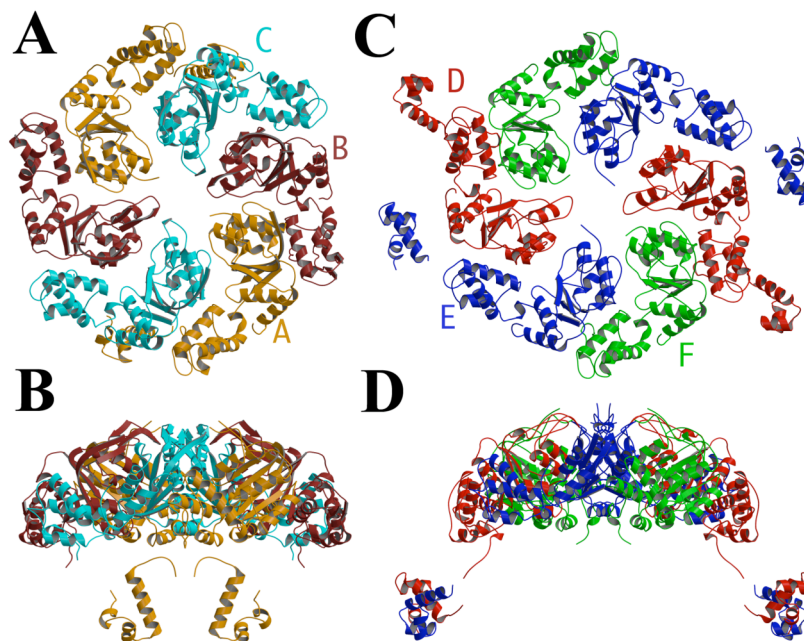


The  $\sigma^{54}$ -dependent transcription in bacteria is associated with various stress and growth conditions. Activators of the  $\sigma^{54}$  protein contain a central domain belonging to the AAA+ superfamily of ATPases, members of which function in diverse cellular processes in both prokaryotic and eukaryotic cells. We describe the X-ray structure of an N-terminal domain deletion of the ZraR protein from *Salmonella typhimurium*, which is a homologue of the general nitrogen regulatory protein NtrC, at 3 Å resolution. The structure reveals a hexameric ring that is typical for AAA+ containing proteins but which differs from the heptameric ring found in the crystal structure of the AAA+ domain of NtrC1 from *Aquifex aeolicus*. The dimerisation interface between DNA binding domains observed in the crystal structure suggests that dodecamers, rather than hexamers, might be the functionally important oligomer.



**Figure.** Overall structure of the AAA+ and DNA-binding domains. (A) and (C) show the two hexamers looking down the crystallographic two-fold parallel to **b**. The non-crystallographic three-fold operator is co-linear with the crystallographic two-fold. The molecules are coloured as follows: A in yellow, B in brown, C in cyan, D in red, E in blue and F in green. (B) and (D) show the rings side on with the DNA binding domains hanging under the mushroom shaped hexamers with the region containing the GAFTGA motif on the opposite side and in the centre of the ring. The other three DNA-binding domains are not visible in the electron density map.

The crystals belong to space group  $P222_1$  with  $a=107.44$ ,  $b=114.74$  and  $c=187.26\text{\AA}$ . The data collection details were reported in (Sallai et al., 2003). Data from the native and Se-Met crystals at the peak and inflection point wavelengths were collected at the ID-14-4 beamline at the ESRF in Grenoble using an ADSC Quantum-4 detector. The wavelengths were chosen on the basis of the X-ray absorption fluorescence spectrum. Data on the mercury derivative were collected at the X13 beamline at the EMBL in Hamburg using MAR165 CCD detector. The data were processed and reduced using the programs DENZO, SCALEPACK (Otwinowski and Minor, 1997) and TRUNCATE from the CCP4 program suite (Collaborative Computational Project, Number 4, 1994).

As described in (Sallai et al., 2003), initial phases for the structure determination were obtained by the SIRAS method, using the data from native and ethyl-mercuric phosphate (EMP) derivative crystals. Solvent flattening and six fold NCS averaging were initially applied to improve the phases and, consequently, the map quality.

### **Publications**

Sallai, L., Hendle, J. & Tucker, P.A.

X-ray crystallographic characterization and phasing of an NtrC homologue.

Acta Crystallogr D Biol Crystallogr 2003 Sep;59(Pt 9):1656-8.

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Crystal structure of the central and C-terminal domain of the sigma(54)-activator ZraR.

J Struct Biol 2005 Aug;151(2):160-70.