

End-Labeling of 3'-recessed restriction sites

1 ug of digested DNA	x.x ul
Klenow fragment (0.5 Unit)	1.0 ul
10x nick translation buffer (See Maniatis)	2.5 ul
P ³² -labelled dCTP (400-3000 uCi/mM)	2.0 ul
0.5 mM dATP	1.0 ul
0.5 mM dTTP	1.0 ul
0.5 mM dGTP	1.0 ul
Water	<u>xx.x ul</u>
Final volume	25.0 ul

Let reaction proceed for 15-30 minutes, bring volume up to 100 ul with STE, and run through a spun column equilibrated with STE (See Maniatis). Measure activity on liquid scintillation counter. For blotting, load 6000 cpm.

See Focus 6(1):6. 1984.