Ligation protocol

Introduction

Get started by giving your protocol a name and editing this introduction.

Materials

- Ice
- > Digested fragments
- > T4 DNA Ligase buffer
- > T4 DNA ligase
- > Sterile H2O
- > Heatblock/PCR machine

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Procedure

Ligation

- 1. Keep all component on ice. Take the Ligase out of the freezer directly to ice only right when you need it and return it to the freezer right after use.
- 2. Use 2µl of each restriction reaction

this is your ligating DNA

- 3. Add 1µl 10x T4 DNA Ligase Buffer
- 4. Add 0,5µl T4 DNA Ligase
- 5. Adjust total volume to 10µl with H2O
- 6. Ligate in rt for 30min
- 7. Inactivate the ligase by incubating in $80 \,{}^{\circ}$ C for 20min
- 8. Use 1-2µl for transformations