

**T7 RNA Polymerase Transcription Buffer Set**

v. 250701

**Catalog number** C15027-K01 / C15027-K02

<b>Package &amp; Component</b>		<b>C15027-K01</b>	<b>C15027-K02</b>
	T7 RNA Polymerase (200 U/μL)	10,000 U	25,000 U
	10X RNA Polymerase Reaction Buffer A	0.5 mL	1 mL
	10X RNA Polymerase Reaction Buffer B	0.5 mL	1 mL
	10X RNA Polymerase Reaction Buffer C	0.5 mL	1 mL
	10X RNA Polymerase Reaction Buffer D	0.5 mL	1 mL
	10X RNA Polymerase Reaction Buffer E	0.5 mL	1 mL
	10X RNA Polymerase Reaction Buffer F	0.5 mL	1 mL
	10X RNA Polymerase Reaction Buffer G	0.5 mL	1 mL
	10X RNA Polymerase Reaction Buffer H	0.5 mL	1 mL
	10X RNA Polymerase Reaction Buffer I	0.5 mL	1 mL
	100 mM DTT	0.5 mL	1 mL

**Description**

The T7 RNA Polymerase Transcription Buffer Set contains a T7 RNA Polymerase and a set of seven 10X RNAP reaction buffers. This set of reagents was specifically designed for the selection of efficient transcription reactions. The user simply combines DNA template, NTPs, T7 RNA polymerase, and the 10X polymerase reaction buffer to proceed following reactions.

Standard RNA synthesis procedures:

1. Below reaction mixture should be prepared under room temperature and combined in the following order:

**Manuel**

<b>Component</b>	<b>Amount</b>	<b>Final concentration</b>
<b>Nuclease-Free H<sub>2</sub>O</b>	X μL	-
<b>Template DNA</b>	0.5-1 μg	-
<b>10X RNA Polymerase Reaction Buffer</b>	2 μL	1 X
<b>ATP (100 mM)</b>	0.6 μL	3 mM
<b>UTP (100 mM)</b>	0.6 μL	3 mM
<b>CTP (100 mM)</b>	0.6 μL	3 mM
<b>GTP (100 mM)</b>	0.6 μL	3 mM
<b>100 mM DTT</b>	2 μL	10 mM

<b>T7 RNA Polymerase (200 U/μL)</b>	1 μL	-
<b>RNase inhibitor (optional)</b>	0.5 μL	1 U/μL
<b>Inorganic Pyrophosphatase (optional)</b>	0.5 μL	0.0025 U/μL
<b>Total reaction volume</b>	20 μL	-

2. Incubate at 37°C for 30 minutes to 2 hours.

3. Above reaction mixture may be scaled up or down proportionately.

**Storage**

Store at -20°C for up to 6 months, and it is recommended to store at -80°C for long-term preservation. Avoid repeated freeze/thaw cycles.

**Handling  
Instruction**

For optimal storage, aliquot the enzyme, reaction buffer and DTT reagent into smaller quantities and store at recommended temperature. Avoid extended exposure to ice; instead, promptly retrieve the required portion and return it to the appropriate storage temperature.

**Notes**

1. Transcription reaction should be performed under RNase free condition. Use nuclease-free tubes, reagents, and water to avoid RNase contamination. Also, wear gloves when working with RNA.
2. To obtain optimal condition, NTP concentration can be titrated between 3 – 5 mM.
3. The volume of T7 RNA Polymerase can be titrated between 1-2 μL in the IVT reaction to optimize your assay.

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