Chapter 3
Antioxidant Properties of Water-Soluble Fullerene Derivatives

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Abstract Due to their inherent electronic properties, fullerenes are considered as radical sponges being capable of effectively quenching reactive oxygen species (ROS). The most promising candidates for potential pharmaceutical applications are therefore water-soluble fullerene derivatives, since they provide reasonable biological availability. In light of these considerations, we give an overview over the most recent concepts for designing and synthesizing real water-soluble fullerene compounds. Several studies concerning the quenching activities against ROS-like superoxide radical anion of some of these novel compounds are reviewed. We finally present first promising investigations about cytoprotective and neuroprotective activities of several carboxyfullerenes in zebrafish embryos as a mammalian model system. By comparing the activities for different addition patterns and other structural changes some first conclusions concerning a structure–function relationship can be drawn.

Keywords Fullerene, water-solubility, antioxidant, cytoprotection

3.1 Introduction

Shortly after the discovery of the fullerenes in 1985 (Kroto et al., 1985) and especially after their accessibility in macroscopic quantities (Kraetschmer et al., 1990) these new carbon allotropes raised great interest in the chemical world due to their unique structural and electronic properties. As a direct consequence of the curved conjugated \( \pi \)-system fullerenes were predicted to be fairly electronegative with the
ability to undergo multiple reversible reduction steps. Indeed this was demonstrated by the fully reversible reduction up to the hexaanion for $C_{60}$, the most prominent and abundant fullerene (Echegoyen and Echegoyen, 1998). As another important chemical property their pronounced affinity towards radicals was pointed out, shortly thereafter (Hirsch and Brettreich, 2005). Upon addition of a large variety of radicals quite stable diamagnetic or paramagnetic adducts were observed and intensively studied.

Summarizing these facts, fullerenes can be considered as very effective ‘radical sponges’ (Krusic et al., 1991; McEwen et al., 1992) being capable of taking part in reversible redox processes. These properties make fullerenes very interesting with respect to potential applications in the field of biomedical or pharmaceutical chemistry. It is widely believed that reactive oxygen species (ROS) like hydroxyl radicals, peroxides and predominantly the superoxide radicals cause significant cell death and are important factors in aging as well as in chronic neurodegenerative diseases like ALS, Alzheimer’s and Parkinson’s. Due to their inherent structural properties and their ability to act as radical sponges, fullerenes and their derivatives were proposed to have potent antioxidant and neuroprotective activities in biomedical applications. In general, two different mechanisms for the quenching of ROS by fullerenes can be considered. The first involves a stoichiometric addition of ROS to the fullerene surface, probably followed by subsequent elimination or addition steps in order to regenerate the parent fullerene and resulting in a catalytic ROS cleavage. The second proposed mechanism is based on several outer sphere electron transfer processes involving reduction and reoxidation steps between the fullerene and radical species.

For successful clinical applications, it is necessary to demonstrate the specific mode of action, the pharmacokinetics and the distribution in various tissues after administration. One severe drawback for fullerenes in this context is the almost complete insolubility of these all-carbon allotropes in water. To overcome these impediments, one of the first goals in fullerene chemistry was to design and synthesize water-soluble derivatives, which retain the unique inherent fullerene properties while achieving reasonable biological availability. Subsequent antioxidant and protective studies using these novel water-soluble fullerenes produced promising results. Studies carried out with antibodies against fullerenes suggested that water-soluble fullerenes can readily pass cell membranes and are preferentially located at or near mitochondria (Foley et al., 2002) – the cellular organelle responsible for aerobic respiration and the site where most of the biological processes concerning ROS take place. These initial studies demonstrated substantial antioxidant activity both in vitro and in vivo, encouraging a flurry of new work in this area.

In this contribution we want to provide a short overview addressing the current state-of-the-art of water-soluble fullerene derivatives and their potential applications as antioxidant or neuroprotective drug candidates. After summarizing the most prominent concepts of designing water-soluble fullerenes in the first chapter we will present some more recent achievements with respect to biological activities of antioxidant fullerenes, with emphasis on our own results.