

## **Related products**

Product	Amount	Cat. no.
Anza™ T4 DNA Ligase Master Mix	50 reactions	IVGN210-4
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
PureLink <sup>™</sup> PCR Purification Kit	50 preps	K3100-01
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

To order additional Anza<sup>™</sup> Restriction Enzymes and Anza<sup>™</sup> Modifying Cloning Enzymes, go to **thermofisher.com/Anza** 

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## Anza™ Alkaline Phosphatase (1 U/µL)

Cat. No.	Size	Lot no.	Exp. Date
IVGN220-4	500 reactions		
IVGN220-8	2000 reactions		
Publication N	<b>lo.</b> MAN00143′	13 <b>Rev.</b> E	3.0

ProductThe Invitrogen™ Anza™ AlkalinedescriptionPhosphatase is used for<br/>dephosphorylating vectors prior to<br/>insert ligation and for removing 5'<br/>phosphates prior to end-labeling.

Components	IVGN210-4	IVGN210-8
Anza™ T4 Alkaline Phosphatase	500 μL	4 × 500 μL
Anza™ 10X Buffer	500 μL	2 × 1000 µL
Anza™ 10X Red Buffer	500 μL	2 × 1000 µL

Storage

Store at –20°C.

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

## General guidelines

- Plasmid DNA should be free of RNA and genomic DNA for efficient dephosphorylation.
- The enzyme can be heat inactivated at 80°C in 5 minutes, eliminating the need for purification prior to ligation.
- If the Anza<sup>™</sup> restriction enzyme is incompatible with the one step protocol, use a two step protocol by performing restriction digestion prior to dephosphorylation.

For detailed instructions on two step protocols, go to **thermofisher.com/Anza** 



## One step digestion and dephosphorylation protocol

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
plasmid DNA	0.2–1 µg
Anza™ 10X Buffer <b>or</b> Anza 10X Red Buffer	2 µL
Anza™ Restriction Enzyme	1 µL
Anza™ Alkaline Phosphatase	1 µL
Final reaction volume	20 µL

- 2. Mix reagents by pipetting up and down.
- 3. Incubate at 37°C for 15 minutes.
- 4. Heat inactivate enzyme by incubating at 80°C for 20 minutes.
- 5. Ligate insert and vector using the Anza<sup>™</sup> T4 DNA Ligase Master Mix.
- Use 1–5 μL of the ligation reaction mixture to transform competent cells.