3 Assemblies

Assembly Name: 2frag

Assembly Size: 21,949 (Recomend: Electroporation)

3 Parts

MpMXBP_pFa pa aq

Reagents (ul)

| ligase_buffer | ligase | bsai | water |
|---------------|--------|------|-------|
| 2 | 0.5 | 0.5 | 1.506 |

Assembly Name: 3frag

Assembly Size: 29,494 (Recomend: Electroporation)

4 Parts

MpMXBP_pFa pa ab bq

Reagents (ul)

| ligase_buffer | ligase | bsai | water |
|---------------|--------|------|-------|
| 2 | 0.5 | 0.5 | 0 |

Assembly Name: 4frag

Assembly Size: 37,286 (Recomend: Electroporation)

5 Parts

MpMXBP_pFa pa ab bc

Reagents (ul)

| ligase_buffer | ligase | bsai | water |
|---------------|--------|------|-------|
| 2 | 0.5 | 1.0 | 0 |

9 Parts

| Part Name | Size | Conc. | Times Used | 30fmol (ul) | Total Volume |
|------------|-------|-------|------------|-------------|--------------|
| MpMXBP_pFa | 8067 | 40 | 3 | 3.737 | 13.453 |
| ра | 14112 | 40 | 3 | 6.538 | 420 |
| aq | 14083 | 50 | 1 | 5.219 | 6.263 |
| ab | 14087 | 33 | 2 | 7.91 | 18.984 |
| bq | 12884 | 40 | 1 | 5.969 | 7.163 |
| bc | 13888 | 60 | 1 | 4.289 | 5.147 |
| cq | 14131 | 25 | 1 | 10.474 | 12.569 |

Total Reagents Volumes Required (ul)

| ligase_buffer | ligase | bsai | water |
|---------------|--------|------|-------|
| 7.2 | 1.8 | 2.4 | 1.807 |

OT2 Set-Up Instructions

Reagent Plate Layout (ul)

| A1 ('ligase_buffer', 7.2) | B1 ('ligase', 1.8) | C1 ('bsai', 2.4) | D1 ('water', 1.807) |
|---------------------------|--------------------|------------------|---------------------|
| A2 ('aq', 6.263) | B2 ('ab', 18.984) | C2 ('bq', 7.163) | D2 ('bc', 5.147) |
| A3 | В3 | C3 | D3 |
| A4 | B4 | C4 | D4 |
| A5 | B5 | C5 | D5 |
| A6 | B6 | C6 | D6 |
| A7 | B7 | C7 | D7 |
| A8 | B8 | C8 | D8 |
| A9 | В9 | C9 | D9 |
| A10 | B10 | C10 | D10 |
| A11 | B11 | C11 | D11 |
| A12 | B12 | C12 | D12 |

| E1 ('MpMXBP_pFa', 13.453) | F1 ('pa.1', 200) | G1 ('pa.2', 200) | H1 ('pa.3', 20) |
|---------------------------|------------------|------------------|-----------------|
| E2 ('cq', 12.569) | F2 | G2 | H2 |
| E3 | F3 | G3 | Н3 |
| E4 | F4 | G4 | H4 |
| E5 | F5 | G5 | H5 |
| E6 | F6 | G6 | H6 |
| E7 | F7 | G7 | H7 |
| E8 | F8 | G8 | H8 |
| E9 | F9 | G9 | Н9 |
| E10 | F10 | G10 | H10 |
| E11 | F11 | G11 | H11 |
| E12 | F12 | G12 | H12 |

Thermocycler Plate with Assemblies

| A1 2frag | B1 3frag | C1 4frag | D1 |
|----------|----------|----------|-----|
| A2 | B2 | C2 | D2 |
| А3 | В3 | С3 | D3 |
| A4 | B4 | C4 | D4 |
| A5 | B5 | C5 | D5 |
| A6 | B6 | C6 | D6 |
| A7 | B7 | С7 | D7 |
| A8 | B8 | C8 | D8 |
| A9 | В9 | C9 | D9 |
| A10 | B10 | C10 | D10 |
| A11 | B11 | C11 | D11 |
| A12 | B12 | C12 | D12 |

| E1 | F1 | G1 | H1 |
|-----|-----|-----|-----|
| E2 | F2 | G2 | H2 |
| E3 | F3 | G3 | Н3 |
| E4 | F4 | G4 | H4 |
| E5 | F5 | G5 | H5 |
| E6 | F6 | G6 | H6 |
| E7 | F7 | G7 | H7 |
| E8 | F8 | G8 | Н8 |
| E9 | F9 | G9 | Н9 |
| E10 | F10 | G10 | H10 |
| E11 | F11 | G11 | H11 |
| E12 | F12 | G12 | H12 |