

# Gibson-Cloning

---

## Introduction

Gibson cloning is an alternative to sticky-end cloning and may work better sometimes

## Materials

- Prepared Gibson-Cloning Mix (contains Polymerase, Ligase, 3'-5'-Exonuclease) (can be bought as Kit)
- Plasmid (cut)
- Insert with corresponding overlaps

## Procedure

1. Calculate the volumes of insert and vector needed. Use 50 ng of vector for a 20  $\mu$ L ligation. Use the 3 fold molar surplus of insert
2. Pipette the following table

Component	Volume in $\mu$ L
Gibson Cloning Mix	15
Vector	x
Insert	x
ddH <sub>2</sub> O	x
<b>total</b>	<b>20</b>

3. Incubate 1 hour at 50°C
4. Use the sample for transformation