$^{32}$P Labeling of Large DNA Fragments:

1. Purify DNA fragment from gel and then $1:10$ with ethanol/CHCl$_3$

extract a couple of times to remove residual agarose

2. Aliquot 200-500ng of DNA into screw cap tube and add ddH$_2$O to a final volume of 40ul

- DNA - 10ul (will vary)
- ddH$_2$O - 30ul
- Primers - 20ul
  (herring/turkey)
- 5x dCTP Buffer - 20ul
- $[^{32}P]-dCTP$ - 20ul
- Klenow - 2ul

\[ \text{total: 102ul} \]

- Incubate tube at $37^\circ \text{C}$ for 15-30 mins
- Add stop 2ul and mix gently
- Centrifuge to get labeled DNA to the bottom
  (will condense in cap)
- Nuclease to clean up
- Count probe 1ul spotted on filter etc.
- Boil 5-10 mins before use.