

Insect Cell-Free Protein Expression

TnT[®] T7 Insect Cell Extract Protein Expression System

- Express protein in only a few hours
- Use DNA-based templates for expression
- Compatible with multiple detection methods



Promega

TnT[®] T7 Insect Cell Extract Protein Expression System

The TnT[®] T7 Insect Cell Extract Protein Expression System is a convenient, quick, single-tube, coupled transcription/translation system for cell-free protein expression. Protein synthesis reactions are initiated by the addition of a DNA template, eliminating the need for time-consuming *in vitro* RNA synthesis.

The extract is made from the commonly used *Spodoptera frugiperda* Sf21 cell line. All components necessary for the transcription/translation are present in the TnT[®] T7 ICE Master Mix. To initiate protein synthesis, the only component that must be added is the DNA template. Reactions are incubated at 28-30°C and are complete within 4 hours (Figure 1).

Proteins are expressed from genes cloned downstream of the T7 promoter. Companion vectors have been designed to achieve optimal yield with TnT[®] T7 ICE System. They contain untranslated region (UTR) sequences at the 5' and 3' ends of the gene coding region to enhance translation efficiency. Using the TnT[®] T7 Insect Cell Extract Protein Expression System and these vectors, 75µg/ml of functional protein can be produced (Figure 3).

Obtain data faster

Protein is expressed in only 4 hours—not days as with cell-based expression (Figure 2)

Complete system

No requirement to purchase additional reagents

Achieve high protein yields

Express up to 75µg/ml of protein for multiple applications

Flexible

Detect expressed proteins by coomassie staining or incorporation of a fluorescence or biotinylated modified tRNA

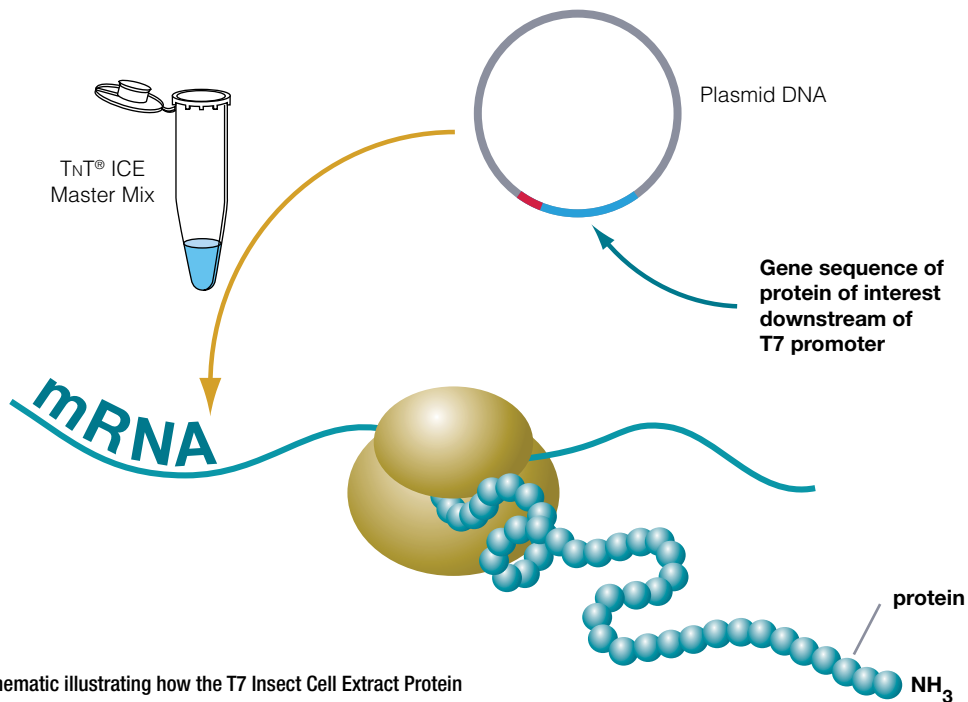


Figure 1. Schematic illustrating how the T7 Insect Cell Extract Protein Expression System functions.

Obtain Data Faster

No need to wait days to determine if your protein is expressed. Efficiently screen multiple clones by just adding DNA template containing the protein-coding sequence of interest downstream of the T7 promoter directly to the TnT[®] T7 ICE Master Mix and incubate for 4 hours (Figure 2).

TnT[®] T7 Insect Cell Extract Protein Expression System

Add DNA directly to TnT[®] T7 ICE Master Mix, express protein

4 Hours

Analyze protein

Total: 4 Hours

Cell-based baculovirus expression

Co-transfect insect cells with linearized viral DNA and recombinant vector

4 DAYS

Select plaque and replate

4 DAYS

Choose plaque for expression and amplify

3 DAYS

Titer viral stock

4 DAYS

Infect insect cells and express protein

3 DAYS

Analyze protein

Total: 18-21 DAYS

Figure 2. Time comparison of the TnT[®] T7 Insect Cell Extract Protein Expression System and cell-based baculovirus expression of recombinant proteins.

**Express
Proteins
in Hours
Not Weeks**

Achieve High Yields

The T7 Insect Cell Extract System is designed to express up to 75µg/ml of protein when used with the pF25 T7 Flexi Vectors. The high yield gives you additional protein for functional screening applications, which would not be possible with low yield standard cell-free eukaryotic systems (Figures 3 and 4).

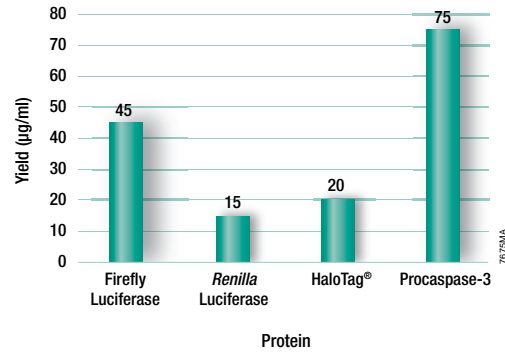


Figure 3. Typical protein yields using TnT[®] T7 Insect Cell Extract Protein Expression System.

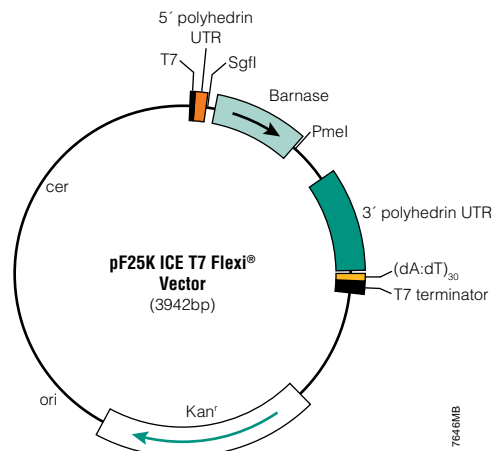
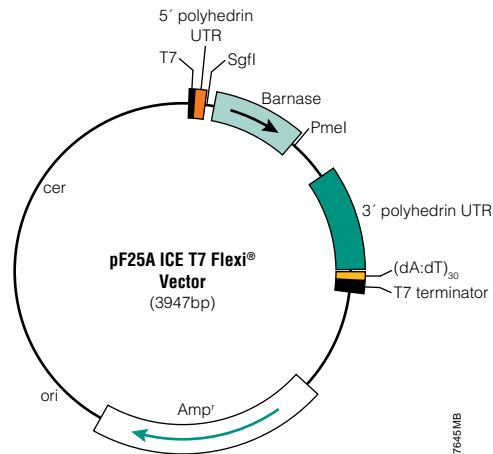


Figure 4. pF25A T7 Flexi[®] and pF25K T7 Flexi[®] vectors.

Ordering Information

Product	Size	Cat.#
TnT® T7 Insect Cell Extract Protein Expression System	40 reactions	L1102
	10 reactions	L1101
pF25A T7 Flexi® Vector	20µg	L1061
pF25K T7 Flexi® Vector	20µg	L1081

Related Products

Product	Size	Cat#
FluoroTect™ Green _{lys} in vitro Translation Labeling System	40 reactions	L5001
Transcend™ Chemiluminescent Non-Radioactive Translation Detection System	1 System	L5080
Transcend™ Colorimetric Non-Radioactive Translation Detection System	1 System	L5070
Transcend™ Biotinylated tRNA	30µl	L5061

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