Chemistry and Pharmacological Properties of Two Gold(I) Carbene Complexes as Potential Anticancer Agents

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Following the introduction of auranofin in the clinics for the treatment of rheumatoid arthritis and the discovery of its remarkable antiproliferative properties *in vitro*, gold compounds were increasingly considered as a possible source of new metal-based anticancer agents. This interest was also fuelled by the observation that gold compounds usually manifest a very different pharmacological profile compared to established anticancer platinums implying the occurrence of original and innovative modes of action. In the last two decades, several promising families of Au-based drug candidates, with the gold centre in the oxidation states +3 or +1, featuring different structural motifs, were prepared and characterised and their biological and pharmacological profiles were initially assessed (Nobili et al, 2010).
The aim of this study was to prepare and characterise two novel gold carbene complexes and to investigate in depth their main chemical and pharmacological features.
The gold(I) carbene complexes **1** and **2** were prepared adapting the procedure developed by Baker and Berners-Price (Baker et al, 2006). The compounds were characterized in the solid state by elemental analysis and IR spectroscopy. The solution chemistry of **1** and **2** was investigated through UV-Vis spectrophotometry and ^{1}H NMR, under physiologically relevant conditions. The cytotoxic effects of **1** and **2** were measured *in vitro* against the human ovarian carcinoma cell line A2780 sensitive to cisplatin (A2780/S) and its cisplatin-resistant cell subline (A2780/R) according to the procedure described by Skehan et al. (1990). For comparison purposes the cytotoxicity of cisplatin was evaluated in the same experimental conditions. Reactivity studies with cytochrome C, lysozyme and Atox-1 were monitored through ESI mass spectrometry (ESI-MS) and UV-Visible spectrophotometry according to protocols previously established (Casini et al, 2006).
Two novel gold carbene compounds, namely chlorido (1-butyl-3-methyl-imidazole-2-ylidene) gold(I) (**1**) and bis(1-butyl -3-methyl -imidazole-2-ylidene) gold(I) (**2**), were prepared and structurally characterised as potential anticancer drug candidates. These compounds consist of a gold(I) center linearly coordinated either to one N-Heterocyclic carbene (NHC) and one chloride ligand (**1**) or to two identical NHC ligands (**2**). Crystal structures were solved for both compounds, the resulting structural data being in agreement with expectations.
Solution studies revealed that these gold carbene complexes are highly stable in aqueous buffers, at physiological pH.
We investigated whether the presence of two tight carbene ligands in **2** might lead to biological properties distinct from those of the mono-carbene complex **1**. In spite of their appreciable structural differences, these compounds manifested a similar potent cytotoxic action *in vitro* when challenged against A2780 cells. Both compounds reached activities in the low micromolar range either in A2780S (<2 μM) and A2780R (<1 μM) after a 72 h drug exposure. Thus, both compounds were able to overcome resistance to cisplatin whose IC_{50} in A2780R was 23.24 μM.
The reactivity protein studies showed no adduct formation even upon long incubation (72h) with the model proteins cytochrome c and lysozyme; in contrast, both compounds were able to metalate the copper chaperone Atox-1 bearing a characteristic CXXC motif.
Based on these findings, it is proposed that the investigated gold carbene compounds are promising antiproliferative agents warranting a wider pharmacological evaluation. It is very likely that these gold compounds produce their potent biological effects mainly through selective metalation and impairment of a few crucial cellular proteins.

Baker et al. (2006) *Dalton Trans*. 3708–371
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Nobili et al. (2010) *Med Res Rev*. 3: 550-580
Skekan et al. (1990) *J Natl Cancer Inst*. 82: 1107-1112