Diruthenium(II,III)-ketoprofen anticancer metallodrug investigated for interaction with transferrin and for effects on the antiproliferative activity, apoptotic and mitotic processes in the A172 human glioma cell line

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Ruthenium compounds have played key role in the development of new antitumor metallopharmaceuticals. Our research is focused on studies of paddlewheel structured complexes bearing the Ru_2(II,III) core coordinated to drugs of clinical use such as the non-steroidal anti-inflammatory drugs (NSAIDs), and also on the investigation of their properties as anticancer agents. The lead-complex of Ibuprofen was found to exert growth inhibitory effects in rat and human glioma cells *in vitro* and also in the rat C6 orthotopic glioma *in vivo* [1,2]. The unique effects of this complex in the models for Glioblastoma (grade IV/WHO), a malignant and aggressive human brain cancer associated to poor patient survival, have opened new perspectives to expand our studies in the field. In previous work, we reported the *in vitro* activity of the Ketoprofen analogue, $[Ru_2(Ket)_4Cl]$ or RuKetCl, Ket = Ketoprofenate anion, in the HT-29 and Caco-2 human colon carcinoma cells [3], and the interaction of this complex with the human serum albumin [4]. In the present study, we have exploited the interaction of RuKetCl with transferrin, and we have also investigated the effects of this metallodrug on the antiproliferative activity and apoptotic and mitotic processes in the A172 human glioma cell line. Data from electronic spectroscopy, circular dichroism, fluorescence and ultrafiltration, accompanied by adduct analysis from ICP-OES and MALDI-MS, reveal that the complex interacts with the protein in its both forms, Apo and Holo, and, moreover, that these interactions seem to not depend on the presence of Fe(III) in the specific binding sites of the transferrin. The secondary structure of the protein was not modified by the presence of the complex. The capacity for retention of Ru was ~ 70 %, and the results suggest weak protein-metallodrug interactions mainly dominated by hydrophobic forces. The RuKetCl complex exhibits dose and time dependent antiproliferative activity in the A172 human glioma cell line with the most significant effect ($p < 0.001$) at 200 µmol L^{-1} metallodrug concentration after 72 h treatment exposition. The cellular apoptotic process was significantly increased while the cellular mitotic process was significantly decreased after the cells were exposed to the metallodrug. The metal cellular uptake was followed by ICP-OES. These findings reveal that the RuKetCl interacts with the serum transferrin and shows promising biological effects in the A172 human glioma cell line, thus broadening the possibility of exploiting diverse Ru_2(II,III)-NSAID drugs to target glioblastoma.

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