Bioorganometallic Chemistry

DOI: 10.1002/anie.201002062

Metal Complexes as Protein Kinase Inhibitors**

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bioorganometallic chemistry · enzymes · iridium · inhibitors · N ligands

Metal complexes currently play a comparably unimportant role in medicinal chemistry and drug development with a few exceptions such as gold complexes for the treatment of rheumatoid arthritis and the blockbuster generation of platinum-based antitumor therapeutics.[1] The broad disregard of metal coordination species in drug design can be traced back to generalizations concerning the low stability of these types of agents for therapeutic purposes and their overall toxicity.

With the development of octahedral ruthenium(II) complexes bearing bidentate pyridocarbazole ligands, the group of Eric Meggers demonstrated that inert metal complexes can provide a powerful tool for the design of novel innovative drugs (Figure 1).[2–4] These complexes are potent inhibitors of protein kinases, a class of enzymes comprising numerous highly relevant biomolecular targets for anticancer drug development. Out of approximately 80 low-molecular-weight protein kinase inhibitors that have reached the clinical trial stage, 11 have already been approved by the U.S. Food and Drug Administration (FDA). The majority of these drugs represent competitive enzyme inhibitors replacing ATP in the respective binding site.[5] As all of the more than 500 different kinases require the co-substrate ATP for enzymatic catalysis and the ATP binding sites of kinases show a broad structural similarity, the design of kinase inhibitors faces a challenging selectivity problem. If other kinases (besides those involved in the uncontrolled growth of tumor tissues) are affected, undesired side effects cannot be avoided. The natural product staurosporine (Scheme 1) represents the lead structure of kinase inhibitors of the so-called indolocarbazole class. Staurosporine itself cannot be applied in clinical therapy as it is an extremely nonselective inhibitor with high general toxicity. However, the heterocyclic staurosporine core provides an excellent template for the development of more selective protein kinase inhibitors. Through appropriate structural variations it is possible to generate new derivatives that exhibit an optimized interaction with different ATP binding sites.[6] This strategy has been followed up by the Meggers group with the use of transition-metal complexes containing bidentate heterocyclic ligands of the indolocarbazole type.

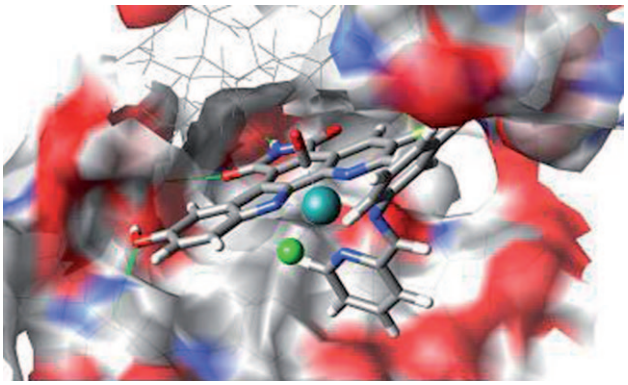
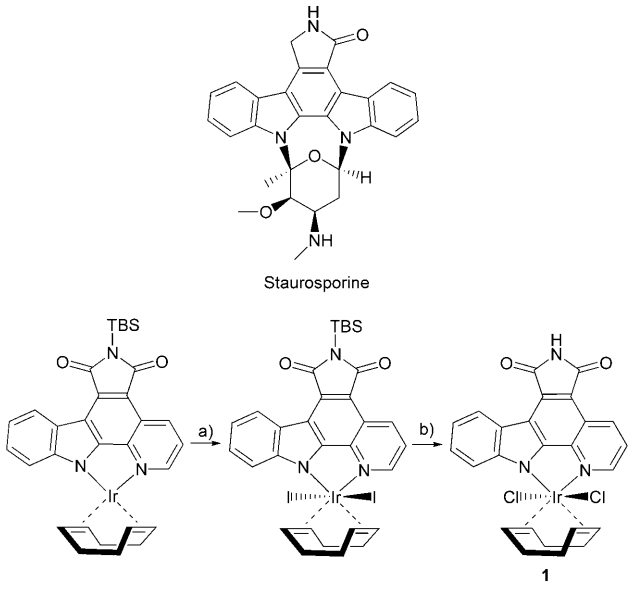


Figure 1. Ruthenium complex Λ-FL172 in the proteinkinase PAK1.[4] Hydrogen bonds are in green. Color code for the ions/atoms: Ru turquoise, Cl green. Visualisation with UCSF Chimera[7] (RCSB PDB file 3FXZ).



Scheme 1. Top: Staurosporine is a nonselective inhibitor of kinases, but pyridocarbazole–metal complexes with appropriately designed ligands achieve selectivity. Bottom: Key steps in the synthesis of **1**; a) oxidative addition, b) ligand exchange and removal of the *tert*-butyldimethylsilyl-protecting (TBS) group.

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[**] This research was supported financially by the Deutsche Forschungsgemeinschaft (research unit FOR630 "Biological Function of Organometallic Compounds").

Figure 1 shows the interaction of a representative ruthenium(II) pyridocarbazole species with the kinase PAK1 in a simplified manner.[4] While the lactam group of the pyridocarbazole ligand forms the characteristic hydrogen bonds linking the kinase and inhibitor, the ruthenium metal center and its additional ligands facilitate an optimized filling of the ATP binding pocket. Accordingly, an extended surface area of the active site can be addressed, which differs among the different kinases, and selectivity can finally be achieved. In this context, it is of interest to note that the metal center plays a purely structural role in orienting the interacting ligands. Consequently, the exact stereochemical position of the coordinated ligands plays a crucial role.

Meggers et al. now report on a series of octahedral iridium(III) complexes, which were obtained by a synthetic procedure featuring an oxidative addition as the key step. (Scheme 1).[8] Coordinative bonds with iridium(III) are generally rather inert and thereby provide stable structure elements for the development of potent enzyme inhibitors. Moreover, the target compounds were prepared by stereospecific oxidative addition reactions from the respective square-planar iridium(I) precursors, allowing excellent control of the required stereochemistry.

In an intensive screening against 229 protein kinases, the iridium complex **1** proved to be a potent and selective inhibitor of the kinase Flt4. This enzyme is involved in the formation of both lymphatic vessels and blood vessels (angiogenesis) and is overexpressed in various cancerous tissues. Increased activity of Flt4 could be related to the formation of lymphatic metastases and is associated with a poor prognosis for the respective patients.[9] The impact of the inhibition of Flt4 on metastasis reduction as well as the role of other kinase enzymes involved in vessel formation is the subject of ongoing intensive research efforts.[10] So far, monoclonal antibodies have been used for comparative studies on the effects of the selective inhibition of Flt4. Selective low-molecular-weight inhibitors such as **1** might significantly facilitate these research studies and provide valuable additional information.

One prerequisite for further research is that the compounds also exert their activity in a biological environment such as cell-based assays and experimental animal models. For this purpose the effects of **1** were investigated in vivo in two established zebrafish embryo models. So-called "zebrafish assays" offer a fast and efficient method to identify active lead compounds and to study the effect of these compounds on relevant biochemical pathways in vivo.[11] In the first model here, 24 h old embryos were treated with the test compounds and then examined under a microscope after an appropriate incubation period for defects in blood vessel formation. In the second model, human tumor cells are initially injected into the embryos. In the developing zebrafish embryos this triggers the formation of additional vessels, which are required to support the now-growing implanted tumor tissue (Figure 2).[12] Upon treatment with anti-angiogenic substances this tumor-cell-induced angiogenesis can again be reduced. As expected, compound **7** triggered significant anti-angiogenic effects in both assays, thus providing the proof of concept that this class of bioorganometallics is also effective in vivo. Overall, these results provide a strong basis for the development of iridium(III) complexes as potent inhibitors of angiogenesis.

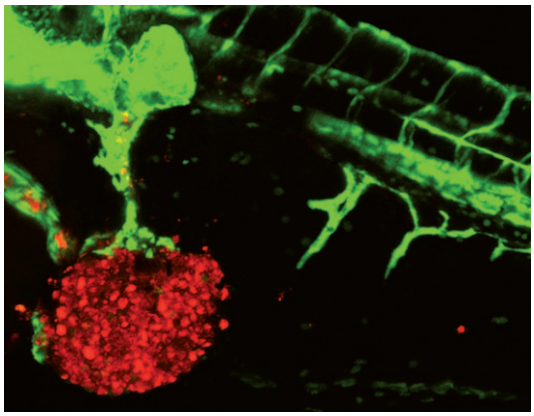

Figure 2. Tumor-cell-induced angiogenesis in the developing zebrafish embryo; the implanted human tumor cells are red fluorescent, blood vessels of the embryo are green fluorescent. The figure was provided by C. P. Bagowski.[12]

The future will show whether the traditional scepticism against the use of metal complexes in medicinal chemistry can be overcome and whether the design of inert metal species as demonstrated by Meggers et al. will become an established strategy for structure-based drug development. However, in any case these metal-based kinase inhibitors should be considered as valuable pharmalogical tools for the investigation of the physiological and pathophysiological roles of certain kinases such as Flt4.

Received: April 7, 2010
Published online: June 29, 2010

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