

What are Open Reading Frames?

To make a protein, ribosomes must perform translation on mRNA and assemble amino acids into a polypeptide chain. Ribosomes initiate translation at the start codon, translocate along the mRNA to make a polypeptide, and stop when they reach the stop codon. Open reading frames (ORFs) are the region of an mRNA from the start codon to the stop codon. Different orders of nucleotides in an ORF code for different proteins and protein expression, so studying these nucleotide orders can reveal why certain proteins are expressed or why cells function in certain ways.

However, not every ORF makes a full protein. In recent years, drug developers have been studying small ORFs (sORFs), which are 100 or less codons long and produce micropeptides. In addition, sORFs impact the gene regulation of the protein coding sequences (CDS) in the transcriptome. There are four main types of sORFs.



Upstream ORFs

Upstream ORFs (uORFs) are in the 5' UTR and regulate translation in the downstream CDS. They regulate translation by preventing scanning ribosomes from reaching the CDS, usually by triggering ribosome dissociation, stalling the ribosome, or initiating mRNA decay. uORFs are the most abundant type of sORFs and found in nearly half of human CDS's.



Downstream ORFs

Downstream ORFs (dORFs) are in the 3' UTR and enhance the translation efficiency of the CDS. The exact mechanism of how dORFs do this is still being studied, but scientists suggest the enhanced translation efficiency may have to do with dORFs ability to recruit translation factors, especially during transcript looping.



Internal ORFs

Internal ORFs (intORFs) are located within the CDS, either in or out of frame of a canonical transcript. They code alternative proteins rather than the primary protein produced in the CDS. intORFs are translated when a ribosome skips the mRNA's 5' start codon and starts translating in the middle of the ORF, but once translation begins, the intORF is translated similarly to other coding regions of the RNA.



Long noncoding ORFs

Long noncoding ORFs (lncORFs) are located on long noncoding RNAs (lncRNAs). lncRNAs do not code for proteins, but scientists are learning that they impact many cellular functions. Scientists are also discovering micropeptides translated from lncRNAs, suggesting that while lncRNAs don't produce proteins, lncORFs produce functional peptides.



Locating small ORFs at Eclipsebio

Since small ORFs impact gene expression, they can offer insights into how an RNA therapeutic functions. They are also promising targets for effective RNA therapeutic use. At Eclipsebio, our **eRibo Pro** assay reveals where translation occurs on an RNA, locating where ORFs are on the transcriptome to understand how translation is happening and potential targets from RNA therapeutics.

Interested in learning about ORFs in your RNA? [Contact us](#) to get started.