

GenBuilder DNA Assembly Kits

Highest Efficiency Seamless Cloning Solution



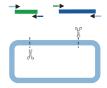




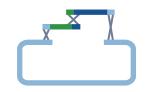
Features	GenBuilder™ Cloning Kit	GenBuil
Cat. No.	L00701	
Highlights	Multi-gene cloning for up to 6 fragments	Multi gene cl
Number of fragments	6	
Cloning efficiency %	>90%	
Plasmid library construction	Suitable	1
Incubation time	15 min	
Cloning unpurified PCR product	Efficient	
Assembly with single stranded oligos	No	
HT cloning	Yes	
Seamless cloning	Yes	
Cloning fragments >5 kb	Yes	
Positive control	Linearized pUC57 with two fragments to reconstitute RFP for visual determination of cloning efficiency	Linearized pl reconstitute F of
Size (No. of Reactions)	10, 50, customized	10

GenBuilder™ Plus Cloning Kit		
L00744		
Multi gene cloning for up to 12 fragments		
12		
>90%		
Recommended		
15 min		
Highly efficient		
Yes		
Linearized pUC57 with two fragments to reconstitute RFP for visual determination of cloning efficiency		
10. 50. customized		

Assembly Workflow



Prepare the DNA fragments with overlaps and the linearized vector for assembly.



2. Add DNA fragments and linearized vectors in the GenBuilder™ master mix. Mix well and incubate at 50°C for 15-50 minutes.

Final construct sequence

3. Transform assembly product into competent cells and plate on selective medium. Obtain colonies containing the assembled DNA.



Highest cloning efficiency

GenBuilder and GenBuilder Plus have the highest cloning efficiency among competitors

A. DNA Assembly with five PCR fragments

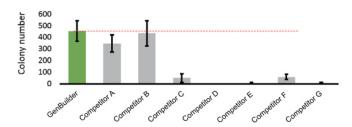
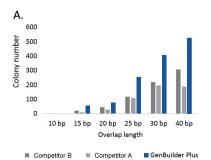
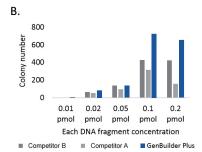


Figure 1. Cloning five or more PCR fragments with GenBuilder, GenBuilder Plus and other cloning kits

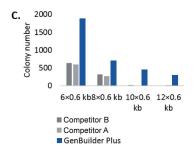
Performance Comparison of Assembly Kits



Graph A. Impact of the overlapping base pair length with cloning of 5 DNA inserts into one vector. Various PCR fragments with overlapping lengths of 10 bp, 15 bp, 25 bp, 30 bp, and 40 bp were assembled into pUCS7 in a single-step assembly reaction. The number of colonies recovered from 1/10th of the cloning reaction were counted per reaction.



Graph B. Impact of DNA fragment concentration with cloning of 5 DNA inserts into one vector. Cloning of 5 DNA inserts of various concentrations into one vector using various DNA Assembly methods. Various concentrations of five PCR fragments (0.01 pmol, 0.02 pmol, 0.05 pmol, 0.1 pmol, and 0.2 pmol) were assembled into pUCS7 in a single-step assembly reaction. The number of colonies recovered from 1/10th of the cloning reaction were counted per reaction.



Graph C. Performance of Assembly kits with the increase of the amount of DNA fragments. Multiple fragment cloning impact with various DNA Assembly methods. 6, 8, 10 and 12 PCR fragments were assembled into linearized pUCS7 vector in a single-step assembly reaction. The number of colonies recovered from \(\frac{1}{1}\) 10th of the cloning reaction were counted for each reaction. GenBuilder™ Plus DNA Assembly had the best assembly performance.

Figure 2. (graphs A, B and C). Comparison study of GenBuilder™ Plus DNA Assembly kit with Competitors A and B.

Reduced Reaction Incubation Time

Successful assembly in only 15 minutes

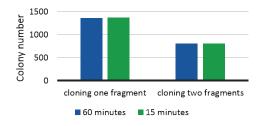


Figure 3. Cloning Efficiency Comparison: Incubation Time with GenBuilder™ Master-Mix

DNA assembly into the pUC57 vector was tested with a 1 kb PCR fragment and two 1.5 kb PCR fragments. GenBuilder™ reactions were incubated at 50°C for 15 or 60 minutes, then transformed into chemically competent DH10B cells. After recovery in 1 mL SOC medium, cells were spread on selective plates. Results showed no significant difference in colony numbers between 15-minute and 60-minute incubations. All colonies were >90% PCR positive.

