Vector Dephosphorylation

This is to be used for VECTORS ONLY. Do NOT dephosphorylate your insert!

1. Gel purify your linearized vector and quantify using Nanodrop before beginning.
2. To your remaining solution, you will need to add Antarctic Phosphatase and the corresponding Ant Phos Buffer. We will always use 1ul of enzyme. Use the following formula to calculate volume of buffer to add:

$$Volume\ of\ buffer\ (\mu L) = \frac{(Volume\ of\ eluant\ in\ mix) + 1}{9}$$

3. Add buffer first, followed by enzyme – make sure to mix both.
4. Calculate & record new concentration of DNA using the following formula:

$$Dephos.\ vector\ conc. = (purified\ dig.\ conc.)\left(\frac{volume\ of\ eluant}{total\ vol.\ of\ eluant + enzyme + buffer}\right)$$

5. Incubate at 37°C for 30-60 minutes, then heat kill at 70°C in water bath for 10-20 minutes. (NOTE: This heat killing step is crucial to a successful ligation!)
6. Proceed directly to ligation (no need to purify) or store at -20°C until needed.