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Anza™ Alkaline Phosphatase Kit	500 reactions	IVGN220-4
Anza™ T4 PNK Kit	50 reactions	IVGN230-4
Anza™ DNA Blunting Kit	100 reactions	IVGN240-4
Anza™ DNA End Repair Kit	20 reactions	IVGN250-4
One Shot™ TOP10 Chemically Competent <i>E. Coli</i>	20 reactions	C4040-03
One Shot™ INV110 Chemically Competent <i>E. Coli</i>	20 reactions	C7171-03
Select Agar, powder	500 g	30391-023

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**ThermoFisher**  
S C I E N T I F I C

## Anza™ T4 DNA Ligase Master Mix

Cat. No.	Size	Lot no.	Exp. Date
IVGN210-4	50 reactions		
IVGN210-8	200 reactions		

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**Product description** The Invitrogen™ Anza™ T4 DNA Ligase Master Mix is a 4X concentrate used to join DNA fragments with sticky or blunt ends, and repair nicks in double-stranded DNA with 3'-hydroxyl and 5'-phosphate ends.

Components	IVGN210-4	IVGN210-8
Anza™ T4 DNA Ligase Master Mix	1 × 250 µL	4 × 250 µL

**Storage** Store at -20°C.

**For research use only. Not for use in diagnostic procedures.**

## General guidelines

- Ligation can be performed with either phosphorylated or dephosphorylated DNA insert.  
**Note:** if using a dephosphorylated DNA insert, the linearized vector needs to be phosphorylated.
- Ligation can be performed with DNA in water, TE, elution buffer, or 1X Anza™ buffers.
- If using electrocompetent cells, perform column purification of ligated DNA prior to transformation.

## DNA ligation protocol

Use this protocol to perform 15-minute ligation of blunt- or sticky-ended DNA into a vector for cloning.

1. Prepare a reaction mix by adding the reagents listed in the following table to a clean microcentrifuge tube:

Reagent	Volume
Nuclease-free water	As required to reach final reaction volume
Linearized vector DNA	10–100 ng
DNA insert	3:1 molar excess over vector DNA
Anza™ T4 DNA Ligase Master Mix	5 µL
<b>Final reaction volume</b>	<b>20 µL</b>

2. Mix reagents by pipetting up and down, then centrifuge briefly.
3. Incubate at room temperature for 15 minutes.
4. Use 1–5 µL of the ligation reaction mixture to transform competent cells.

**Note:** the ligation reaction mixture can be stored at 0–4°C until required for transformation.