ClonExpress® One Step Cloning Kit

ClonExpress® II One Step Cloning Kit	25/50 rxn	C112-01/02	One Step Cloning for Single Fragment
ClonExpress® MultiS One Step Cloning Kit	10/25 rxn	C113-01/02	One Step Cloning for Multi-fragment
ClonExpress® Ultra One Step Cloning Kit	25/50 rxn	C115-01/02	One Step Cloning for 1 ~ 5 Fragments

Cloning Can be as Simple as You Want

ClonExpress® Seamless Cloning Technology

ClonExpress technology is a simple, fast and efficient seamless DNA cloning technology, which can quickly and directionally clone the inserted fragments to any site of any vector. Linear the vector, and introduce the linearized clone vector terminal sequence at the 5' end of PCR primer of the inserted fragment, so that the 5' end and 3' end of PCR products of the inserted fragment have the same sequence (15 - 20 bp) corresponding to the two ends of the linearized clone vector, respectively. After mixing the PCR products with the linearized clone vector in a certain proportion, under the catalysis of Exnase, the reaction can be completed in 5 - 30 min and the positive rate of cloning is more than 95%.

- **Widely Applicable** — It is suitable for targeted cloning to almost any vector at any site.
  It can efficiently clone 50 bp - 10 kb fragments and carry out one step multi-fragment seamless cloning.
- **Design Simple** — Enzyme digestion is unnecessary.
  It only needs to introduce the homologous sequence at the end of the vector at the 5' end of the primer.
- **Convenient Operation** — The linearized clone vector and PCR products can be cloned directly without purification.
  It only takes 5 - 30 min to complete the reaction.
- **Highly Efficient Cloning** — Efficient cloning (100 cfu/ng Vector) can also be achieved by using inefficient competent cells (10^7 cfu/μg).
  Independent of ligase and phosphatase, the positive rate of cloning is more than 95%.

Vazyme BioTech Co.,Ltd.
Sales: global@vazyme.com
Support: global@vazyme.com

Progress of homologous recombination experiment

A Vector linearization: the linearized vector can be obtained by enzyme digestion or reverse PCR.

B Get insert fragments*:

① The 15 - 20 bp sequences at the end of the linearized vector was used as the homologous sequence (blue and red markers) and added to the 5' end of the gene-specific forward/reverse primers respectively, so as to amplify the insertion fragments with homologous sequences.

② The primers need to add homologous sequences at the 5' end (marked with dark blue, green, light blue and red in the figure), so that there are 15 - 20 bp homologous sequences between products and between products and vector.

C Recombination reaction: the linearized vector and the inserted fragments were mixed in proportion, and the recombination reaction was complete at 37˚C ~ 50˚C for 5 - 30 min under the catalysis of Exnase.

D Transformation of competent cells: The cloning products can be directly transformed to competent cells with a true positive rate >95%.

* ① represents single fragment cloning; ② represents multi-fragment cloning

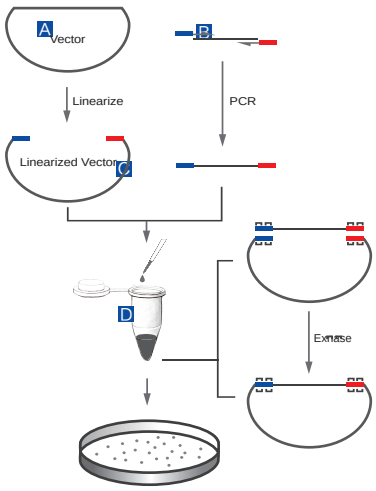

Fig. 1 Schematic diagram of rapid cloning technology

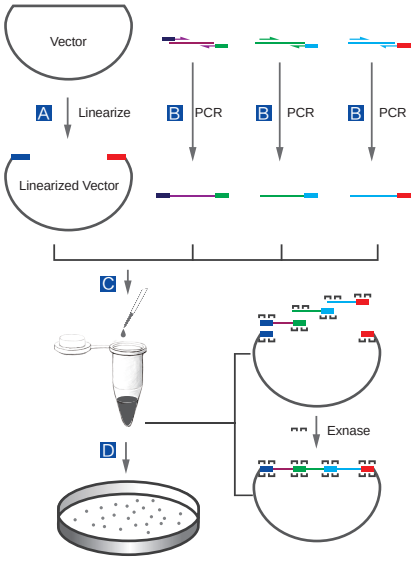

Fig. 2 Schematic diagram of multi-fragment splicing cloning technology

Cloning efficiency

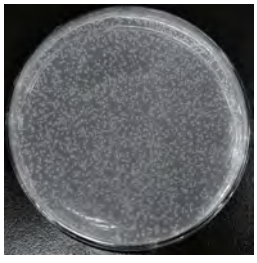

plate of recombination and transformation reaction

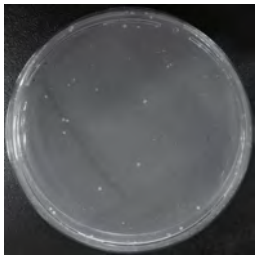

Plate of negative transformation control

Fig 3. The plate of overnight culture

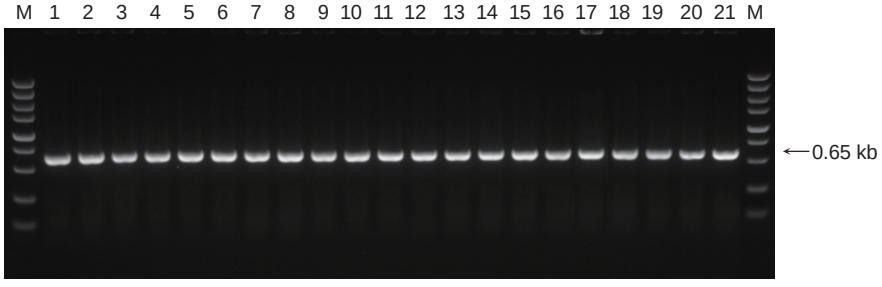

Fig 4. Agarose gel electrophoresis detection of colony PCR

M：DL5,000 Marker；1 - 21：21 positive clones

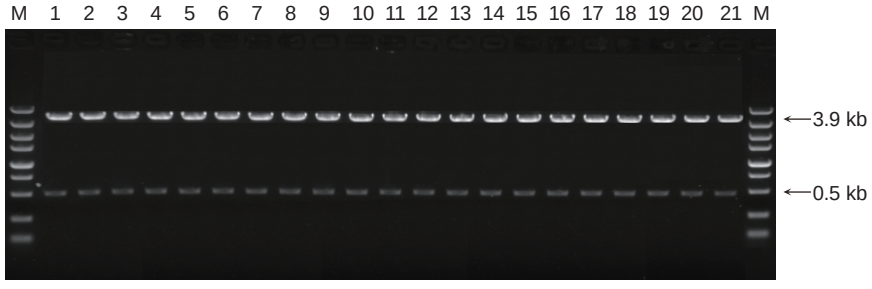

Fig 5. Agarose gel electrophoresis detection of enzyme-digested products

M：DL5,000 Marker；1 - 21：21 positive clones