

Automated MetClo Assembly Plan

OT2 Set-Up Instructions

Welcome to the MetCloOT2 plan specific, to your assemblies!

This documentation is a userguide that instructs the setup of the OT2 robot for multiplexed MetClo based DNA assembly.

MATERIALS

> Software:

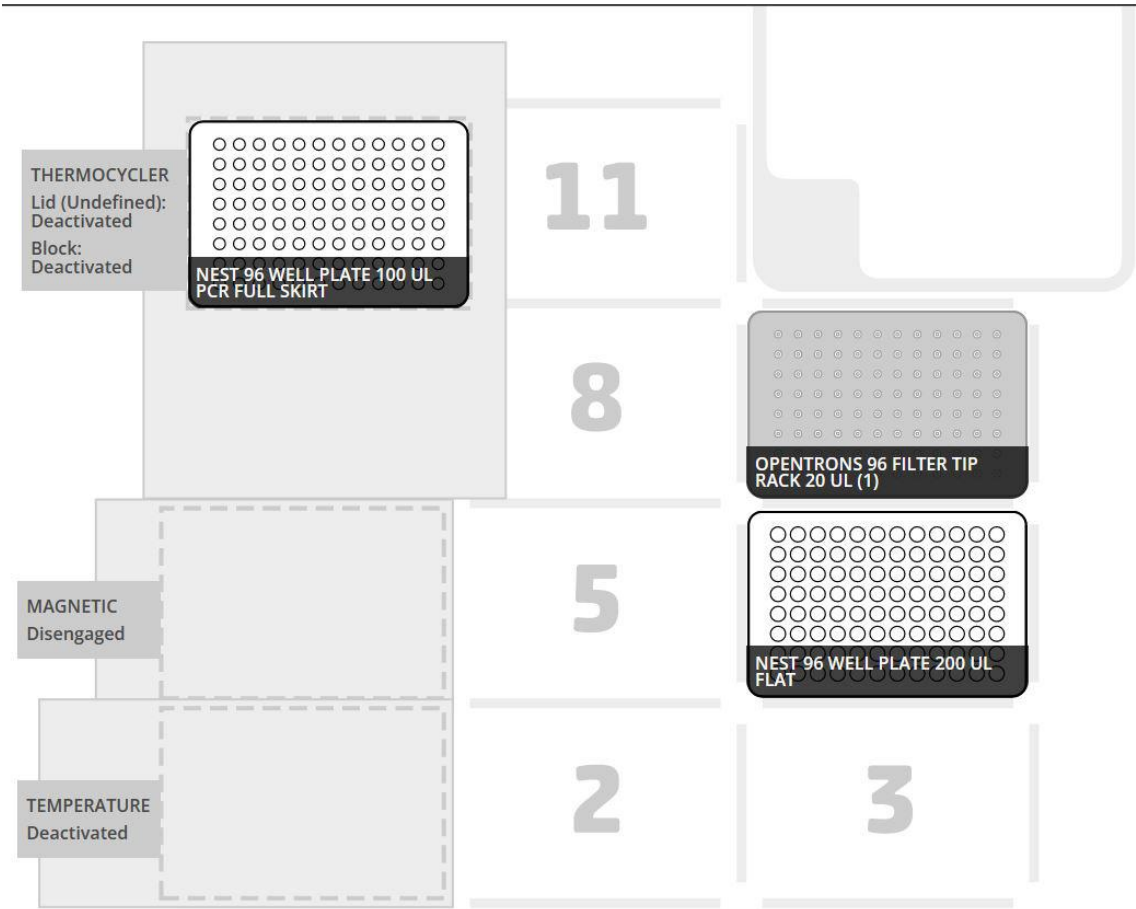
- OpenTrons OT-2 App
- Python 3

> Hardware:

- OpenTron OT-2
- OpenTrons P20 Single Channel Electronic Pipette
- Opentrons Thermocycler
- Opentrons 96 Tip Rack 20ul

Automated MetClo Assembly Plan

OT2 Layout



Automated MetClo Assembly Plan

Reagent Plate Layout (ul)

A1 ('ligase_buffer', 7.2)	B1 ('ligase', 1.8)	C1 ('bsai', 2.4)	D1 ('water', 1.807)
A2 ('bq', 7.163)	B2 ('bc', 5.147)	C2 ('cq', 12.569)	D2
A3	B3	C3	D3
A4	B4	C4	D4
A5	B5	C5	D5
A6	B6	C6	D6
A7	B7	C7	D7
A8	B8	C8	D8
A9	B9	C9	D9
A10	B10	C10	D10
A11	B11	C11	D11
A12	B12	C12	D12

E1 ('MpMXBP_pFa', 13.453)	F1 ('pa', 23.537)	G1 ('aq', 6.263)	H1 ('ab', 18.984)
E2	F2	G2	H2
E3	F3	G3	H3
E4	F4	G4	H4
E5	F5	G5	H5
E6	F6	G6	H6
E7	F7	G7	H7
E8	F8	G8	H8
E9	F9	G9	H9
E10	F10	G10	H10
E11	F11	G11	H11
E12	F12	G12	H12

Automated MetClo Assembly Plan

Thermocycler Plate with Assemblies

A1 2frag	B1 3frag	C1 4frag	D1
A2	B2	C2	D2
A3	B3	C3	D3
A4	B4	C4	D4
A5	B5	C5	D5
A6	B6	C6	D6
A7	B7	C7	D7
A8	B8	C8	D8
A9	B9	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	H3
E4	F4	G4	H4
E5	F5	G5	H5
E6	F6	G6	H6
E7	F7	G7	H7
E8	F8	G8	H8
E9	F9	G9	H9
E10	F10	G10	H10
E11	F11	G11	H11
E12	F12	G12	H12

Automated MetClo Assembly Plan

3 Assemblies

Assembly Name: 2frag

Assembly Size: 21,949 (Recomend: Electroporation)

3 Parts

MpMXBP_pFa	pa	aq
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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
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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
cq			

Reagents (ul)

ligase_buffer	ligase	bsai	water
2	0.5	1.0	0

Automated MetClo Assembly Plan

7 Parts

Part Name	Size	Conc.	Times Used	30fmol (ul)	Total Volume
MpMXBP_pFa	8067	40	3	3.737	13.453
pa	14112	40	3	6.538	23.537
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

Automated MetClo Assembly Plan

Total Reagents Volumes Required (ul) *1.2

ligase_buffer	ligase	bsai	water
7.2	1.8	2.4	1.807