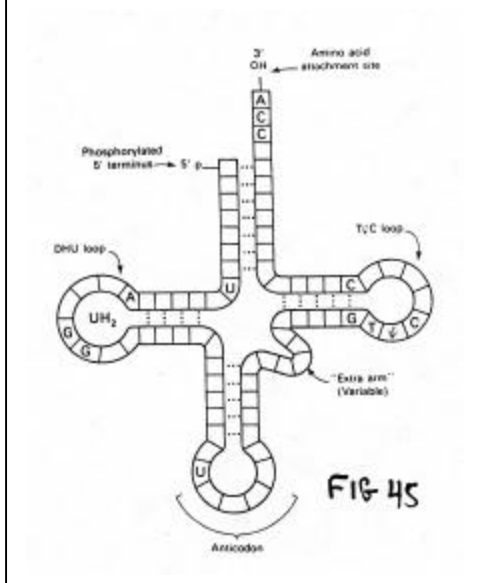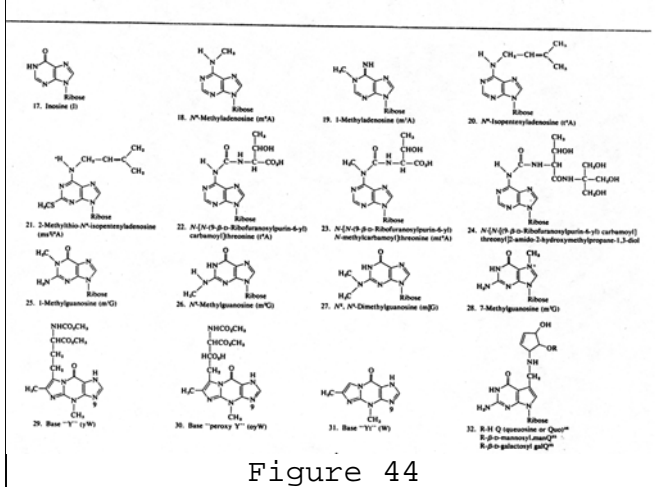RNA structure

The Structure of tRNA.

The role of tRNA is two fold, bind to and accept the correct amino acid from the appropriate tRNA synthetase and recognize and bind the ribosome-mRNA complex to deliver the amino acid to the growing polypeptide chain. Because of its central role in gene expression (and its relative abundance), tRNA is one of the best understood, most thoroughly studied biological macromolecules.
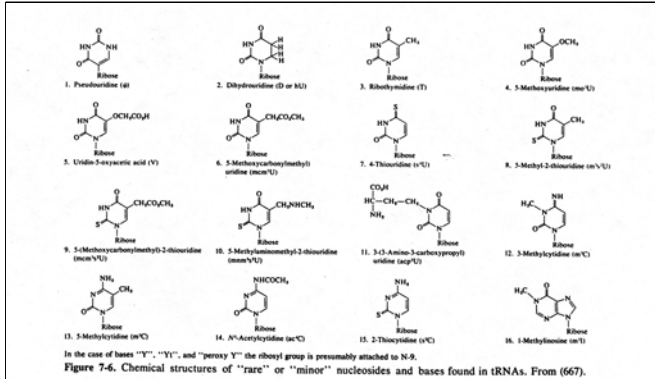
tRNAs are generally 75-90 nucleotides in length and contain not only the four normally occurring RNA bases, but also rare or minor variant bases. Some of these bases are given in the **(Figure 44)**. Most characteristic are the products of alkylating adenine guanine and cytosine amino groups; alkylations of the ring N of these bases, glycosylation and saturation of C=C bonds. As we shall see these modifications often restrict the type of base pairing these bases can assume and has interesting consequences for the tRNA tertiary structure.

Figure 44

FIG 45

Secondary structure

Based on base-base complementarity, the secondary structure of tRNA can be drawn as the familiar cloverleaf structure of 4 stems, each consisting of four to seven Watson-Crick type base pairs. Five regions of the tRNA are not base paired, the CCA acceptor stem, the D-loop (named for the presence of dihydrouridine), the anti-codon loop which interacts with the mRNA, the "extra arm" and the TYC loop (named for the presence of the pseudouridine base. **(FIGURE 45)**.
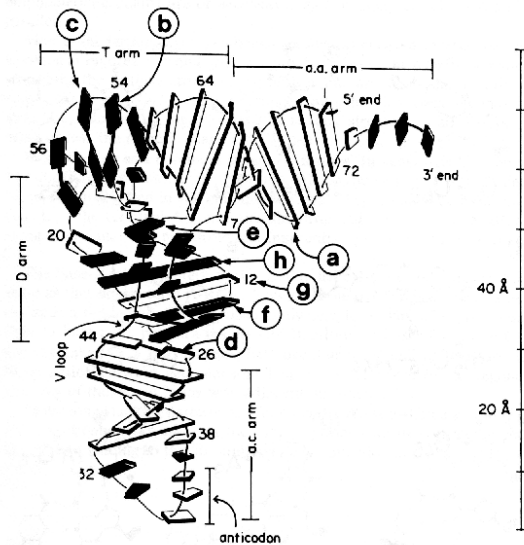
Tertiary Structure.

Figure 15-3 Illustration of base-pairing and stacking interactions in yeast tRNA^Phe.

Figure 46

The three dimensional structure of the tRNA molecule is not a cloverleaf, the molecule instead folds back on itself, forming two segments of double helix and most of the upaired bases in the T and D loops form base pairs with each other **(FIGURE 46)**. One of the two segments of double helix are formed by the stacking of bp region of the T and acceptor stems, the other by the stacking of the D-arm base paired region on the base paired part of the anticodon loop. This tertiary structure of the molecule is L-shaped and is held together by base pairs between the bases in the upaired regions of the stems and by unique stacking interactions **(Figure 47)**.
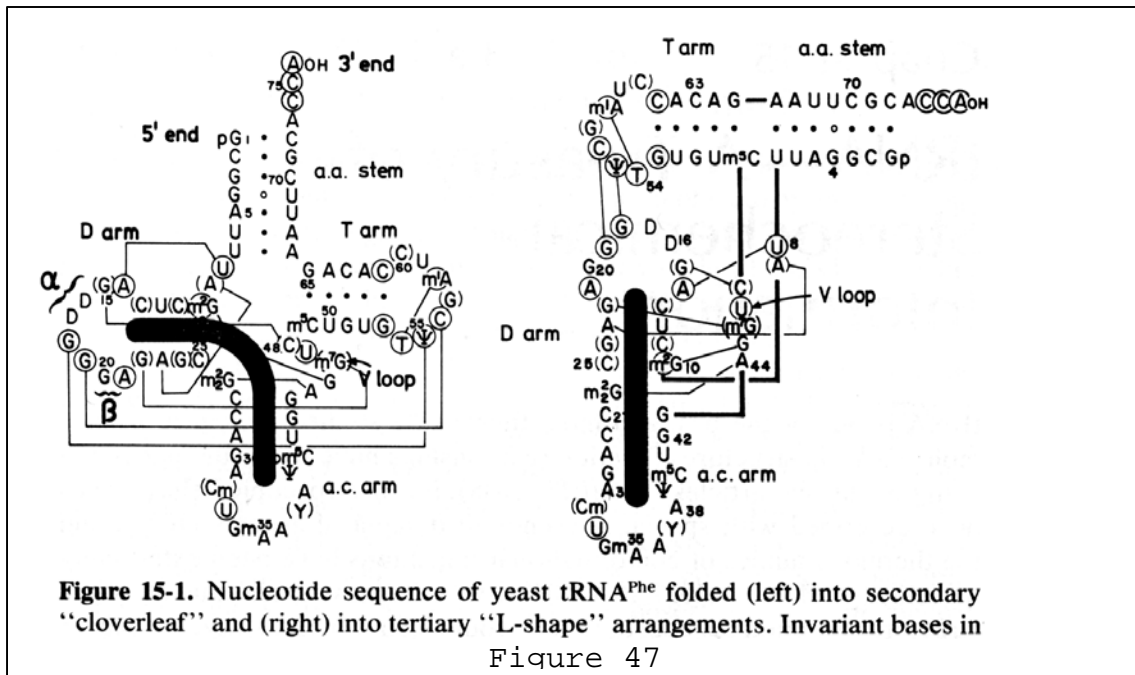

Figure 15-1. Nucleotide sequence of yeast tRNA^Phe folded (left) into secondary "cloverleaf" and (right) into tertiary "L-shape" arrangements. Invariant bases in

Figure 47

Base modifications and base pairings (see Figure 48)

m^1A_{58}-T_{54} This base pair is a reversed Hoogstein type, with the chains from each base pair running in an antiparallel direction. The m^1A is unable to form a normal Watson-Crick base pair because the N1, normally a W-C acceptor is methylated, rendering it refractory to H-bonding.

$m^2_2G_{26}A_{44}$ is a purine-purine base pair who normally pair in a wobble fashion. However, this configuration is altered due to the presence of the bulky methyl groups on G N2. This pushes this base pair further apart, more than even in the usual wobble situation. This base pair is at the junction of the anticodon and D-stems, and is stacked with both and is thus responsible for the $26°$ kink between these two helices

2

Figure 15-4 Unusual base–base interactions found in yeast tRNA^{Phe}. For their positions in the tertiary structure, see Figure 15-3. All other base-pairs are of the Watson–Crick type. Flags at glycosyl links indicate backbone orientations. They are parallel in c, e, f(m²G₁₀, G₄₅), g(A₉, A₂₃), h(C₁₃, m⁷G₄₆) and antiparallel otherwise. All of the dimeric base–base interactions are already contained in Figure 6-1.
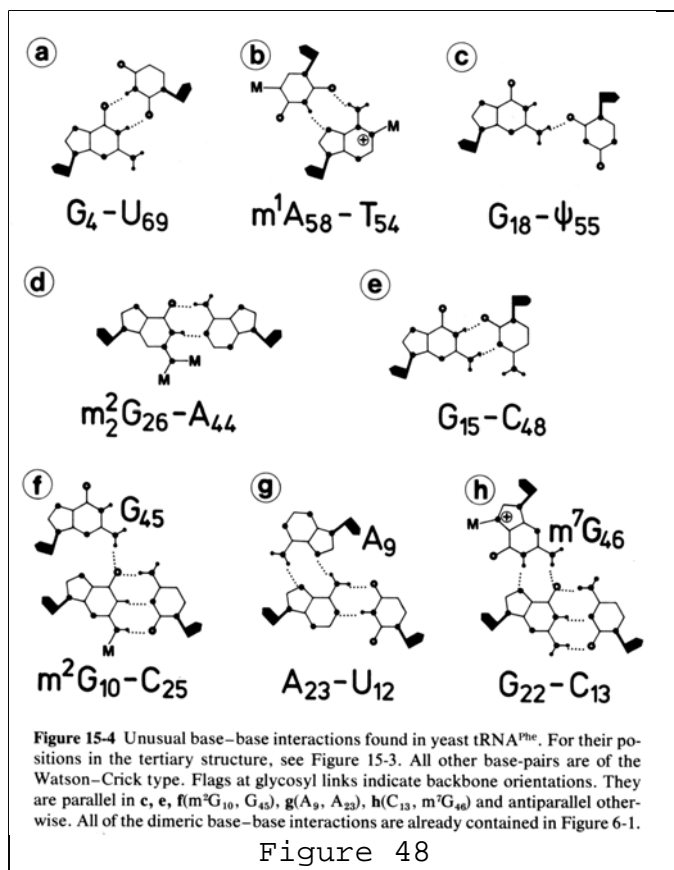
Figure 48

A number of triplet interactions are formed which also help hold the structure together **(Figure 48)**.

Primary sequence conservatism gives all tRNAs the same structure. An examination of the places in tRNA where the base are conserved turns out ot be in the non-base paired loops. As we have juest seen, these are the bases which are involved in stabilizing the tertiary structure of the tRNA molecules **(Figure 47 & 48)**.

Base stacking

The strength and utiliity of base stacking to hold together the structure of a polynucleotide is illustrated by tRNA. Only 42 of the 76 bases in the yeast tRNA^{phe} are involved in A-RNA double helical structures. Yet, however, 71 of the 76 are involved in stacking interactions. Of the five that are not, one is the terminal A in the acceptor stem and the two others are the modified dihydrouridines in the D-stem, which are non-planar, non-aromatic and hence oppose stacking. The base stacking in the double helical regions resembles that we have seen in A-DNA. A very interesting stacking interaction is seen where the individual bases of one strand are tucked between the bases of an adjacent strand. These interactions occur where three strands meet; inside the L corner and in the T-loop region.
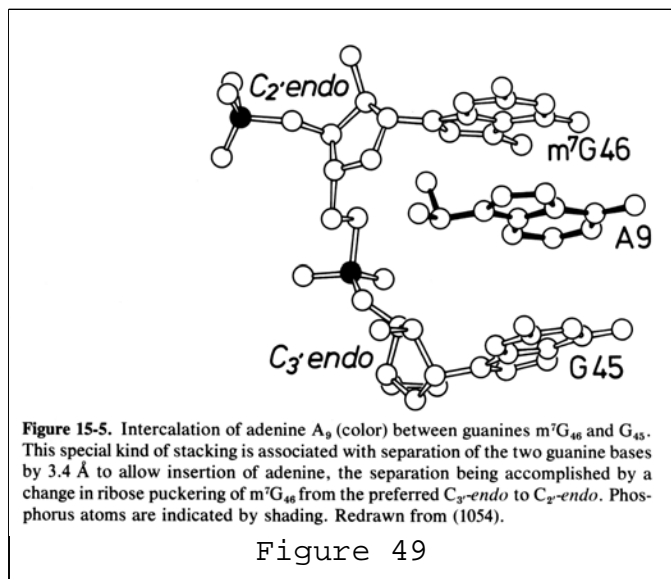

Figure 15-5. Intercalation of adenine A₉ (color) between guanines m⁷G₄₆ and G₄₅. This special kind of stacking is associated with separation of the two guanine bases by 3.4 Å to allow insertion of adenine, the separation being accomplished by a change in ribose puckering of m⁷G₄₆ from the preferred C₃'-endo to C₂'-endo. Phosphorus atoms are indicated by shading. Redrawn from (1054).

Figure 49

FIGURE 49 shows the case of A₉ intercalated between the bases G₄₅ and m⁷G₄₆. Because the base inserts between the two others, and the base has finite vander Waals distance (this is a stacking interaction after all), the receiving bases must move apart by at least 3.4A. For this to occur, the ribose phosphate backbone must readjust accordingly. The most obvious way for the chain to readjust is by changing its sugar pucker, such that the bases can separate-a change from the A-RNA C3'endo to the more B-DNA-like C2'endo. As you should recall this increases the intrastrand phosphate-phosphate distance from 5.9 to 7.0 (just like in B-DNA).