Pseudouridine Modification in RNA

Introduction of pseudouridine

Many RNAs in nature have undergone post-transcriptional modifications, which play an important role in the structure, function, and metabolism of RNA. More than 170 types of RNA modifications have been discovered, among which pseudouridine modification is the earliest and most abundant RNA modification, so it is also called the "fifth base". Pseudouridine is the 5-position ribose isomer of uridine (see figure 1), which is widely present in a variety of RNAs (tRNA, rRNA, snRNA, snoRNA, etc.). Pseudouracil modification generally results from the isomerization of uridine. With the development of sequencing technology in recent years, it has been found that there are abundant pseudouridine modifications on mRNA. Pseudouridine is a natural structural analogue of uridine. Compared with uridine, ribose is not connected to uracil N1, but to C5 of the pyrimidine ring. The isomerization of pseudouridine does not affect the classic base pairing, but it turns the N1 position into a proton donor. This may be because pseudouridine has different characteristics from other bases and can make many RNA structures stable.

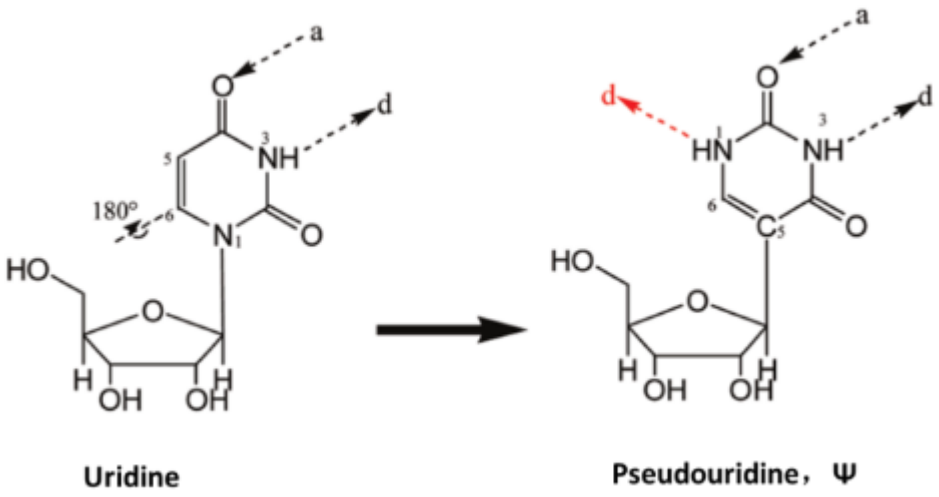

Figure 1

"Uridine and pseudouridine: d is the hydrogen bond donor, and a is the acceptor.

(DOI: 10.13376/j.cbls/2014037)

Functions of pseudoUridine modification

1. Pseudouridine modification in rRNA

   Pseudouridine modifications in rRNA are commonly found in the large and small subunits of rRNA, and are mainly distributed in functionally

important regions, such as the peptidyl transferase center (PTC), decoding center, and A-site finger region (ASF). The distribution implies that pseudouridine modification can affect the function of rRNA. Pseudouridine modification in these regions may affect the folding of rRNA, the assembly of ribosomes, and the maintenance of the corresponding high-level structure.

2. Pseudouridine modification in mRNA

   Existing studies have shown that pseudouracilylation of mRNA has three main functions: changing codons, enhancing transcript stability and stress response. The process of mRNA pseudouracil modification is catalyzed by pseudouracil synthases (PUS), which changes the chemical structure of uracil nucleotides (U) to form pseudouracil nucleotides. In 2011, Yi-Tao Yu's laboratory at Rochester University discovered that the introduction of pseudouridine modification in the stop codon can turn the stop codon into a sense codon. After the in vitro transcribed mRNA containing the pseudouridine modification is injected into the body, the translation rate and stability of the RNA containing the modification will increase compared with the mRNA without the pseudouridine modification.

3. Pseudouridine modification in snRNA

   The spliceosome formed by snRNA and a series of proteins plays an important role in the splicing process of pre-mRNA. Pseudouridine modification has occurred in basically all snRNAs, and these modifications are highly variable among different species. Pseudouridine modifications in snRNA are mainly distributed in functionally important areas. And the current research shows that the pseudouridine modification in snRNA mainly plays a role in the shearing process through the interaction between RNA and RNA. The pseudouridine modification in snRNA plays an important role in the formation of snRNP, the assembly of spliceosome, and the splicing of pre-mRNA.

Application of pseudouridine modification

When synthesizing RNA in vitro, use 5-methylcytidine instead of cytidine and pseudouridine instead of uridine to modify the bases of RNA, and then add interferon inhibitors to the cell culture medium. These measures can limit the activation of the host cell's innate immune system by exogenous nucleic acid, and make the exogenous nucleic acid play a corresponding role in the cell. When the mRNA vaccine is injected into the human body, the human body's immune response to the mRNA vaccine is mainly related to uridine (partly composed of uracil). Using pseudouracil instead of uracil can reduce the immune system's recognition of mRNA. mRNA vaccine manufacturers Moderna and BioNTech

both use pseudouracil modification to maintain mRNA stability. Pseudouridine has been widely used in the production of mRNA drugs. ψ-seq can locate the modification sites of pseudouracil with single nucleotide accuracy. However, the exploration of mRNA modification sites has just begun, and the limitations of detection technology still need to be broken through.