

1.6 Recipes

CB5X

	RRL		WG	
	volume	final	volume	final
1 M HEPES-K pH 7.5	400 uL	80 mM	400 uL	80 mM
1 M mgCl ₂	75 uL	15 mM		
1 M MgAc ₂	70 uL	14 mM		
0.1 M spermidine	100 uL	2 mM	100 uL	2 mM
water		425 uL		430 uL
TOTAL		1 mL		1 mL

4 NTP (5X)	UNCAPPED	CAPPED
0.1 M ATP	150 uL	150 uL
0.1 M CTP	150 uL	150 uL
0.1 M UTP	150 uL	150 uL
0.1 M GTP	150 uL	15 uL
WATER	400 uL	535 uL
TOTAL	1.0 mL	1.0 mL

adjust pH to 7.0 with 2 M Tris base

SP6 pol dilution buffer

50 mM Tris-Cl pH 7.5	250 uL of 2 M
100 mM NaCl	250 uL of 4 M

0.1 mM EDTA	2.0 uL of 0.5 M
7.0 mM DTT	70 uL of 1.0 M
50% glycerol	2.5 mL
0.1% Triton X-100	5.0 uL
Add water to total = 5.0mL	

Cap (10X)L: 25 A250 units = 1.2 mg 300 æL 20 mM Tris pH 8

DTT (10X) 0.1 M

tRNA 10 mg/mL

SP6 RNA polymerase:

Storage buffer

50mM Tris-HCl pH 7.9	250uL of 2M
100mM NaCl	250uL of 4M
0.1mM EDTA	2uL of 0.5M
7mM DTT	70uL of 1M
50% glycerol	2.5mL
0.1% Triton X-100	5uL make to 5mL with dH2O