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Fullerene C_{60}–Conjugated Doxorubicin for Advanced Tumor Therapy

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After heart disease and stroke, cancer is the third leading cause of death worldwide, accounting for an estimated 9.6 million deaths in 2018 [1] and breast cancer is one of the most deadly diagnosed cancer amongst women. The anthracycline antibiotic – doxorubicin (DOX) is a commonly used antineoplastic agent in chemotherapy of a wide range of cancers e.g. leukemia, but also in the treatment of solid tumors such as breast cancer [2]. However, doxorubicin may induce cardiotoxicity and congestive heart failure, which justifies an increasing demand for developing of new strategies minimizing its side effects. In this context, nanomaterials seem to be attractive agents that might allow to keep a balance between therapeutic efficacy and adverse after effects.

A fullerene–doxorubicin conjugate (Ful–DOX) was obtained using the malonic diester scaffold *via* the modified procedure described by Lu et al. [3]. An alcoholysis reaction of malonyl chloride was followed by functionalization with the fullerene C60 (Bingel–Hirsch addition) and the selective hydrolysis of tertiary esters. Next, activation reaction in the presence of DCC (*N*,*N*'-dicyclohexylcarbodiimide) and NHS (*N*-hydroxysuccinimide) led to the bis-*N*-succinimide activated diester, which was coupled with doxorubicin to give the final conjugate. The identity of the obtained product was confirmed with ^{1}HNMR and mass spectrometry. The Ful–DOX conjugate was investigated by UV-Vis spectroscopy, spectrofluorimetric measurements and dynamic light scattering (DLS). In order to determine biological activity of Ful–DOX, the MTT assay and confocal microscopy were employed.

Intra- and inter-molecular interactions in conjugate solutions lead to the substantial quenching of DOX fluorescence. It can be related, at least partially, to the aggregation of conjugate, as indicated by its DLS hydrodynamic diameter equal to 825.0 nm. The results of biological assays, carried out using the MCF-7, T47D and MDA-MB 231 cell lines, confirma substantially higher cytotoxicity of free DOX compared to the conjugate. This result seems to be in line with confocal fluorescence images, which suggest that the distribution of free DOX and conjugate in the studied cells differs dramatically. The former accumulates in the nucleus, while the latter concentrates on nuclear membrane, which probably prevents the cytotoxic activity of DOX.

In summary, DOX covalently conjugated with fullerene could be a potential drug delivery system to alleviate anthracycline toxicity. Our future efforts will goes in to development of a conjugate which will release the drug Specifically on target, i.e. cancer cells.

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Biography:

Kamila Butowska, M.Sc. in 2017 she defended Master's thesis at the Faculty of Chemistry, University of Gdańsk and specialty: Chemical Analytics and Diagnostics, under the supervision of Professor Janusz Rak. In the same year she commenced her PhD study at the Intercollegiate Faculty of Biotechnology of the University of Gdańsk and Medical University of Gdańsk under the supervision of Professor Jacek Piosik. During her Masters she won "AMBER" contest and as consequence, played a role of principal investigator for the project entitled "Radio sensitizers with high affinity to electron. Physicochemical and cellular research" founded by European Social Found of EU under Inno-Agro Chem Ośinitiative. She was also an intern in the group of Professor Andreas Kornath at the Department of Chemistry and Pharmacy at the Ludwig Maximilians University in Munich. Her main scientific interests are chemical synthesis and studies on biological activity of conjugates of metalloproteinase-2 and metalloproteinase-9-sensitive peptides with doxorubicin to be used for targeted anticancer therapy.