

# ENFINIA™ IVT Ready DNA

## Specifications and Submission Guidelines

ENFINIA IVT Ready DNA is linear, double-stranded, NGS-verified DNA manufactured in the United States by Elegen and ready to ship in as few as 10 business days. Our proprietary cell-free DNA synthesis platform delivers sequences of 1 kb to 5.5 kb in length, complete with poly(A) tail, eliminating the need to assemble, clone, sequence, and linearize DNA. These ready-to-transcribe DNA templates will enable you to rapidly synthesize and screen mRNA with confidence.

### SPECIFICATIONS

1. ENFINIA IVT Ready DNA can be ordered at a standard synthesis yield of 10 µg as measured by Thermo Quant-iT assay.
2. ENFINIA IVT Ready DNA is available in lengths ranging from 1,000 - 5,500 bp, excluding the poly(A) tail.
3. ENFINIA IVT Ready DNA can be produced with a continuous or segmented poly(A) tail. When placing an order on our portal, these options will be available in a dropdown menu.

Continuous Tails	A70	A90	A100	A110	A120	A130
Segmented Tails	A30-Linker-A70 (two deoxyadenosine homopolymers separated by a 10 bp UGC Linker, 5'-GCATATGACT-3') <sup>1</sup>			A30-Linker-A90		

4. ENFINIA IVT Ready DNA is delivered as dried-down double-stranded DNA in a 96-well microplate, one sequence per well.
5. Each ENFINIA IVT Ready DNA sequence includes a 22 bp adaptor on the 5' end (5'-GCGAGTCTTAGCCTGCGACGCT-3'). There is also a 3-T overhang on the 5' strand of the poly(A) tail.
6. Each ENFINIA IVT Ready DNA sequence is NGS-verified before shipment.

1. Chaudhary, N. 2021. *mRNA vaccines for infectious diseases: principles, delivery and clinical translation*.

## SEQUENCE SUBMISSION ACCEPTANCE CRITERIA

	Standard Complexity	High Complexity
Sequence Length (without poly(A) tail)	1,000 - 5,500 bp	1,000 - 5,500 bp
Overall GC Content	25 - 65%	25 - 75%
100 bp GC Content	22 - 75%	12 - 83%
Local GC Variation	up to 60%	up to 70%
Repeats	up to 20 bp	up to 150 bp
Homopolymers* (not including poly(A) tail)	Up to 7 bases for G/C Up to 8 bases for A/T	Up to 15 bases for G/C Up to 30 bases for A/T

1. DNA sequences must be 1,000 - 5,000 bp including the promoter, ORF, and both UTRs, but excluding the poly(A) tail. For sequences shorter than 1,000 bp, a random sequence of nucleotides can be added upstream of the promoter to extend the length to 1,000 bp. Please note that this added sequence will not be transcribed to mRNA.
2. The DNA sequence may contain either a **PaqCI** recognition site (5'-**CACCTGC**-3' or 5'-**TACCTGC**-3') or a **BsaI** recognition site (5'-**GGTCTC**-3'), but not both. Sequences containing the **BspQI** recognition site (5'-**GCTCTTC**-3') are not permitted. The above also applies to the reverse complements of the restriction enzyme recognition sites.
3. The DNA sequence must not contain a homopolymer of 10 or more deoxyadenosine (A) bases within the last 30 bp of a sequence.
4. The provided sequence (comprising the promoter, UTRs, and ORF) cannot end in an deoxyadenosine (A).

*\*Elegen's QC methods currently used for ENFINIA IVT Ready DNA may not reliably measure homopolymer sequences longer than 10 bp, except for the poly(A) tail. An accepted sequence containing a homopolymer that is longer than 10 bp may contain molecules where the homopolymer is shorter or longer than expected.*

