The small guanine nucleotide binding (GNB) protein Ras is a central component in cellular signalling networks controlling proliferation, differentiation or apoptosis. Alternating between an inactive GDP- and an active GTP-bound form Ras acts as molecular switch, which is regulated by guanine nucleotide exchange factors (GEFs) and GTPase activating proteins (GAPs). Ras with GTP bound strongly interacts with effectors such as Raf-kinase, RalGDS, and PI3K that transmit the incoming activation signal to downstream targets. $^{31}$P NMR spectroscopy allowed to identify two distinct conformational states when a nucleoside triphosphate (T) is bound, called state 1(T) and state 2(T). Ras in state 1(T) has a more than 2 orders of magnitude smaller affinity for effectors than Ras in state 2(T). In cancer research Ras represents a target of high interest due to its critical involvement in about 30% of all human malignancies (Bos 1989). Oncogenic Ras can neither be switched off by its intrinsic GTPase activity nor by GAPs leading to uncontrolled cell growth and thus to tumour formation.

Stabilizing the weak effector binding state 1(T) by small compounds represents a promising novel strategy for the inhibition of oncogenic Ras signalling. In fact, metal(II)-cyclens and their peptide conjugates exclusively recognize conformational state 1(T). We have determined the NMR-structure of this complex by n-dimensional NMR-spectroscopy [1].

The affinity of metal(II)-cyclens is much too small for pharmaceutical applications. A solution is fragment based drug design were two low-affinity ligands that bind in close proximity on the surface of the protein are linked to give a new compound with high affinity. In the PhD thesis these fragments will first be identified by in-silico ligand screening with LUDI, then verified by STD-NMR and the interaction sites will be identified by HSQC-NMR-spectroscopy and crystallography. New compounds will be designed by structure-aided drug design. In another PhD-thesis they will be synthesized (in cooperation with Prof. B. König) and tested for biological activity.


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