyethyl)-bis(1,2-dicarbollide)-3,3'-cobaltate (III), a new type of oligofunctionalized derivate of COSAN. This derivate was used to obtain nanoparticles of third generation. All of the above-mentioned derivates and chemical research constitute original contributions to boron clusters chemistry.

Moreover, similar to 1,2-dicarba-*closo*-dodecaborane, using ^{11}B-NMR and ^{1}H-NMR spectroscopy I analyzed the influence of incorporation of substituents into metallacarborane structure on its properties change connected with the changing distribution of electron density and stereochemistry of obtained derivates. The complexity of these processes for metallacarboranes is even bigger than for 1,2-dicarba-*closo*-dodecaborane. The results of my research may contribute to its better understanding.

Next, I described, in short, the use of obtained oligofunctionalized boron clusters in 1) synthesis of building blocks of nanoparticles, composites of boron clusters and anti-sense DNA-oligonucleotides by incorporation of DNA-oligomer to functionalized boron cluster, 2) assemble of nanoparticles by annealing of obtained building blocks, 3) research on

physicochemical properties and biological activity of nanoparticles. The first part was carried out in the cooperation with Laboratory of Nucleic Acid Therapeutics (Centre of Molecular and Macromolecular Studies), and the second and the third parts were performed in our Laboratory in the Institute of Medical Biology of Polish Academy of Sciences, mainly by dr Katarzyna Bednarska-Sczepaniak and dr Gabriela Gajek. A more detailed description of this research can be found in Publication 2 (position 9 of the Bibliography) of which I am a co-author. This work along with Publication 1 (position 94 of the Bibliography) constitute the basis of my thesis. The work positioned as 101 in the Bibliography part sums up the latest results of research described in part 2.2.6 and is in preparation for publication.

In the last part of my thesis, I described the results of preliminary research on the synthesis of genistein analogs containing 1,2-dicarba-*closo*-dodecaborane in the structure. This project is not directly connected with oligofunctionalizaion of boron clusters but refers to the title of the thesis in the context of the synthesis of bioactive compounds. Genistein is a natural compound belonging to flavonoid group with a wide spectrum of activities. I was mostly interested in its hormone-like activity and affinity to β-estrogen receptor (ER). I obtained two analogs of genistein containing boron cluster that show different in vitro cytotoxicity in cell lines Huh70 (cytotoxicity tests were carried out by dr Katarzyna Bednarska-Sczepaniak). Currently, the obtained derivates are being examined as potential selective α- and β- estrogen modulators within collaboration with Dr Geert A. Daudey of the Center for Research in Molecular Medicine and Chronic Disease University of Santiago de Compostela, Spain.