



Fast Ligation Kit

Catalog Number FLK-01

Components	100 reactions
Fast DNA Ligase	100 ul
2X Reaction Buffer	1 ml

Storage

The kit is stable for 12 months at -20°C.

Application

Suitable for all common ligation reactions including cohesive and blunt end ligations.

Features

Fast - 10 minute protocol for both cohesive and blunt end ligations.

Protocol

1. Add de-ionized water or Tris-EDTA (TE) buffer to 50ng of vector and 15 ng insert (**3 fold excess of insert is recommended**), to a final volume to 9ul.
2. Add 10ul of 2X reaction buffer and mix by inverting tube several times.
3. Add 1ul of Fast DNA Ligase and mix thoroughly by inverting tube several times. Briefly centrifuge to bring the contents to the bottom of the tube.
4. Incubate the ligation mixture at room temperature for 10 minutes¹.
5. If transforming competent cells (**HIT DH5 alpha Catalog Number RH617**) store ligation reaction on ice until transformation. Alternatively the reaction can be stored at -20°C for extended periods of time.

Notes: **1.** Increasing the incubation time from 10 to 20 minutes has been shown to increase ligation efficiency for certain DNA fragments. **2.** Inactivation of ligase by heating is not recommended, as it may reduce transformation efficiency. **3.** Avoid repeated freeze/thaw cycles for maximum ligase efficiency..

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