

Protocol : Restriction enzyme and buffer table

Enzymes	Buffer 1	Buffer 2	Buffer 3	Buffer 4
EcoRI	√	√	√	√
PstI			√	
XbaI		√		√
SpeI		√		√
NdeI		√		√
MfeI				√

Enzymes	Buffer 1	Buffer 2	Buffer 3	Buffer 4
EcoRI & PstI			√	
EcoRI & SpeI		√		√
XbaI and PstI		√	√	
EcoRI, PstI & DpnI			√	

Enzyme concentrations:

Enzymes	Amount
EcoRI-HF	20 U/μl
PstI	20 U/μl
XbaI	20 U/μl
SpeI	10 U/μl

It is possible to reduce the volume of EcoRI-HF; PstI and XbaI to 0.5 μl as 5-10 units is sufficient for a 50 μl digestion mixture.

By definition, 1 unit of restriction enzyme will completely digest 1 μg of substrate DNA in a 50 μl reaction in 60 minutes.