Echo550 Liquid Handler

Standard Operating Protocol

I. Preparation

1. Use Echo qualified source plate (PP-0200, LP-0200).

	PP plate (#PP-0200)	**LDV plate (#LP-0200)**
Vol range	15-65 uL	3-12 uL

***Depends on the liquid property, see Appendix Table 1.*

2. Prepare the sample and load adequate volume (include dead volume) to the Echo-qualified source plate.
3. Centrifuge the plate at 4000rpm for 1 min to remove trapped air bubble.
4. Clean the bottom of the plate with Kimwipe.

II. Initialization

5. Log in PPMS tracker.
   Launch the software *Echo Plate Reformat v1.6*, select ***Connect***. Make sure the software *Echo Liquid Handler* is on. If not, please contact staff for assistance.
6. In Protocol setup, select appropriate options in (a) source plate, (b) destination plate and (c) mapping mode.

(a) Source Plate

Plate format: select ***384PP*** or ***384LDV***.

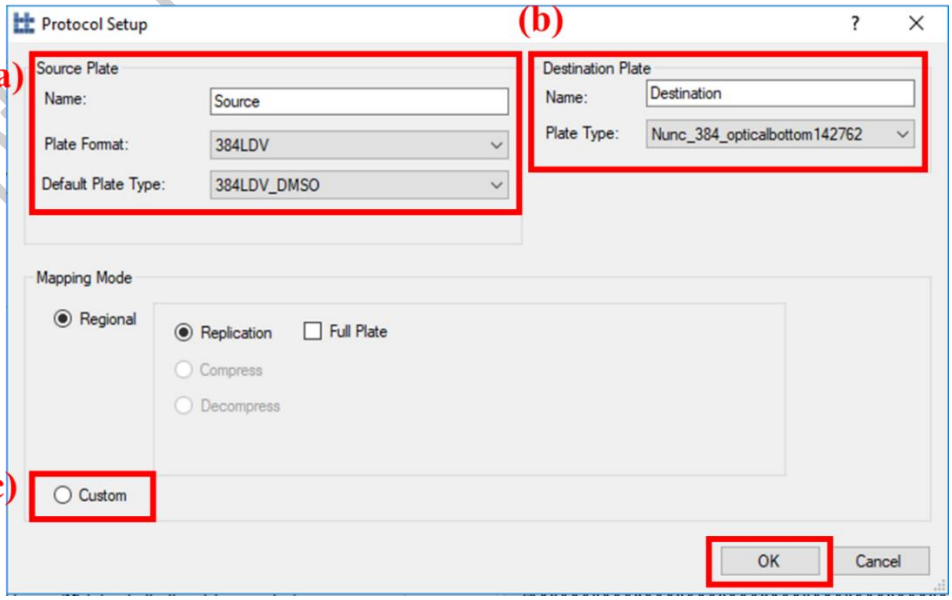
- Default Plate Type: select appropriate calibration protocol based on liquid properties. (Check ***Appendix, Table 1***)

(b) Destination Plate

- Plate type: select appropriate destination plate type (for SSIBio, Bio-rad and Roche plate, choose ***Roche 384 PCR plate***).

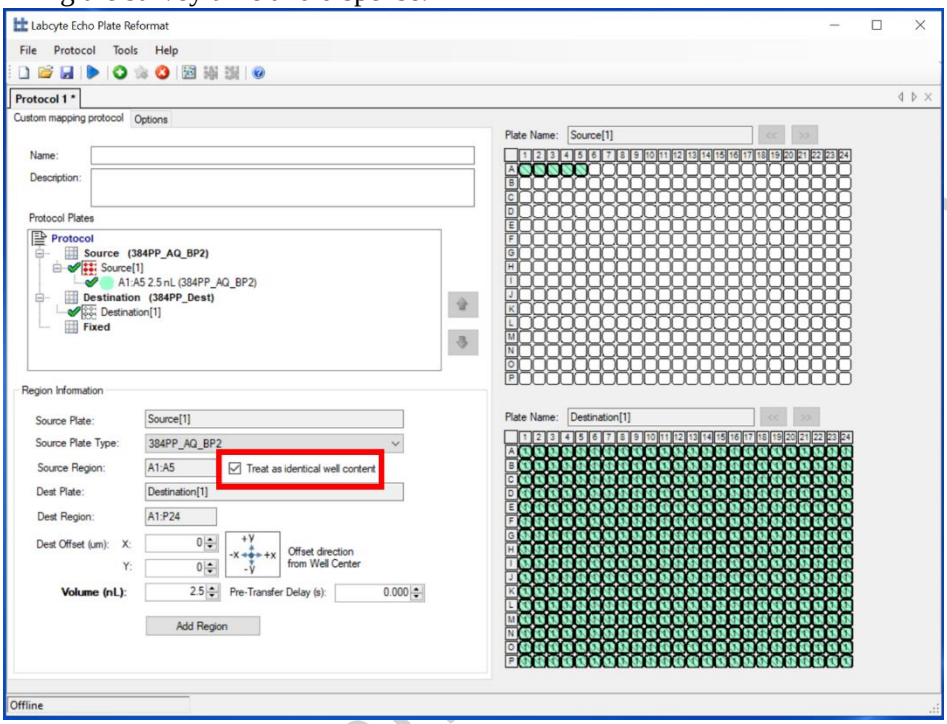
(c) Mapping mode

- select Custom.

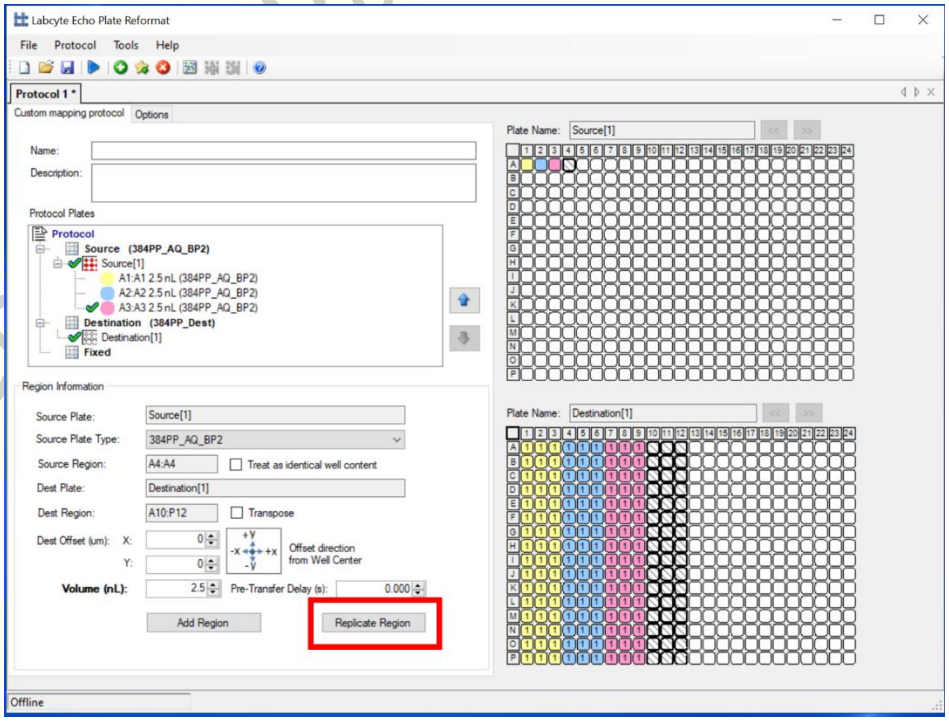


III. Transferring liquid

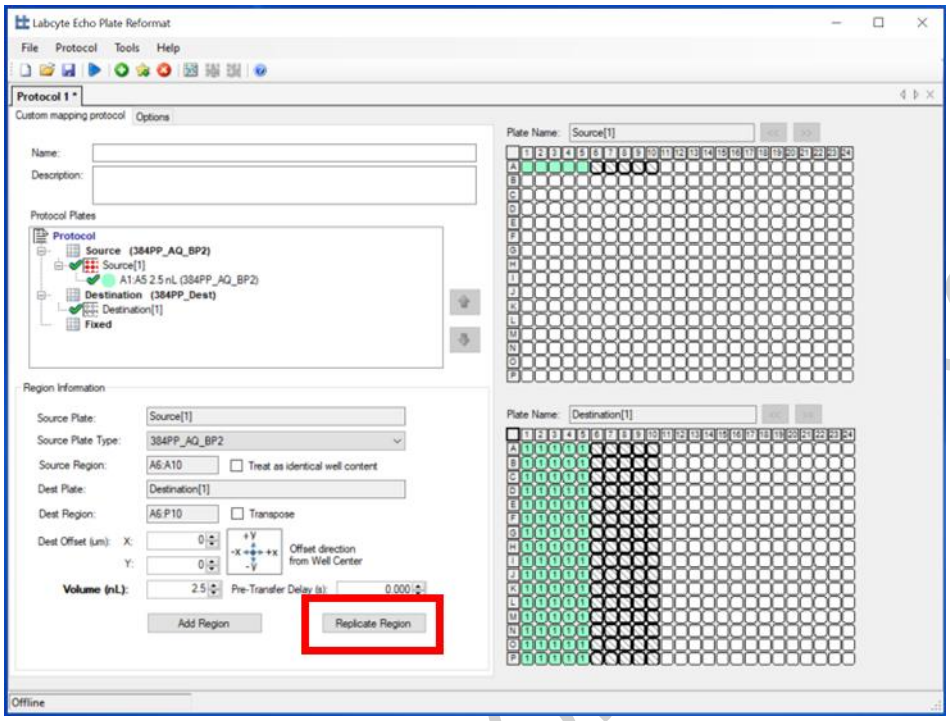
7. Create a new protocol.
8. Select the well(s) in the sources plate which contains the fluid to be transfer and select the well(s) in destination plate to receive the transfer volume.
    a. If multiple wells have same content (e.g. mastermix), check the box ***Treat as identical well content*** for optimizing the survey time and dispense.



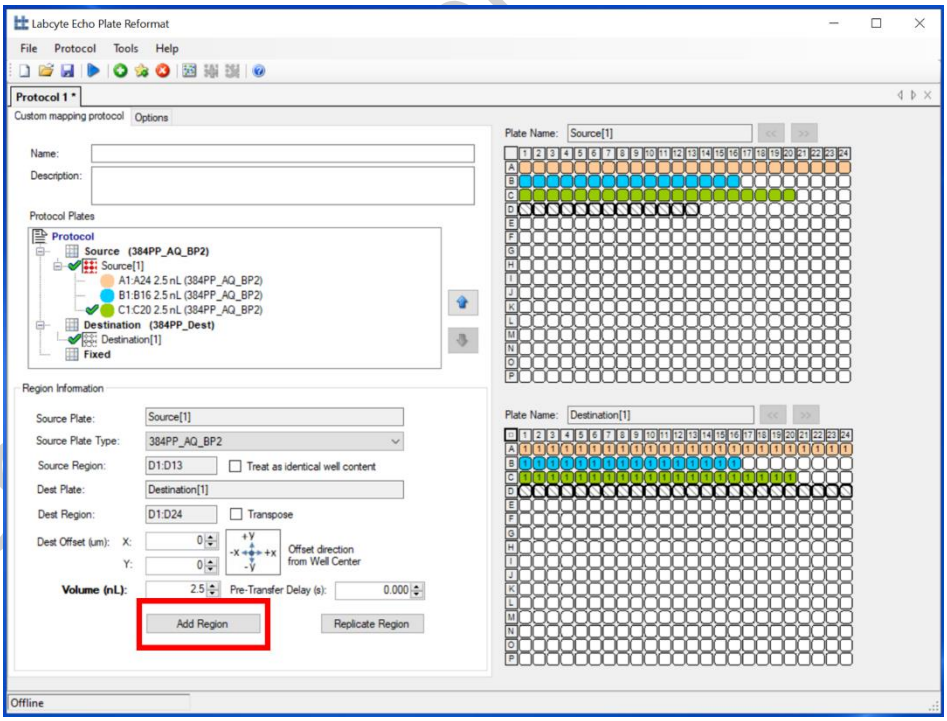
    b. If a sample from one well will be dispensed to multiple wells (e.g., cDNA), select ***Replicate Region***.



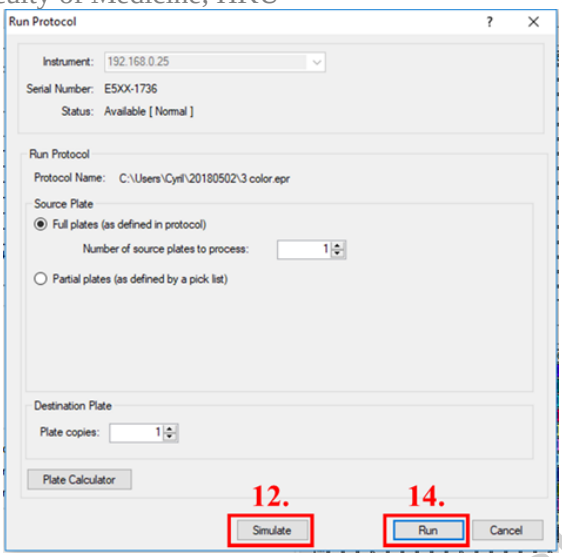
c. If multiple dispenses of samples from A1 to A5 for 16 times (i.e. across the whole column), each at A1 to P1 and A5 to P5 respectively, select ***Replicate Region***.



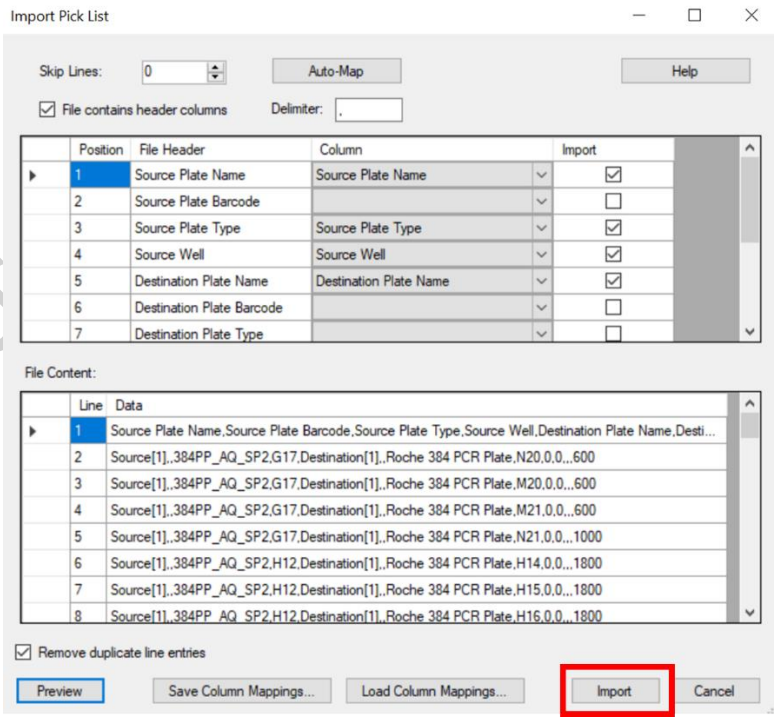
d. If samples will be replicated to the destination plate, select ***Add Region***.



9. Input the volume to be transferred.
   Save the Protocol in C:\Users\PI's Name\User Name. Click on ***Start*** button.
10. Use ***Simulate*** to verify the protocol was set up properly. Adjust the delays in Animation if necessary. Skip the animation if needed. Check whether the volume and event counts in source and destination wells are as expected.



11. Close the Simulation tap. Amend the protocol for any issues being identified and repeat the simulation again if needed.
12. Click Start in Echo software and click ***Run***.
13. Change the adapter if required by the alert message.
14. Put source plate on the reservoir plate gripper. Click ***OK***
15. Put the destination plate in the receiver plate gripper. Click ***OK*** to start the transfer.
    When the transfer finishes, remove the plates from Echo550 and put a cover on it or put it on ice if needed. Click ***OK*** after removal of each plate.
16. Go to "Labcyte Transfer Report" folder and check the transfer record Make up for any transfer exception by importing a pick list (by editing the exception report) to transfer the makeup volume. To import excel file for transferring, click a new dispense file and click ***File> Import Region Defination***. Choose the amended excel file. Click ***Import***.



17. Apart from the software "*Liquid Handler*", close all other software and log out PPMS tracker.
18. Sign on the logbook.

IV. Appendix

Table 1. Calibration Protocol based on liquid properties.

LDV plate

Calibration	Summary	Example fluids	Vol (uL)
384LDV_DMSO	384plate, DMSO	Solution reported in %DMSO hydrate (70%-100%)	2.5 - 12
1536LDV_DMSO	1536 plate, DMSO		1 – 5.5
384LDV_AQ_B	384 plate, buffer	PBS, cell culture media w/o protein, cDNA, primers/probes	3 – 12
384LDV_AQ_P	384 plate, protein	Buffers, reagents containing protein (no surfactants), e.g. Enzymes, serum, antibodies	6 – 14

PP plate

Calibration	Summary	Example fluids	Vol (uL)
384PP_DMSO2	DMSO	Solution reported in %DMSO hydrate (70%-100%)	15 – 65
384PP_AQ_SP2	surfactant/ protein	Buffers, reagents with surfactant, e.g. PCR mastermix, lysis buffer, Triton X-100, Tween-20, SDS, NP-40, with or w/o plasma or serum	15 – 65
384PP_AQ_BP2	buffer / protein	Reagents w/o surfatants. Can contain DNA/protein/serum/plasma. Solutions reported in % Glycerol (<30%)	15 – 65
384PP_AQ_CP	crystallography reagents, salts, etc.	Reagents w/o surfatants. MPD/PEGS/Osmotic solutions. Solutions reported in Mrayl.	25 – 50
384PP_AQ_GP	glycerol + buffer/protein	Enzymes/Antibodies/Proteins stored in glycerol. Solutions reported in % Glycerol (15%-55%)	15 – 65