



Fig. 2. Directional cloning strategy.

5. 3M Sodium acetate.
6. 100% (v/v) Ethanol.

2.4. Ligating the Adaptors

1. 70% (v/v) Ethanol.
2. Adaptors (4 μ g at 0.4 μ g/ μ L).
3. 5% Nondenaturing acrylamide gel.
4. 10X Ligation buffer: 500 mM Tris-HCl, pH 7.4, 100 mM MgCl₂, 10 mM dithiothreitol (DTT).
5. 10 mM rATP.
6. T4 DNA ligase (4 Weiss U).

2.5. Phosphorylating the Adaptors

1. 10X Ligation buffer (*see* Subheading 2.4., item 4).
2. 10 mM rATP.
3. T4 polynucleotide kinase (10 U).