

# pGGDestTol2LC-sgRNA ligation mixture

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## **Abstract**

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#### **Protocol**

## Step 1.

2μl 10x CutSmart buffer

## Step 2.

2 μl T4 DNA ligase buffer

## Step 3.

100ng of each pU6x-sgRNA plasmid

#### Step 4

50ng empty pGGDestTol2LC-sgRNA vector of choice (see plasmid list and diagrams in Guidelines),

#### Step 5.

1µl Bsal

#### Step 6.

1μl T4 DNA ligase

#### Step 7.

adjust volume to 20µl with H2O