X-ray structure of a hydroxynitrile lyase from *Adenia racemosa*

N. Staunig, W. Höffken[1], K. Gruber and C. Kratky

Dept. of Chemistry, University of Graz, Heinrichstr. 28, 8010 Graz, Austria
[1]BASF-AG, GVC/C - A 30, 67056 Ludwigshafen, Germany

Hydroxynitrile lyases (HNLs) are plant enzymes that catalyse the cleavage of cyanohydrins into hydrocyanic acid and the respective aldehydes or ketones. HNLs possess a high potential for industrial applications, because they also catalyse the inverse of the natural reaction *i.e.* the enantioselective synthesis of cyanohydrins.

The (S)-HNL from Adenia racemosa (*Ar*HNL) is a glycosylated protein (Figure 1) with a molecular mass of 12.4 kDa. It is homologous (with 31% sequence identity and 60% similarity) to Boiling Stable Protein SP1 (PDB-entry 1TRO).

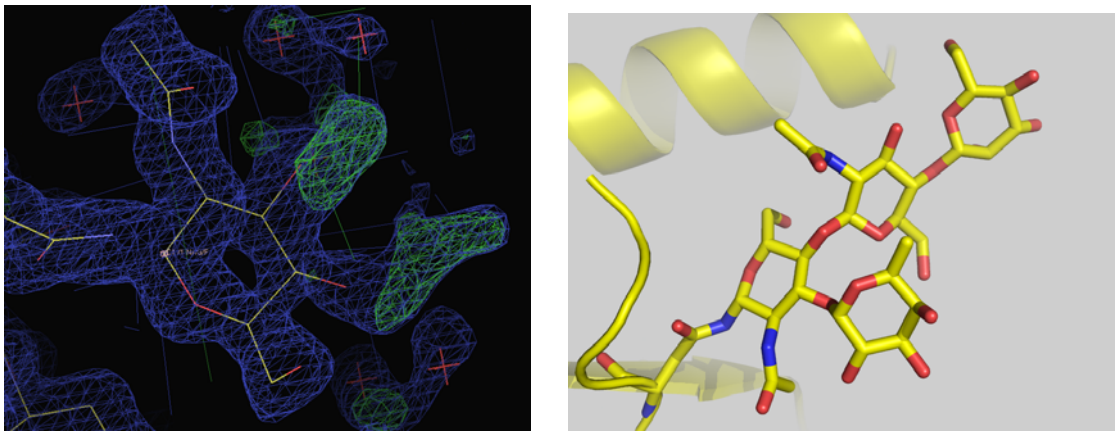

Figure 1: N-glycosylation at Asn-108 of *Ar*HNL.

*Ar*HNL was successfully crystallised by the sitting–drop technique at room temperature with sodium acetate, trifluoroethanol and PEG-6000 as precipitating agents. The thus obtained monoclinic crystals belong to the space group *C*2 with cell dimensions *a*=138.0 Å, *b*=52.3 Å and *c*=87.0 Å, β=126.70°. The structure was solved by molecular replacement and the refinement against data extending to a crystallographic resolution of 1.3 Å is still in progress. The current crystallographic R values are: R= 18.6% and R_{free}= 21.8%.

In the crystal, ArHNL forms a homo-dimer, with each monomer consisting of a four-stranded anti-parallel β-sheet packed against 3 α-helices. The main contact between the monomers occurs via the β-sheets (Figure 2). The relative content of α- and β-structures was also confirmed by CD-measurements.

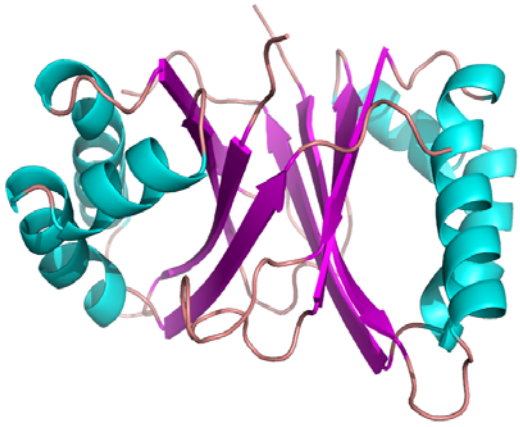

Figure 2: Molecular structure of *Ar*HNL.

Soaking experiments are in progress in order to elucidate the crystal structure of the enzyme substrate complex.

References