INTRODUCTION

Transfer ribonucleic acids (tRNAs) play a central role in the complex mechanism of protein synthesis. In that process their chief function is to carry amino acids to the ribosomes, to decode the messenger RNA and to incorporate the correct amino acid into the growing polypeptide chain.

The tRNAs fulfill this function through a series of interactions with their biological partners:

- with the aminoacyl-tRNA synthetases, enzymes which catalyze the specific activation of an amino acid and its transfer to the cognate tRNA;
- with elongation factor Tu, which through the ternary complex aminoacyl-tRNA/GTP/EF-Tu, carries the aminoacyl-tRNA to the ribosome;
- with the mRNA involving the codon-anticodon interaction;
- with the ribosome at the A site in the pretranslocation step or at the P site in the posttranslocation step.

Due to these multiple interactions, tRNAs represent a particularly exciting model for the understanding at the molecular level of the mechanisms of recognition between nucleic acids and proteins and of structure-function relationships. In this chapter, we will focus on the tRNA-aminoacyl-tRNA synthetase interaction, which is essential because the correct attachment of amino acids to the 3'-end of tRNA, which is necessary for the fidelity of translation, relies on accurate recognition between these two macromolecules.

We will concentrate on a specific tRNA-aminoacyl-tRNA synthetase system, which has been extensively studied in our Institute: the tRNA\textsuperscript{Asp}-aspartyl-tRNA synthetase system from yeast. This system is particularly interesting because both partners, tRNA\textsuperscript{Asp} and aspartyl-tRNA synthetase, have been crystallized in their free and, more important, in their complexed state. In addition, tRNA\textsuperscript{Asp} could be a tempting model for studying tRNA-mRNA interaction, particularly since it has a self-complementary GUC anticodon. The resulting anticodon-anticodon interaction which mimicks the anticodon-codon interaction exists in solution and in the tRNA\textsuperscript{Asp} crystal structure. For all these reasons, the aspartic acid system from yeast is ideal for studying various types of fundamental tRNA recognition processes.
Fig 1: The nucleotide sequence of yeast tRNA<sub>Asp</sub>. For convenience the numbering system of the nucleotides is that of yeast tRNA<sub>Phe</sub>; in the 75-nucleotide long tRNA<sub>Asp</sub>, position 47 in the variable loop has been omitted. Non-classical Watson-Crick base pairs are indicated by broken lines.

**STRUCTURE OF tRNA**

The three-dimensional structures of two tRNA, yeast tRNA<sub>Phe</sub> and tRNA<sub>Asp</sub>, have been solved at high resolution. Both are elongator tRNAs with a short extra-loop. In this section, we will focus on the structure of tRNA<sub>Asp</sub> with reference to that of tRNA<sub>Phe</sub>.

**Primary structure**

The nucleotide sequence<sup>2</sup> of yeast tRNA<sub>Asp</sub>, shown in Figure 1, presents some characteristic features. It contains a high number of G-C base pairs, except in the D-stem where two G-U base pairs are present. The variable loop is made of four nucleotides versus five in tRNA<sub>Phe</sub> (for convenience of comparison we kept the same numbering, assuming a deletion at position 47). The D-loop has the same length as that of tRNA<sub>Phe</sub> but the two conserved Gs, which are crucial for D- and T-loops tertiary interactions, are at positions 17 and 18 instead of 18 and 19, thus making α and β regions of the D-loop quite symmetrical. Last but not least, the anticodon GUC presents the peculiarity to be self-complementary, with a slight mismatch at the uridine position. This feature was first noted by Grosjean et al.<sup>3</sup> who showed the existence of a significant interaction in solution and suggested it to be a tempting model to study tRNA-mRNA recognition.

**Crystal structure**

For yeast tRNA<sub>Asp</sub> two structures have been solved from a multiple