Similarities and differences in the modes of recognition of tRNAs and amino acids by their cognate aminoacyl-tRNA synthetases will be discussed with reference to crystal structures of three Thermus thermophilus class II synthetases: seryl-, lysyl- and histidyl-tRNA synthetases. The crystal structure at 2.7 Å resolution of T. thermophilus lysyl-tRNA synthetase complex co-crystallised with a seryl-adenylate analogue, reveals how the synthetase interacts with the acceptor stem of the cognate tRNA. A novel hydrophobic interaction between Phe-262 in the motif II loop and bases U68 and C69 in the major groove of the tRNA discriminates pyrimidines from purines in these positions. The mode of recognition is rather similar to that observed in the closely related synthetases will be discussed with reference to crystal structures of Bacillus stearothermophilus.

We are using isomorphous replacement with selenomethionine-substituted TrpRS (Doublié et al., 1994, Acta Cryst. A50:164-182), together with non-crystallographic symmetry averaging and maximum entropy solvent flattening to supplement phases from positioned fragments of the known model for structure determination. Conformational differences between the different TrpRS structures, including a detailed analysis of the composition and properties of nonpolar nuclei and microclusters by the method of Ilyin (1994 Prot. Eng. 7:1189-1198) will be described.

(These results will be described by NG GM48519-02)

PS04.06.10 STRUCTURES OF TRYPTOPHANYL-TRNA SYNTHETASE-LIGAND COMPLEXES Xin Huang and Charles W. Carter, Jr., Department of Biochemistry and Biophysics, CB 7260 University of North Carolina at Chapel Hill, Chapel Hill, NC 27514 USA

The crucial role of aminoacyl-tRNA synthetases (aaRSs) in maintaining the fidelity of the genetic code has motivated intense study of the sources of their specificity for cognate amino acids and tRNAs. Conclusions of the structural basis for coupling of specificity and catalysis of aaRSs can best be drawn by examining a series of different complexes involving the same enzyme. Bacillus stearothermophilus Tryptophanyl-tRNA synthetase (TrpRS) provides such a series. The success of the first structure in this series, that of TrpRS complexed with tryptophanyl-5′AMP has inspired us to study the structures of the complexes with other ligands such as Tryptophan (substrate) and 5′-O-[N-(L-tryptophanyl)sulfamoyl]adenosine (a stable analog of the tryptophanyl adenylate intermediate). We have synthesized 5′-O-[N-(L-tryptophanyl)sulfamoyl]adenosine and inhibition measurements showed that it is a strong inhibitor with K_i in the nanomolar range. Co-crystallization of this analog with TrpRS is underway. Translation and rotation searches are being carried out on the data from monoclinic crystals (space group P2_1) grown in the presence of tryptophan. Contrary to previous expectation (Carter et al., 1990, Acta Cryst. A46:57-68), our initial results revealed that what we previously interpreted as a non-crystallographic three-fold axis is actually a three-fold screw axis. We hope to make progress in solving these two structures.

(These results will be described by NIH GM48519-02)