# TECAN / ECHO Full Experiment (5,10,16,24,48,96+ Assembly Samples)

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#### **Introduction:**

To fully ensure that the TECAN / ECHO conversion is fully complete, I (along with Luis and Eric and Mary) made a total of 35 experiments using 12 plates, with up to 125 Assembly Samples and also up to 750 aspirate / dispense commands (ignoring reservoir, reagent, and wash commands). Here are the following results below:

#### a) Procedure:

- 1) Create a sample using a combination of 5 various parts:
  - Promoter
  - RBS (5' UTR)
  - Gene
  - Destination Vector (DVA / DVK)
  - Terminator
- 2) Use the following website: https://www.mocloassembly.com/ (Spice Rack Dashboard) to create the assemblies. If successful, the website will generate 3 main features:
  - A PDF file detailing the summary of the experiment: including reservoir well volume (in microliters), Master Mix Reagent Plate volume, and the number of plates used plus reactions and output assemblies (combinations of the parts)
  - A TECAN. gwl input file (Macbook reads the. gwl file as a textfile (.txt))
  - ZIP File (that one can "unzip" in the Finder Folder or in Terminal)
- 3) Terminal Command (as long as the TECAN.gwl file is put exactly in the directory of either tecan converter final.py or echo converter final.py):

Conversion from TECAN to ECHO (provided no errors exist in the file or in the conversion):

```
user$ python tecan_converter_final.py [Assembly_n_Tecan.gwl]
[Assembly n Tecan.csv]
```

Conversion from ECHO to TECAN (provided no errors exist in the file or in the conversion):

```
user$ python echo_converter_final.py [Assembly_n_Tecan.csv]
[Assembly Tecan.gwl]
```

4) If the conversion is still successful, then the respective CSV or TECAN GWL file will be created.

### b) Experiment:

\*\* Note: The experiment is so far only made for up to 750 commands, since another Setup Plate will be required to create a sample with more 96 wells. Generally, the samples are built en-sillico (in bulk) \*\*

Tier 1: 5/96 Wells Tested (only 1 assembly created)

Part Diagram of Plates 1-5:

Promoter	5' UTR / RBS	CDS	Terminator
Destination Vector			

### Plate 1)

Coordinate	Part	Volume	Order
A1	C0040_CD	2.0 μL	3
B1	DVK_AE	2.0 μL	Destination Vector
C1	B0015_DE	2.0 μL	4
D1	B0032m_BC	2.0 μL	2
E1	J23100_AB	2.0 μL	1
		$\Sigma = 10.0 \mu L$	

Statistics:

Reservoir: 14.0 µL

Master Mix  $6.0 \mu L$  (Reagent Plate 1) 4 plates used / 1 reaction reagent

### Coordinate Conversions:

### 5 commands total

TECAN			ЕСНО
Source	Destination	Source	Destination
2	1	B1	A1
5	1	E1	A1
4	1	D1	A1
1	1	A1	A1
3	1	C1	A1

Result: 5/5 commands passed!

# Plate 2)

Coordinate	Part	Volume	Order
A1	C0062_CD	2.0 μL	3
B1	DVK_AE	2.0 μL	Destination Vector
C1	B0015_DE	2.0 μL	4
<b>D1</b>	B0033m_BC	2.0 μL	2
<b>E1</b>	J23102_AB	2.0 μL	1
		$\Sigma = 10.0 \mu L$	

### Statistics:

Reservoir: 14.0 µL

Master Mix 6.0 µL (Reagent Plate 1) 4 plates used / 1 reaction reagent

Coordinate Conversions:

5 commands total

TECAN ECHO

Source	Destination	Source	Destination
2	1	B1	A1
5	1	E1	A1
4	1	D1	A1
1	1	A1	A1
3	1	C1	A1

# Plate 3)

Coordinate	Part	Volume	Order
A1	cre_CD	2.0 μL	3
<b>B</b> 1	DVK_AF	2.0 μL	Destination Vector
C1	B0015_DF	2.0 μL	4
D1	BCD2_BC	2.0 μL	2
<b>E1</b>	J23103_AB	2.0 μL	1
		$\Sigma = 10.0 \mu L$	

# Statistics:

Reservoir: 14.0 µL

Master Mix  $6.0 \mu L$  (Reagent Plate 1) 4 plates used / 1 reaction reagent

# Coordinate Conversions:

### 5 commands total

<u>TECAN</u> E			ЕСНО
Source	Destination	Source	Destination
2	1	B1	A1
5	1	E1	A1
4	1	D1	A1
1	1	A1	A1
3	1	C1	A1

# Plate 4)

Coordinate	Part	Volume	Order
A1	C0062_CD	2.0 μL	3
B1	DVK_AE	2.0 μL	Destination Vector
C1	B0015_DE	2.0 μL	4
D1	B0033m_BC	2.0 μL	2
<b>E1</b>	J23100_AB	2.0 μL	1
		$\Sigma = 10.0 \mu L$	

# Statistics:

Reservoir: 14.0 µL

Master Mix  $6.0 \,\mu L$  (Reagent Plate 1) 4 plates used / 1 reaction reagent

# Coordinate Conversions:

# 5 Commands Total

TECAN			ЕСНО
Source	Destination	Source	Destination
2	1	B1	A1
5	1	E1	A1
4	1	D1	A1
1	1	A1	A1
3	1	C1	A1

Plate 5) [FAILED due to excess amount of promoters [user input error]]

Coordinate	Part	Volume	Order
A1	N/A	0.0 μL	N/A
<b>B</b> 1	N/A	0.0 μL	N/A
<b>C1</b>	N/A	0.0 μL	N/A
D1	N/A	0.0 μL	N/A
<b>E1</b>	N/A	0.0 μL	N/A
<b>F</b> 1	N/A	0.0 μL	N/A
		$\Sigma = 0.0 \ \mu L$	

### Statistics:

Reservoir:  $0.0~\mu L$ 

Master Mix  $0.0~\mu L$  (Reagent Plate 1) 0 plates used / 0 reaction reagent

### Coordinate Conversions:

### 0 Commands Total

TECAN			ЕСНО
Source	Destination	Source	Destination

(Not Applicable since the MoClo Assembly Failed)

Tier 2: 10/96 Wells Tested (2+ assemblies created)

# Plate 6)

Coordinate	Part	Volume	Order
A1	C0062_CD	4.0 μL	3
B1	DVK_AE	4.0 μL	Destination Vector
<b>C</b> 1	B0015_DE	4.0 μL	4
D1	B0033m_BC	4.0 μL	2
<b>E1</b>	J23100_AB	2.0 μL	1a
F1	J23102_AB	2.0 μL	1b
		$\Sigma = 20.0 \mu L$	

### Statistics:

Reservoir:  $28.0~\mu L$  (water tank capacity) Master Mix  $12.0~\mu L$  (Reagent Plate 1) 4 plates used / 2 reactions reagent

### Coordinate Conversions:

10 Commands total

TECAN			ЕСНО
Source	Destination	Source	Destination
2	1	B1	A1
5	1	E1	A1
4	1	D1	A1
1	1	A1	A1
3	1	C1	A1
2	2	B1	B1
6	2	F1	B1
4	2	D1	B1
1	2	A1	B1
3	2	C1	B1

# Plate 7)

Coordinate	Part	Volume	Order
A1	B0034m_BC	4.0 μL	3
B1	C0080_CD	4.0 μL	Destination Vector
C1	DVK_AE	4.0 μL	4
D1	B0015_DE	4.0 μL	2
<b>E1</b>	J23103_AB	2.0 μL	1a
F1	J23106_AB	2.0 μL	1b
		$\Sigma = 20.0 \mu\text{L}$	

### Statistics:

Reservoir:  $28.0~\mu L$  (water tank capacity) Master Mix  $12.0~\mu L$  (Reagent Plate 1) 4 plates used / 2 reactions reagent

# Coordinate Conversions:

# 10 Commands Total

TECAN			ЕСНО
Source	Destination	Source	Destination
3	1	C1	A1
5	1	E1	A1
1	1	A1	A1
2	1	B1	A1

4	1	D1	A1
3	2	C1	B1
6	2	F1	B1
1	2	A1	B1
2	2	B1	B1
4	2	D1	B1

# Plate 8)

Coordinate	Part	Volume	Order
A1	C0040_CD	4.0 μL	3
B1	DVK_AF	4.0 μL	Destination Vector
C1	B0015_DF	$4.0~\mu L$	4
D1	B0033m_BC	4.0 μL	2
<b>E1</b>	J23100_AB	2.0 μL	1a
F1	J23107_AB	2.0 μL	1b
		$\Sigma = 20.0 \ \mu L$	

# Statistics:

Reservoir:  $28.0~\mu L$  (water tank capacity) Master Mix  $12.0~\mu L$  (Reagent Plate 1) 4 plates used / 2 reactions reagent

# Coordinate Conversions:

# 10 Commands Total

**TECAN** 

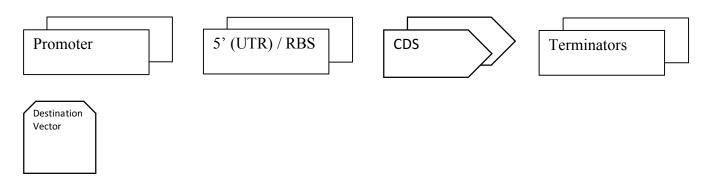
Source	Destination	Source	Destination
2	1	B1	A1
5	1	E1	A1
4	1	D1	A1
1	1	A1	A1
3	1	C1	A1
2	2	D1	D1

1	1	Al	Al
3	1	C1	A1
2	2	B1	B1
6	2	F1	B1
4	2	D1	B1
1	2	A1	B1
3	2	C1	B1

**ECHO** 

# Plate 9) (increased to 9 total parts)

# Part Diagram of Plate 9:



Coordinate	Part	Volume	Order
A1	C0040_CD	12.0 μL	3a
B1	C0062_CD	12.0 μL	3b
<b>C1</b>	DVK_AF	24.0 μL	Destination Vector
D1	B0015_DF	24.0 μL	4
<b>E1</b>	B0032m_BC	12.0 μL	2a
F1	B0033m_BC	12.0 μL	2b
G1	J23100_AB	8.0 μL	1a
H1	J23103_AB	8.0 μL	1b
A2	J23106_AB	8.0 μL	1c
		$\Sigma = 120.0 \mu\text{L}$	

### Statistics:

Reservoir:  $163.0 \mu L$  (water tank capacity) Master Mix  $72.0 \mu L$  (Reagent Plate 1)

4 plates used / 12 reactions

### Coordinate Conversions:

### 62 Commands Total

source plate, source well, destination plate name, destination well, Transfer Volume  $\,$ 

SetupPlate1,C1,Output1,A1,100 SetupPlate1,G1,Output1,A1,100 SetupPlate1,E1,Output1,A1,100

```
SetupPlate1, A1, Output1, A1, 100
SetupPlate1, D1, Output1, A1, 100
SetupPlate1, C1, Output1, B1, 100
SetupPlate1, G1, Output1, B1, 100
SetupPlate1, E1, Output1, B1, 100
SetupPlate1,B1,Output1,B1,100
SetupPlate1, D1, Output1, B1, 100
SetupPlate1, C1, Output1, C1, 100
SetupPlate1, G1, Output1, C1, 100
SetupPlate1,F1,Output1,C1,100
SetupPlate1, A1, Output1, C1, 100
SetupPlate1, D1, Output1, C1, 100
SetupPlate1, C1, Output1, D1, 100
SetupPlate1, G1, Output1, D1, 100
SetupPlate1, F1, Output1, D1, 100
SetupPlate1, B1, Output1, D1, 100
SetupPlate1, D1, Output1, D1, 100
SetupPlate1,C1,Output1,E1,100
SetupPlate1, H1, Output1, E1, 100
SetupPlate1, E1, Output1, E1, 100
SetupPlate1, A1, Output1, E1, 100
SetupPlate1, D1, Output1, E1, 100
SetupPlate1, C1, Output1, F1, 100
SetupPlate1, H1, Output1, F1, 100
SetupPlate1, E1, Output1, F1, 100
SetupPlate1, B1, Output1, F1, 100
SetupPlate1, D1, Output1, F1, 100
SetupPlate1, C1, Output1, G1, 100
SetupPlate1, H1, Output1, G1, 100
SetupPlate1, F1, Output1, G1, 100
SetupPlate1, A1, Output1, G1, 100
SetupPlate1, D1, Output1, G1, 100
SetupPlate1, C1, Output1, H1, 100
SetupPlate1, H1, Output1, H1, 100
SetupPlate1, F1, Output1, H1, 100
SetupPlate1, B1, Output1, H1, 100
SetupPlate1, D1, Output1, H1, 100
SetupPlate1, C1, Output1, A2, 100
SetupPlate1, A2, Output1, A2, 100
SetupPlate1, E1, Output1, A2, 100
SetupPlate1, A1, Output1, A2, 100
SetupPlate1, D1, Output1, A2, 100
SetupPlate1, C1, Output1, B2, 100
SetupPlate1, A2, Output1, B2, 100
SetupPlate1, E1, Output1, B2, 100
SetupPlate1, B1, Output1, B2, 100
SetupPlate1, D1, Output1, B2, 100
SetupPlate1, C1, Output1, C2, 100
SetupPlate1, A2, Output1, C2, 100
SetupPlate1,F1,Output1,C2,100
SetupPlate1, A1, Output1, C2, 100
SetupPlate1, D1, Output1, C2, 100
SetupPlate1, C1, Output1, D2, 100
SetupPlate1, A2, Output1, D2, 100
SetupPlate1, F1, Output1, D2, 100
SetupPlate1, B1, Output1, D2, 100
SetupPlate1, D1, Output1, D2, 100
```

Tier 3: 24/96 Wells Tested (up to 125 assemblies created)

Plate 10)

Part Diagram of Plate 10:

Promoter (3x)

5' (UTR) / RBS (3x)

CDS(3x)

Terminators (3x)

Destination Vector (2x)

Coordinate	Part	Volume	Order
A1	C0040_CD	18.0 μL	3a
B1	C0062_CD	18.0 μL	3b
<b>C</b> 1	DVK_AF	36.0 μL	Destination Vector
D1	B0015_DF	36.0 μL	4
<b>E1</b>	B0032m_BC	12.0 μL	2a
<b>F</b> 1	B0033m_BC	12.0 μL	2b
G1	BCD2_BC	12.0 μL	2c
H1	J23100_AB	12.0 μL	1a
A2	J23102_AB	12.0 μL	1b
B2	J23103_AB	12.0 μL	1c
		$\Sigma = 180.0 \mu\text{L}$	

### Statistics:

Reservoir: 244.0  $\mu$ L (water tank capacity) Master Mix 108.0  $\mu$ L (Reagent Plate 1)

4 plates used / 18 reactions

Coordinate Conversions: (final feasible conversion command to print out)

### 90 Commands Total

source plate, source well, destination plate name, destination well, Transfer Volume

SetupPlate1,C1,Output1,A1,100

SetupPlate1, H1, Output1, A1, 100

```
SetupPlate1,E1,Output1,A1,100
SetupPlate1, A1, Output1, A1, 100
SetupPlate1, D1, Output1, A1, 100
SetupPlate1, C1, Output1, B1, 100
SetupPlate1, H1, Output1, B1, 100
SetupPlate1, E1, Output1, B1, 100
SetupPlate1, B1, Output1, B1, 100
SetupPlate1, D1, Output1, B1, 100
SetupPlate1, C1, Output1, C1, 100
SetupPlate1, H1, Output1, C1, 100
SetupPlate1, F1, Output1, C1, 100
SetupPlate1, A1, Output1, C1, 100
SetupPlate1, D1, Output1, C1, 100
SetupPlate1, C1, Output1, D1, 100
SetupPlate1, H1, Output1, D1, 100
SetupPlate1,F1,Output1,D1,100
SetupPlate1, B1, Output1, D1, 100
SetupPlate1, D1, Output1, D1, 100
SetupPlate1, C1, Output1, E1, 100
SetupPlate1, H1, Output1, E1, 100
SetupPlate1, G1, Output1, E1, 100
SetupPlate1, A1, Output1, E1, 100
SetupPlate1, D1, Output1, E1, 100
SetupPlate1, C1, Output1, F1, 100
SetupPlate1, H1, Output1, F1, 100
SetupPlate1, G1, Output1, F1, 100
SetupPlate1, B1, Output1, F1, 100
SetupPlate1, D1, Output1, F1, 100
SetupPlate1, C1, Output1, G1, 100
SetupPlate1, A2, Output1, G1, 100
SetupPlate1, E1, Output1, G1, 100
SetupPlate1, A1, Output1, G1, 100
SetupPlate1, D1, Output1, G1, 100
SetupPlate1, C1, Output1, H1, 100
SetupPlate1, A2, Output1, H1, 100
SetupPlate1, E1, Output1, H1, 100
SetupPlate1, B1, Output1, H1, 100
SetupPlate1, D1, Output1, H1, 100
SetupPlate1, C1, Output1, A2, 100
SetupPlate1, A2, Output1, A2, 100
SetupPlate1, F1, Output1, A2, 100
SetupPlate1, A1, Output1, A2, 100
SetupPlate1, D1, Output1, A2, 100
SetupPlate1, C1, Output1, B2, 100
SetupPlate1, A2, Output1, B2, 100
SetupPlate1, F1, Output1, B2, 100
SetupPlate1, B1, Output1, B2, 100
SetupPlate1, D1, Output1, B2, 100
SetupPlate1, C1, Output1, C2, 100
SetupPlate1, A2, Output1, C2, 100
SetupPlate1, G1, Output1, C2, 100
SetupPlate1, A1, Output1, C2, 100
SetupPlate1, D1, Output1, C2, 100
SetupPlate1, C1, Output1, D2, 100
```

SetupPlate1, A2, Output1, D2, 100 SetupPlate1, G1, Output1, D2, 100 SetupPlate1, B1, Output1, D2, 100 SetupPlate1, D1, Output1, D2, 100 SetupPlate1, C1, Output1, E2, 100 SetupPlate1, B2, Output1, E2, 100 SetupPlate1, E1, Output1, E2, 100 SetupPlate1, A1, Output1, E2, 100 SetupPlate1, D1, Output1, E2, 100 SetupPlate1, C1, Output1, F2, 100 SetupPlate1, B2, Output1, F2, 100 SetupPlate1, E1, Output1, F2, 100 SetupPlate1, B1, Output1, F2, 100 SetupPlate1, D1, Output1, F2, 100 SetupPlate1, C1, Output1, G2, 100 SetupPlate1, B2, Output1, G2, 100 SetupPlate1,F1,Output1,G2,100 SetupPlate1, A1, Output1, G2, 100 SetupPlate1, D1, Output1, G2, 100 SetupPlate1, C1, Output1, H2, 100 SetupPlate1, B2, Output1, H2, 100 SetupPlate1, F1, Output1, H2, 100 SetupPlate1, B1, Output1, H2, 100 SetupPlate1, D1, Output1, H2, 100 SetupPlate1, C1, Output1, A3, 100 SetupPlate1, B2, Output1, A3, 100 SetupPlate1, G1, Output1, A3, 100 SetupPlate1, A1, Output1, A3, 100 SetupPlate1, D1, Output1, A3, 100 SetupPlate1, C1, Output1, B3, 100 SetupPlate1, B2, Output1, B3, 100 SetupPlate1, G1, Output1, B3, 100 SetupPlate1, B1, Output1, B3, 100 SetupPlate1, D1, Output1, B3, 100

### Plate 11)

Coordinate	Part	Volume	Order
A1	C0012m_CD	16.0 μL	3a
B1	C0062_CD	16.0 μL	3b
<b>C1</b>	DVK_AF	16.0 μL	3c
D1	B0015_DF	16.0 μL	3d
<b>E1</b>	B0032m_BC	64.0 μL	Destination Vector
F1	B0033m_BC	64.0 μL	4
G1	BCD2_BC	32.0 μL	2a
H1	J23100_AB	32.0 μL	2b
A2	J23102_AB	16.0 μL	1a
B2	J23103_AB	16.0 μL	1b

C2	16.0 μL	1c
<b>D2</b>	16.0 μL	1d
	$\Sigma = 320.0 \mu L$	

### Statistics:

Reservoir: 433.0  $\mu$ L (water tank capacity) Master Mix 192.0  $\mu$ L (Reagent Plate 1) 4 plates used / 32 reactions

# Coordinate Conversions:

160 Commands Total

# Plate 12)

Coordinate	Part	Volume	Order
A1	B0034m_BC	50.0 μL	4
B1	C0012m_CD	50.0 μL	5a
<b>C1</b>	C0040_CD	50.0 μL	5b
D1	C0062_CD	50.0 μL	5c
<b>E1</b>	C0080_CD	50.0 μL	5d
<b>F1</b>	cre_CD	50.0 μL	5e
G1	DVK_AF	200.0 μL	Destination Vector
H1	DVK_AF	50.0 μL	Destination Vector
A2	B0015_DF	200.0 μL	6a
B2	B0015_DF	50.0 μL	6b
C2	B0032m_BC	50.0 μL	2a
D2	B0033m_BC	50.0 μL	2b
E2	BCD2_BC	50.0 μL	3a
F2	BCD8_BC	50.0 μL	3b
G2	J23100_AB	50.0 μL	1a
H2	J23100_AB	50.0 μL	1b
A3	J23103_AB	50.0 μL	1c
B3	J23106_AB	50.0 μL	1d
C3	J23107_AB	50.0 μL	1e
		$\Sigma = 1250.0 \mu L$	

#### Statistics:

Reservoir: 1691.0 µL (water tank capacity)

Master Mix 1: 198.0  $\mu$ L (Reagent Plate 1) Master Mix 2: 198.0  $\mu$ L (Reagent Plate 1) Master Mix 3: 198.0  $\mu$ L (Reagent Plate 1) Master Mix 4: 198.0  $\mu$ L (Reagent Plate 1)

Total ( $\Sigma$ ): 750.0  $\mu$ L

4 plates used / 32 reactions

Coordinate Conversions:

626 Commands Total

### c) Conclusions:

### **Thresholds / Testing Benchmarks**

Using R Program / Prism Software (collaboration with Eric and Luis)

#### • Remarks:

- No failed tests (without human error) up to 48 wells in SetupPlate.
- 1 failed test (Plate 5, Test 3) due to improper user input (extra amount of promoter for a 5-well reaction)
- Passed 34/35 tests (97%, with margin of error less than 5%)

#### Overall:

- o 12 plates, 35 tests total
- Up to 750 aspirate / dispense commands converted successfully (in terms of source well, destination well, and transfer volume)
- o twice the number of JoVe Article 2017

### Mathematical Intuition / Pseudocode:

c1) Converting from TECAN Coordinates to ECHO (with floating arithmetic and modulo operators):

$$f(E) = \begin{cases} row = int(TECAN\ Number) - 1) / 8 + 1\\ column = int(TECAN\ Number) - 1) \% 8 \end{cases}$$

```
conversion dictionary = {["0", "A"],["1", "B"], ["2", "C"],
["3", "D"], ["4", "E"], ["5", "F"], ["6", "G"], ["7", "H"]}
# concatenate (or put together the strings)
```

s = convert to string(conv[int(column number])] + str(row)

### Example run:

```
tecan to echo(20) -> "D3"
```

c2) Converting from ECHO to TECAN (with floating arithmetic and mod operators):

$$f(T) = 8 * (ECHO \ column \ number - 1) + conv[ECHO \ row \ number]$$

# Example run: