



HP D300 Digital Dispenser

Operating Manual

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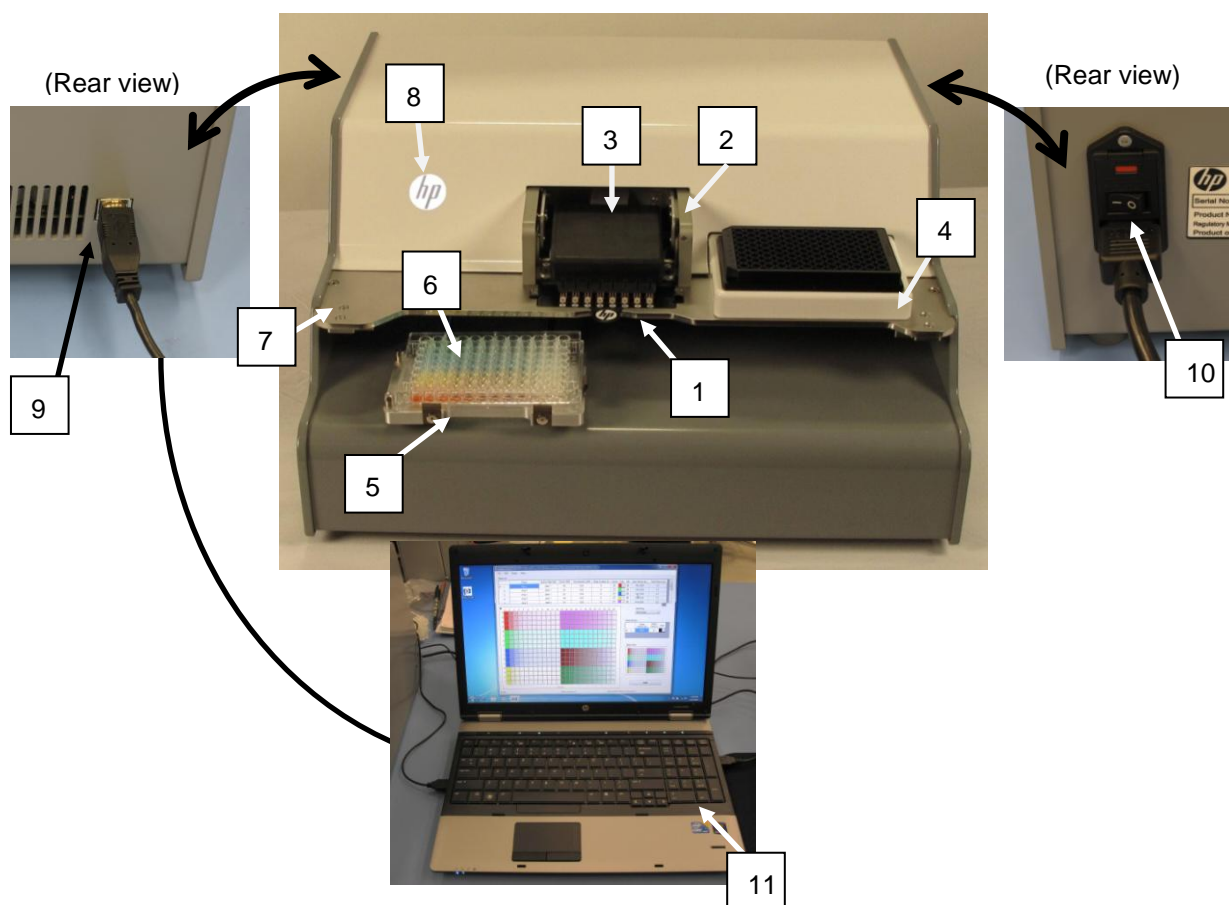
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1 Product description

The HP D300 Digital Dispenser dispenses precise amounts of dilute small molecule compounds in DMSO into standard 96- and 384-well plates. The dispenser is intended to be used in the lab environment for drug discovery purposes with small molecules in DMSO.



- 1 Dispensehead Cassette
- 2 Pogo mechanism
- 3 Pogo block
- 4 Source plate holder
- 5 Motorized stage and destination plate holder
- 6 Destination plate
- 7 Deck
- 8 Power indicator light (HP logo)
- 9 Dispenser USB connection
- 10 Power switch
- 11 PC with D300 software

1-1 System requirements

1-1-1 Hardware

- Dimensions of Dispenser (W x D x H): 47cm x 44cm x 24cm
- Grounded electrical outlet (2 required, one for dispenser, 1 for PC):
 - 120 V (US) or 230 V (Europe) AC power
- 1 GHz or faster 32 bit (x86) or 64 bit processor
- 1 GB (32-bit) or 2 GB (64 bit) RAM
- 16 GB (32-bit) or 20 GB (64-bit) available disk space
- Display: 1024 x 768, 256 colors minimum
- USB ports

1-1-2 Software

- Microsoft Windows 7, Vista or XP
- Microsoft .NET Framework 3.5
 - .NET 3.5 is included with Windows 7
 - .NET 3.5 is available for Windows XP and Vista (www.microsoft.com)
- Microsoft Excel 2003 or later
- Adobe Acrobat Reader (www.adobe.com/downloads)
- Do not adjust text size in Windows – use 100% only

1-1-3 Environmental operating conditions

- Temperature: 20C - 30C laboratory conditions (68 - 86F)
- Humidity: 30% - 80% RH

1-1-4 Shipping and storage conditions

- Temperature: 10C - 32C (50F – 90F)
- Humidity: 20% - 80% RH


2 Safety

The HP D300 Digital Dispenser is intended to be used in the lab environment for drug discovery purposes with small molecules in DMSO only. Other uses of the dispenser are not recommended at this time because they may introduce safety or performance issues.

Under no circumstances should the dispenser be used to dispense bacteria, viruses, cells, bio-fluids, biologicals, pathogens, contagions, infectious agents, potent toxins, oncogenes, radioactive materials or other potentially hazardous substances. The dispenser should not be exposed to Bio Safety Level 3 (BSL3) or greater conditions, contaminants or locations.

The dispenser should be used in accordance with all applicable safety rules that are in effect in the lab in which it is installed. Usage of the dispenser should also follow common sense safety guidelines for chemical, mechanical, and electrical safety.

NOTE: If this unit requires servicing, or you are unsure about any aspect of operation or safety, contact your Tecan Service Help Desk. The phone number can be found at <http://www.tecan.com/customersupport>.

 **CAUTION:** Do not remove any of the dispenser covers. Contact your Tecan Service Help Desk if the dispenser is not working properly. Do not attempt to repair the dispenser yourself.

2-1 General safety rules

- Place the dispenser on a work surface that supports all four feet securely.
- Plug the dispenser into a standard, grounded, electrical outlet. Note the power requirement (120V in North America or 230V in Europe) listed on the back label of the dispenser.
- Avoid the moving parts of the dispenser during operation.

2-2 Personal protective equipment

The HP D300 Digital Dispenser dispenses chemicals normally used by your company. The same safety precautions and personal protective equipment (PPE) specified by your employer for fluid transfer operations, such as pipetting into bio-assay well plates, should be used when operating the dispenser and handling these materials.

2-3 Possible inhalation hazard

The dispenser was designed with chemical safety in mind; however, it does dispense picoliter droplets of fluid and generates a small amount, less than 1% by volume, of very small aerosol droplets that might not be trapped within the well plate that you are dispensing into. If the fluids are known or suspected to be an inhalation hazard or contact hazard, consider using the dispenser in an exhausted enclosure such as a lab fume hood.

2-4 Pinch hazard

The dispenser was designed with mechanical safety in mind; however, there are potential pinch points between the pogo block and either the Dispensehead Cassette, the Dispenser deck or the top cover. This pinch hazard is labeled on the pogo block.

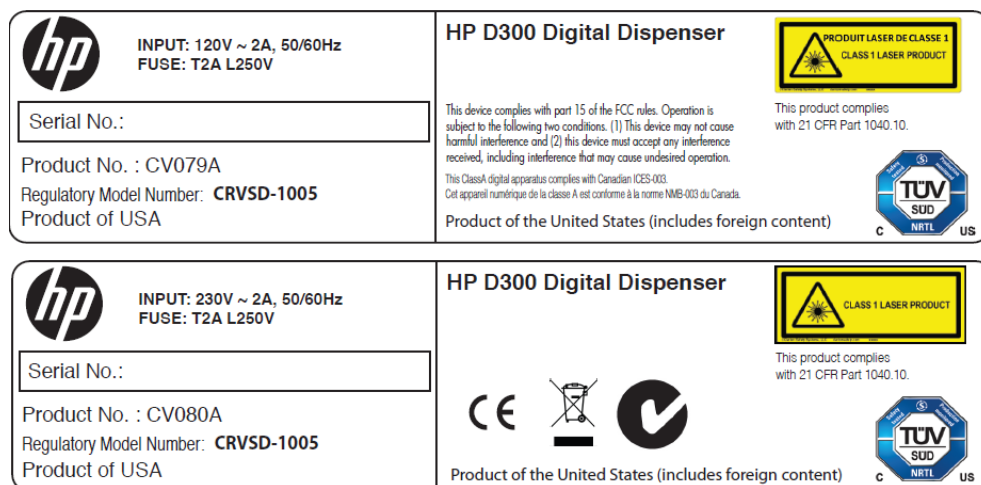
 **CAUTION:** Do not place fingers under or on top of the pogo block at any time.

2-5 Laser safety

A Class 1 laser is used in the dispenser to determine the height of the destination plate. This Class 1 Laser follows the requirements of Laser Notice No. 50 of the FDA (CDRH), Accession No. 0520365-02. This product complies with 21 CFR Part 1040.10 for laser products. See [Figure 2-1](#).

WARNING: Do not look at the laser beam directly. Similarly, do not continue to look at laser light reflected on a highly reflective substance for a long duration.

Figure 2-1 D300 Digital Dispenser rear placards indicating safety certifications for North America (top) and Europe (bottom)



2-6 Electrical safety

The HP D300 Digital Dispenser has passed a TUV certification for a CUE marking signifying compliance with applicable electrical regulations in the EU and North America. As shown in [Figure 2-1](#), a placard is attached to the rear of the dispenser, noting this certification and the electrical specifications of the dispenser: voltage 120V or 230V AC, 1 phase/3 wire, 50/60 Hertz, 2A full load amperage, and short circuit interrupt rating 5000A. The dispenser includes two user-replaceable T2A L250V (time lag, low breaking capacity 2A, 250V) fuses contained in the power inlet module at the rear of the dispenser.

2-7 Electrical emissions safety

The HP D300 Digital Dispenser has passed an FCC/EMC test for electrical emissions safety. This device complies with part 15 of the FCC Rules. Operation is subject to the following two conditions: (1) This device may not cause harmful interference, and (2) this device must accept any interference received, including interference that may cause undesired operation. Any changes or modifications to this equipment not expressly approved by the Hewlett-Packard Company may cause harmful interference, and void your warranty.

This equipment has been tested and found to comply with the limits for a Class A digital device, pursuant to part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this equipment in a residential area is likely to cause harmful interference in which case the user will be required to correct the interference at their own expense. Also, this Class A digital apparatus complies with Canadian ICES -003.

2-8 Decontamination prior to servicing

You may be required by Tecan service personnel to certify that appropriate cleaning, decontamination, and other abatement of potential threats to Tecan service personnel have been performed prior to shipping the dispenser for servicing. Recommended cleaning and decontamination procedures are described in the “[Clean the dispenser](#)” section of this manual.

2-9 Safe disposal of Dispensehead Cassettes and pipette tips

Disposal of used pipette tips and HP Dispensehead Cassettes (DHCs) should be performed in accordance with your company's chemical waste procedures. The DHCs do not require any special disposal procedures except for those required for the chemicals which have been placed in them by your laboratory.

2-10 Ergonomic safety

The HP D300 Digital Dispenser typically requires a single manual pipetting operation for each dispensed compound. In most situations, using the HP dispenser in the intended manner decreases the frequency of shoulder, arm, and wrist motions and so decreases the stress on these joints.

As with any work task, ergonomics are heavily influenced by workstation setup. To minimize ergonomic risks associated with the use of the dispenser, consider the following work area design ideas:

- Place the dispenser on a work bench at a height that allows for proper visual and physical access for operators. Having the dispenser too high can lead to greater shoulder elevation when pipetting into the Dispensehead and having the dispenser too low can lead to increased neck and back flexion.
- If an operator is too short to comfortably access the dispenser, consider moving the dispenser to a lower bench height. Alternatively, use a footstool or high lab stool that adjusts to the proper work height.
- Place the dispenser close to the front of the work surface to minimize the distance the operator must reach to pipette into the Dispensehead.
- Place the pipette tip disposal receptacle close to the dispenser.
- Place the computer keyboard close to the dispenser to minimize reach distance.
- Adjust the computer monitor height and screen angle for good physical and visual access during dispenser operation.
- Use a separate mouse rather than the laptop's touchpad whenever possible.
- Perform data analysis at a separate ergonomically designed workstation whenever possible rather than at the dispenser computer.

2-11 Additional assistance

Contact your local Tecan Service Help Desk for additional assistance. The phone number can be found at <http://www.tecan.com/customersupport>.

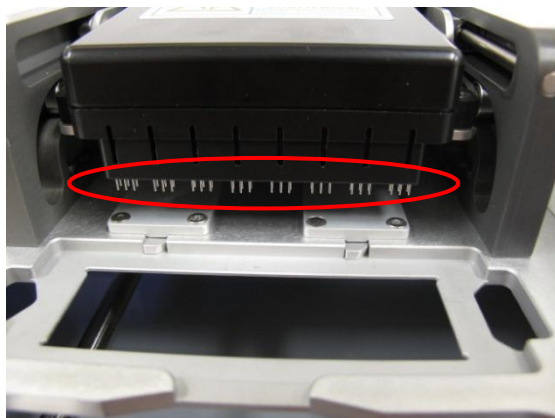
3 Clean the dispenser



WARNING: Observe the following cautions when cleaning the dispenser:

- Do not spray cleaning fluid directly onto the Dispenser when a Dispensehead Cassette is installed. Always avoid exposing unused Dispenseheads to cleaning fluids.
- Do not spray or wipe the ionizer bar that is mounted under the deck between the plate opening and the Dispenseheads.
- Do not spray or wipe the sensors under the Dispenser deck. Doing so could cause the sensors to become misaligned.
- Do not wipe the pogo pins. See [Figure 3-2](#).

Figure 3-2 Pogo pin region of pogo block which should not be sprayed or wiped with cleaning fluid



As with any lab instrument, it is a best practice to routinely clean areas of the Dispenser that come into contact with chemicals.

1. Turn off the dispenser as instructed in the “[Turn off the HP D300 Digital Dispenser](#)” section of this document.
2. Remove any unused Dispensehead Cassettes from the Dispenser.



CAUTION: Exposing unused Dispensehead Cassettes to cleaning fluids may compromise their performance.

3. Spray a cleaning solution, such as a bleach or a 70% ethanol solution, onto a fabric Texwipe or other cloth that does not generate particles.
4. Wipe down all accessible Dispenser surfaces, including the following (See the “[Product description](#)” section for the location of described areas):
 - The top and bottom surfaces of the dispenser deck, avoiding the ionizer bar in the cutout on the right side of the bottom of the deck and the sensors at the front corners of the deck
 - The pogo mechanism housing, excluding the pogo pin area as described above
 - The destination plate holder
 - The source plate holder
 - All of the outside surfaces of the dispenser

4 Get started

The HP D300 Digital Dispenser runs on standard AC power, either 120V or 230V, depending on the Dispenser version. HP Product Number CV079A runs on 120V and CV080A runs on 230V. The Product Number and the voltage requirement is shown on the label on the rear of the Dispenser.



WARNING: Be sure the dispenser matches your outlet voltage. The voltage requirement is shown on the label on the rear of the dispenser. Do NOT plug the dispenser in to the incorrect voltage. If the labeled voltage on your dispenser does not match your standard outlet, contact your Tecan Service Help Desk. The phone number can be found at <http://www.tecan.com/customersupport>.

Two electrical outlets are required, one for the dispenser and another for the computer. A dedicated circuit is not required as the dispenser draws less than two amps.

The dispenser requires a level bench space which is 50 cm (19 inches) wide, 46 cm (18 inches) deep, and 26 cm (10 inches) high. This does not include space for the required computer.

4-1 Start up the dispenser

1. Unpackage the dispenser and remove the four pieces of the shipping bracket.
2. Connect the separately packaged power cord to the rear of the dispenser. See **Figure 4-3**.
3. Plug the other end of the power cord into the appropriate power outlet. See **"Get started."**
4. Turn the dispenser on using the main power switch adjacent to the power cord connection on the rear of the Dispenser. Press the "1" on the switch to turn the dispenser on. See **Figure 4-3**.
5. The HP logo on the front of the dispenser will illuminate and the pogo block LEDs will flash, indicating the beginning of the Dispenser initialization routine.
6. Use the USB cable that came with the dispenser to connect the computer to the dispenser. The USB port is on the rear of the dispenser. See **Figure 4-4**.

Figure 4-3 On/off switch on rear of dispenser. Depress "1" to turn on.



Figure 4-4 USB port on rear of dispenser



7. The first time the Dispenser is connected to the computer, Device Manager will ask you for a device driver.
8. Insert the CD that was enclosed with the Dispenser into your computer's disk drive.
9. If your computer does not autorun the installer, use Windows Explorer to locate the setup.exe file on the CD disk drive.
10. Run setup.exe and follow the on-screen instructions to install **both** the device driver **and** the HP D300 Digital Dispenser software.

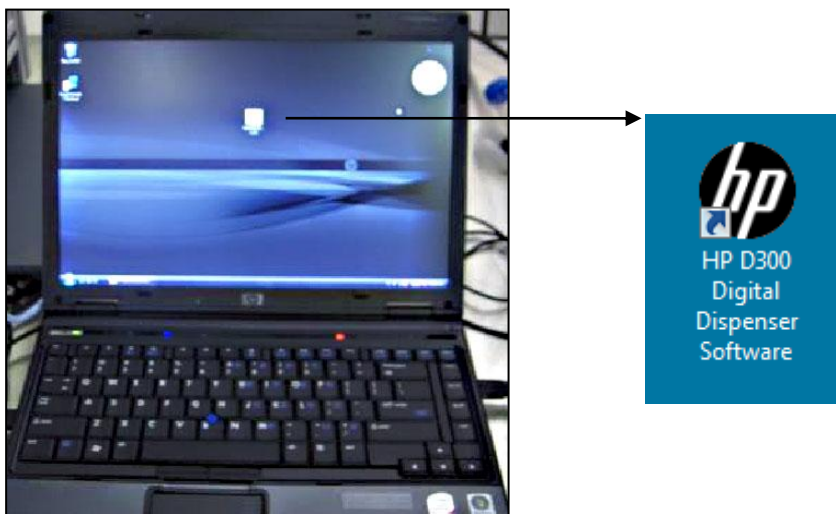
11. Check to see if an update is available for your software. Look for the latest version available and run the *.exe file for it on the Tecan FTP site: <ftp.tecan.com>

Username: HP-D300

Password: HP+Tecan

12. Click on the HP D300 Digital Dispenser Software program icon on the computer desktop to start up the HP D300 Digital Dispenser software. See [Figure 4-5](#).
13. Wait for the Dispenser to go through the software startup routine. The stage will move to the home position every time the HP D300 Digital Dispenser software is started.

Figure 4-5 Laptop computer showing the HP D300 Digital Dispenser software logo

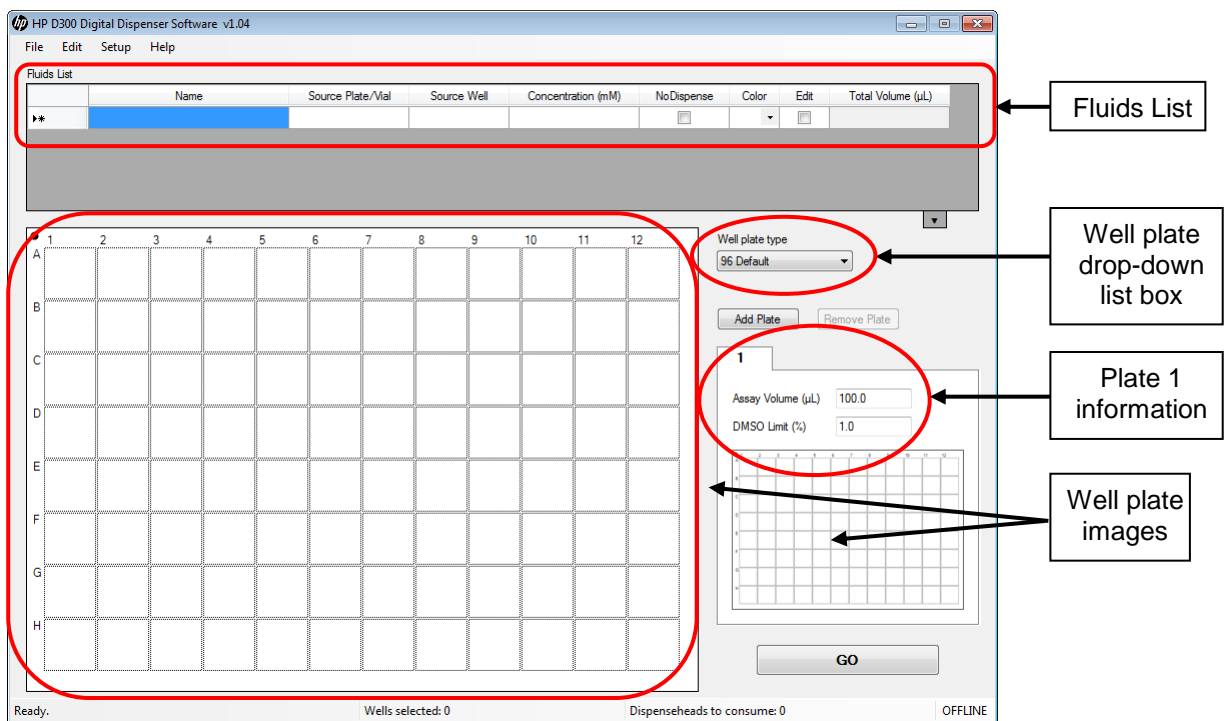


4-2 Get familiar with the software

Every time the software starts, if the computer is connected to a Dispenser, the computer checks the Dispenser USB communication, verifies that the dispenser components are operational, and moves the stage and the pogo mechanism to their home positions. If the computer is not connected to the Dispenser, the software is fully functional but the **GO** operation works in a demonstration mode.

Once the startup routine is complete, the HP D300 Digital Dispenser main operating window appears, showing an empty Fluids List and an image of an empty well plate. See [Figure 4-6](#).

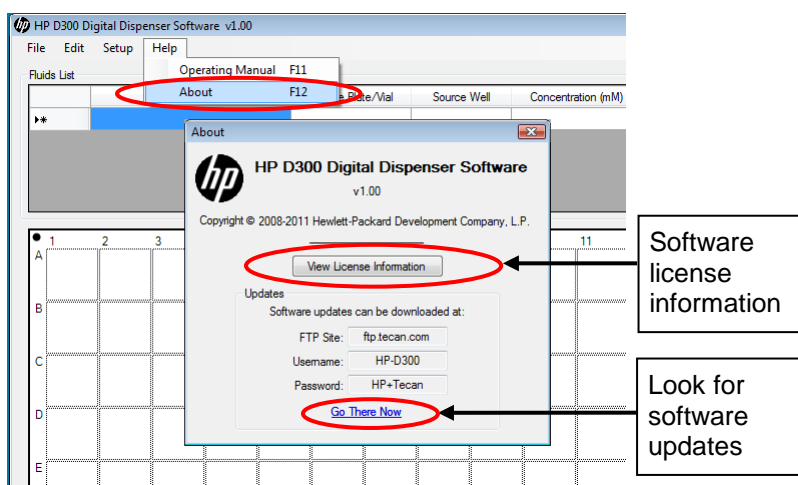
Figure 4-6 HP D300 Digital Dispenser main operating window



The application has a standard MS Windows[®] look and feel. Once you get started, you can work in different areas of the main operating window by clicking in them, you can select cells by left-clicking, control-clicking or shift-click combined with dragging, you can right-click to perform context sensitive operations, or see information about wells that contain fluids by hovering over them in the well plate.

1. Note that this Operating Manual is accessible two different ways, either by opening the pdf file in the C:\Program Files\HP-D300 directory, or by clicking on Help/Operating Manual from the main operating window as shown in **Figure 4-7**.

Figure 4-7 Help menu and Help/About Window




2. Note that the About window also provides access to the License information and to the Tecan FTP site that always contains the latest version of the software. Click on **Go There Now** to directly access the FTP site from the D300 Digital Dispenser software.
3. Select either 96 or 384 from the **Well Plate** drop-down list box.

4. Put at least one fluid in the Fluids List, by entering any characters in the Name cell, or by copying and pasting a list of compounds from an Excel file. The order of the fluids in the Fluids List is the order in which the fluids will be dispensed.
5. Learn how to select and deselect wells in the plate design:
 - Select any number of wells by clicking and dragging in the well plate image. Any well which is partially selected becomes one of the selected wells. Give it a try!
 - Select nonadjacent wells by holding down the CTRL key while selecting wells.
 - Deselect wells by clicking on them again while holding down the CTRL key.
 - Deselect all selected wells by clicking on other wells or clicking on the edge of the well plate (outside the wells) while not holding down the CTRL key.
6. Just for fun, select some wells, then right-click on them to open the Edit Well Plate menu. Click on **Set Volume** or **Set Concentration**, enter the number **1** in the volume or concentration box and click **OK**. The wells you selected will turn red because they now contain fluid 1 in your plate design.

In general, this is a great way to learn to use the software—try things out, right- or left-click when you are unsure, and move the mouse over areas to check them out.

Don't worry, you won't break anything!

NOTE: Either **Set Volume** or **Set Concentration** is available on the well plate options list depending on whether **Volume** or **Concentration mode** was selected in the Options: Edit: Units. See "[Options: Edit: Units](#)" for how to set Volume or Concentration mode.

 **TIP:** If the Set Volume or Set Concentration option is grayed out, the fluids table is empty, so follow step 4 above to add a fluid to the fluid table.

7. Now that you have set up some wells, place the cursor over a colored well and observe the information that appears in the well pop-ups.

Note that the first three values (assay volume, total volume, and DMSO content) regard the entire contents of the well. Below these values, information is given separately for each fluid in that well. With the exception of assay volume, there are two values for each well pop-up entry: the first value and then a second value enclosed by square brackets, for example [0.960 nL]. The first value is the volume or concentration that was requested, and the second bracketed value is what will actually be dispensed since only an integer number of drops can be dispensed. This is where the name Digital Dispenser comes from, an integer number of ultrasmall droplets.

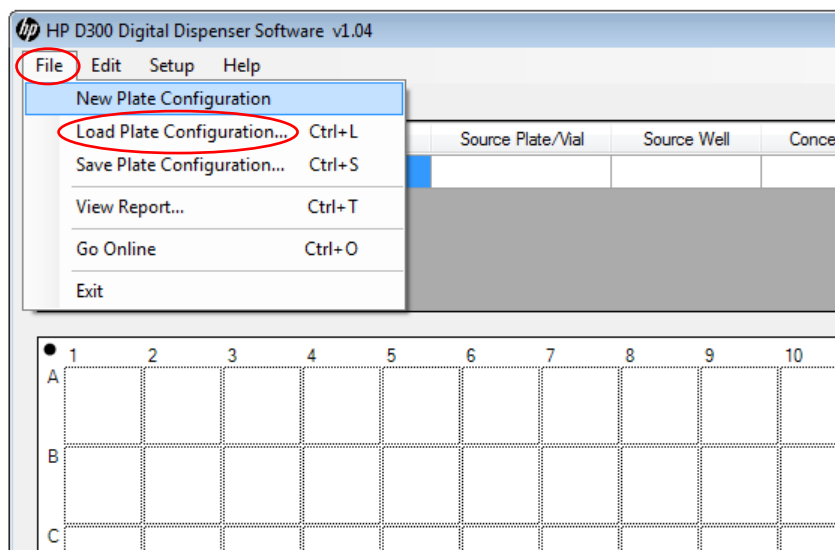
NOTE: Two different drop volumes are used in the dispenser. For dispense volumes less than 3.5nL, the dispenser uses the closest number of either 13pL or 20pL drops to dispense as close as possible to the requested volume. For volumes greater than 3.5nL, the dispenser uses the closest number of 400pL bursts (20 drops simultaneously dispensed from 20 nozzles which are each 20pL) in order to dispense very close to the requested volume very rapidly.

4-3 Load an existing plate configuration

NOTE: To build a new plate configuration, see "[Create plate configurations](#)".


8. On the HP D300 Digital Dispenser main operating window, click **File** and then **Load Plate Configuration**, then locate and select a plate configuration (.xls) that has been previously saved on your computer. Example configurations are in C:\Program Files\HP-D300\Configs. Many of the settings, including the Fluids List, assay volume, DMSO limit, plate layout and the plate jitter option, will change to reflect the configuration you select. See [Figure 4-8](#). Up to five plates can be included in a Plate Configuration for multiplate configurations.

Figure 4-8 Click **File** and then **Load Plate Configuration**



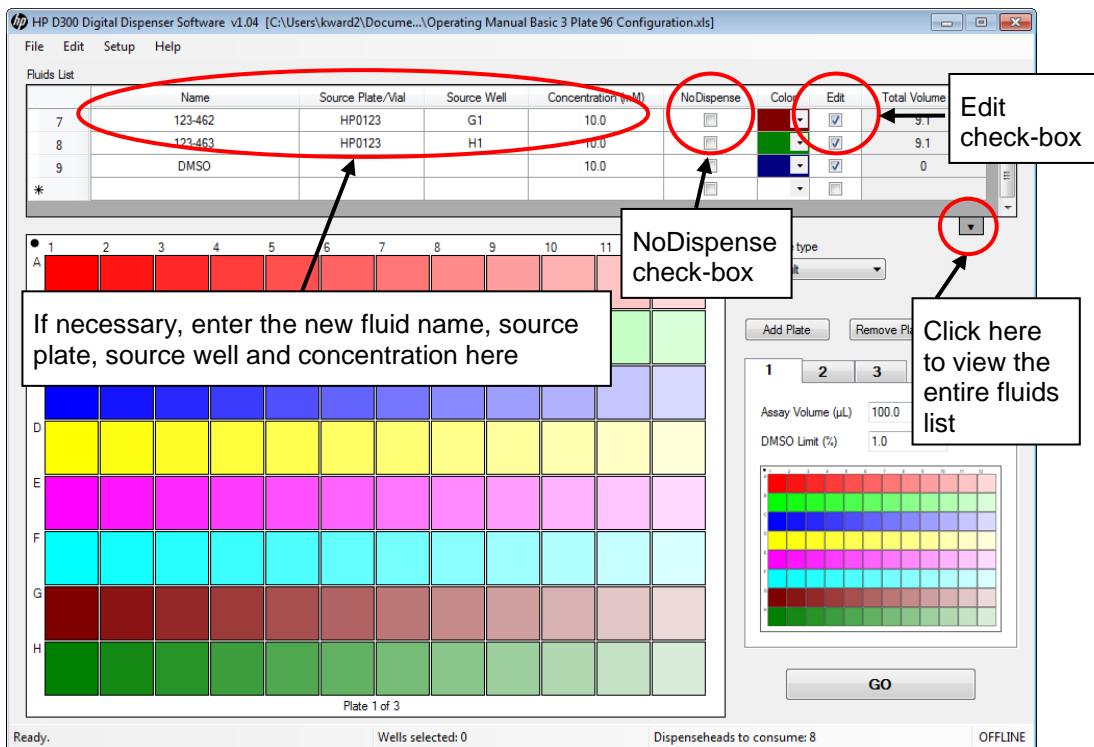
9. If the Fluids List needs to be modified, enter the new fluid name, source plate, source well and concentration in the Source Plate/Vial, Source Well and Concentration columns in the Fluids List. Only the fluid name and concentration are required. See [Figure 4-9](#).

NOTE: These identifiers may be typed into the Fluids List individually, or they may be entered by scanning a barcode, or pasted in from an Excel or other tab-separated file that has a compatible layout. To copy and paste information from another file, the information must be arranged in columns as the Fluids List is, but it is not necessary that the originating table contain all of the columns that the Fluids List contains. Once you have pasted information into the Fluids List, if certain cells (such as those in the Concentration column) do not contain the proper numerical data, then default values will be used for those cells.

 **TIP:** Paste information from another file into the Fluids List just as you would in any other Windows program: click in the upper left-most cell that you want to paste into, then right-click and select **Paste**.

- If the **NoDispense** check box is selected, you can program that fluid just like the other fluids, but it will not be dispensed. This may be useful when you want to maintain information in the output report file about wells that have not been dispensed into. See [Figure 4-9](#).
 - If the **Edit** check box is deselected, that fluid cannot be added to the configuration and will not be displayed in the plate layout. This fluid will still be displayed in the Global View layout if it was added to any wells prior to deselecting the Edit check box, and it will still be dispensed. Deselecting the Edit check box may be useful when you have added multiple fluids to individual wells and want to view or manipulate one or more fluids in the well, but not all the fluids in the well.
10. Click on the arrow at the lower-right corner of the Fluids List to display the entire Fluids List. See [Figure 4-9](#).

Figure 4-9 Main operating window showing Fluid Table options.

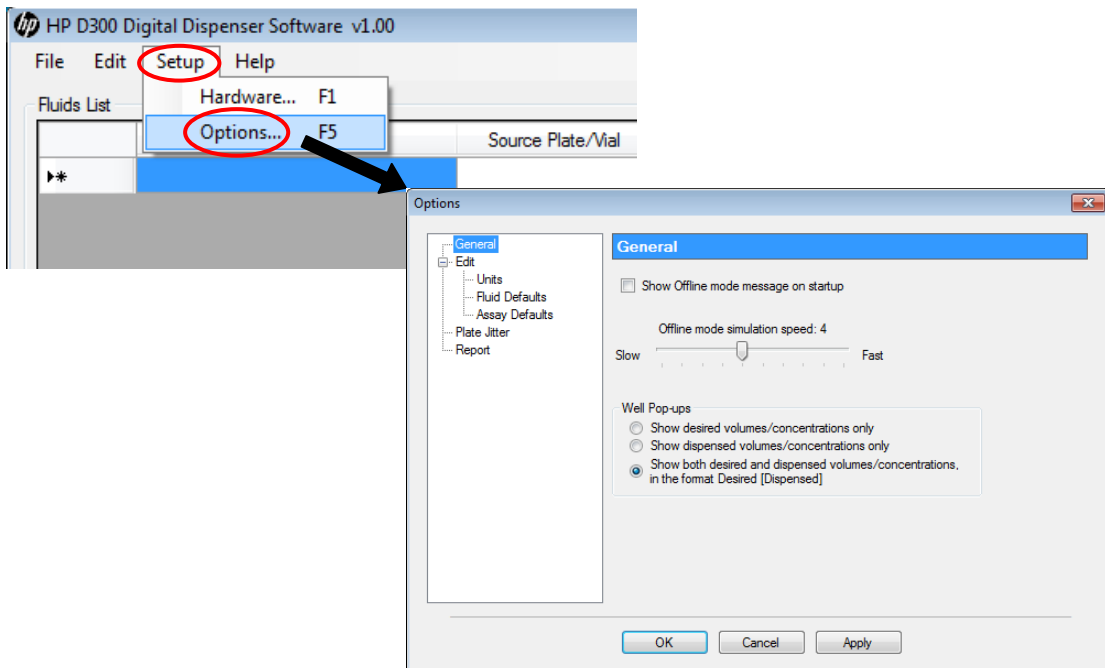


NOTE: The Fluids List can also be managed by clicking **Edit** and then **Fluids List**.

4-4 Get to know the Options dialog box

At the HP D300 Digital Dispenser main operating window, click **Setup** and then **Options** to open the Options dialog box.

Figure 4-10 Click **Setup** and then **Options** to open the Options dialog box

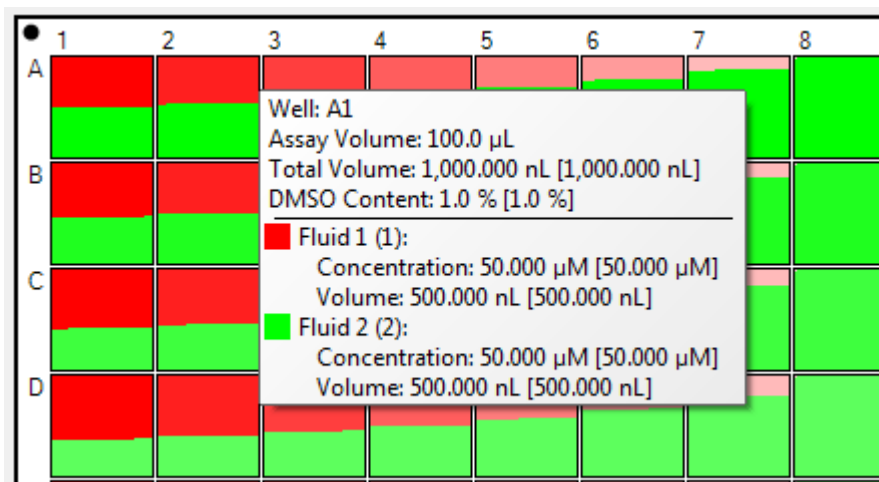


4-4-1 Options: General

From the main operating window, click **Setup**, then **Options**, then **General** to open the General pane of the Options dialog box. See [Figure 4-10](#). In the General pane, you can choose whether to show the Offline mode message when you use the software when not connected to an HP D300 Digital Dispenser. When in Offline mode, you can choose how rapidly to “pseudo-dispense” upon using the Go button using the Offline mode simulation speed. You can also choose whether to show the requested volume or the dispensed volume, or both, in the Well Pop-ups, as described below. For small volumes, the dispensed volume may be slightly different than the requested volume due to the integer number of drops resolution of the Digital Dispenser.

Well pop-ups are the information about the contents of a well that is displayed when you move the mouse over the well plate image on the main operating window. Note that when “Show both requested volume and dispensed volumes/concentrations” is selected, the format of the pop-up is Requested [Dispensed], as shown in [Figure 4-11](#), such that the requested volumes and concentrations are shown first and the dispensed volumes and concentrations are shown next, enclosed in brackets.

Figure 4-11 Example of a well pop-up that displays both requested and [dispensed] volumes and concentrations

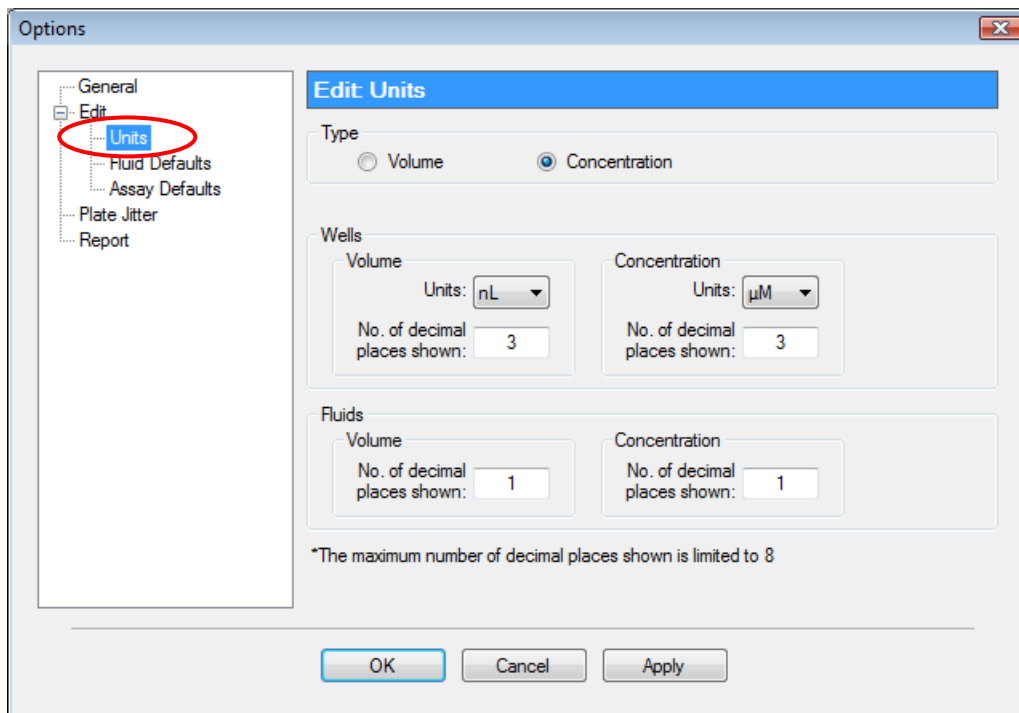


4-4-2 Options: Edit: Units

In the **Options** dialog box, click **Edit** and then Units to open the Edit: Units pane of the Options dialog box. See [Figure 4-12](#).

- The Type area of the dialog box allows you to determine whether the setup mode for titrations is specified using concentration or volume.
- The Wells area allows you to specify the unit type for volume and concentration and to specify the number of decimal places used in plate setup.
- The Fluids area allows you to specify the number of decimal places used in the Fluids List.

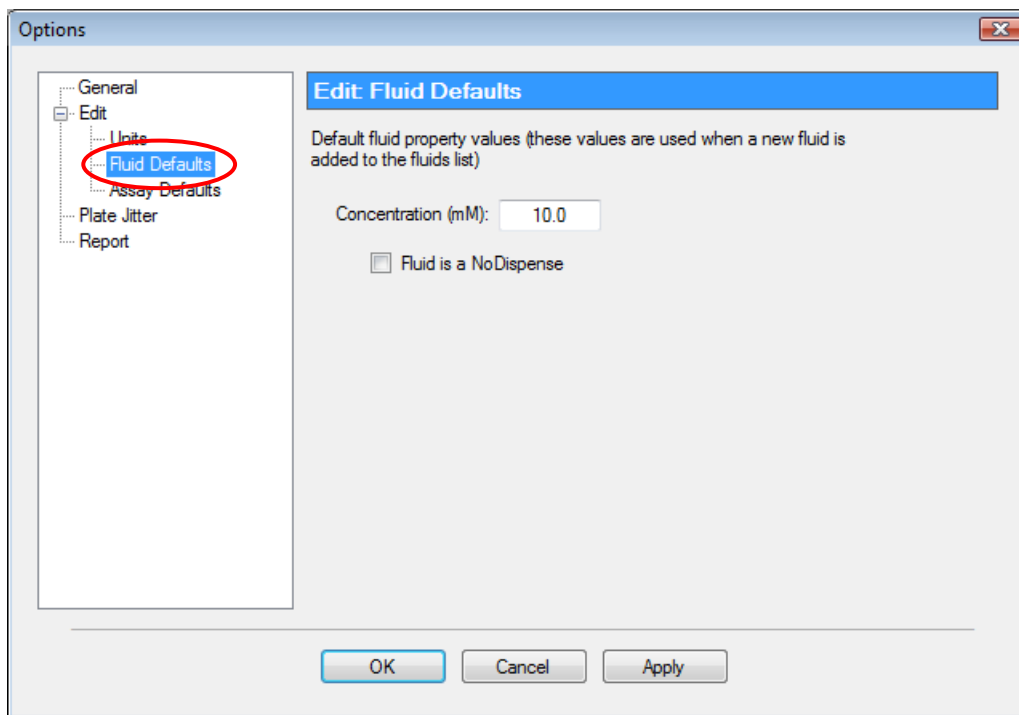
Figure 4-12 Edit: Units pane of the Options dialog box



4-4-3 Options: Edit: Fluid Defaults

In the Options dialog box, click **Edit** and then **Fluid Defaults** to open the Edit: Fluid Defaults pane of the Options dialog box. Set the default concentration for new fluids and specify whether default fluids will be dispensed. See **Figure 4-13**.

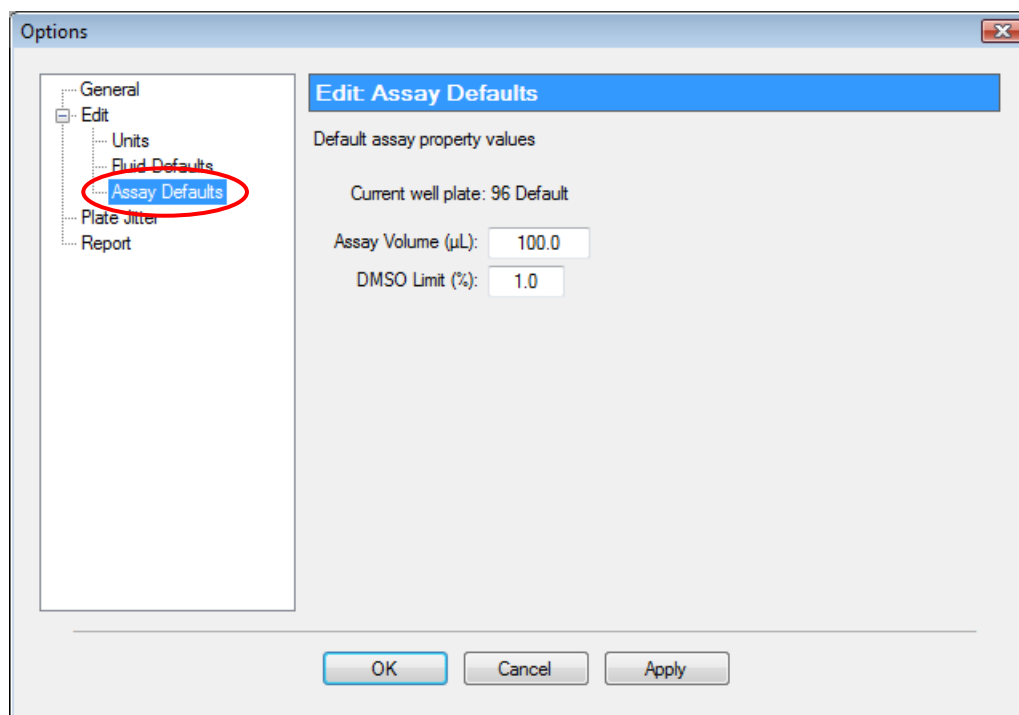
Figure 4-13 Edit: Fluid Defaults pane of the Options dialog box



4-4-4 Options: Edit: Assay Defaults

In the Options dialog box, click **Edit** and then **Assay Defaults** to open the Edit: Assay Defaults pane of the Options dialog box. Set the default assay volume and the default DMSO limit here. See [Figure 4-14](#).

Figure 4-14 Edit: Assay Defaults pane of the Options dialog box



4-4-5 Options: Plate Jitter

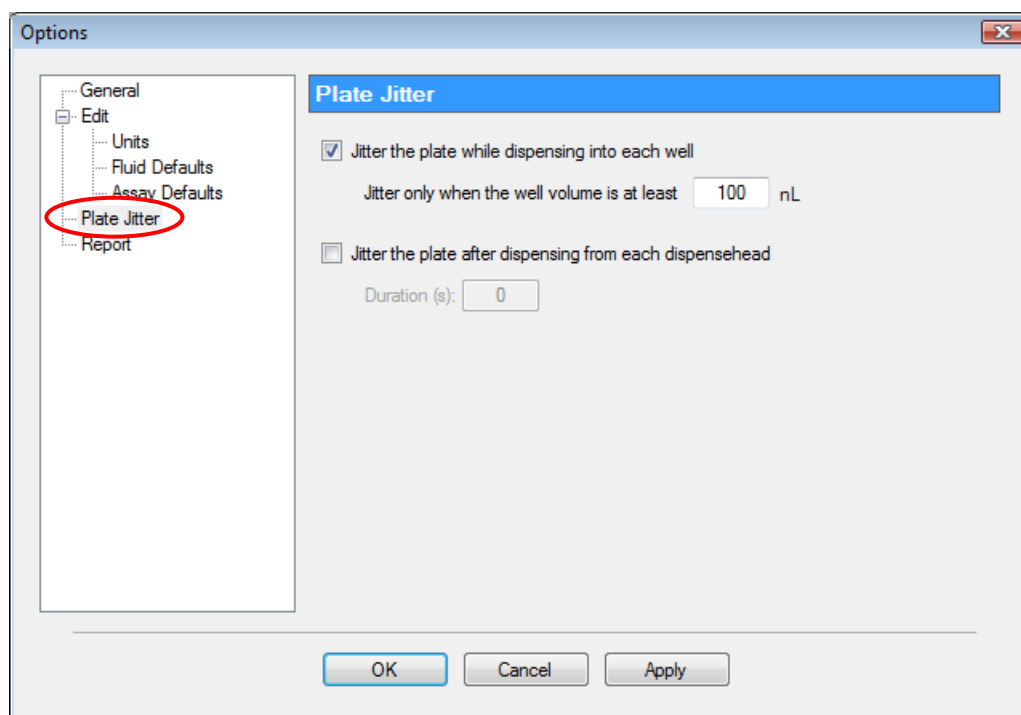
In the Options dialog box, click **Plate Jitter** to open the Plate Jitter pane of the Options dialog box. See [Figure 4-15](#). Use plate jitter if shaking the destination plate is desired either during and/or after dispense, for example if you are dispensing into wells containing cells.

Select **Jitter the plate while dispensing into each well** to shake the destination plate during the dispense. When this option is chosen, shaking only above a specified volume may be entered. Typically, only dispense volumes greater than 100nL have a deleterious effect on plated cells.

Additionally, **Jitter the plate after dispensing from each Dispensehead** may be selected to shake the destination plate post-dispense. If this option is utilized, enter the number of seconds to shake after each Dispensehead has completed dispensing.

! IMPORTANT: Plate Jitter while dispensing is highly recommended when dispensing directly into plates containing live cells to reduce the deleterious effect of a DMSO bolus on the cells.

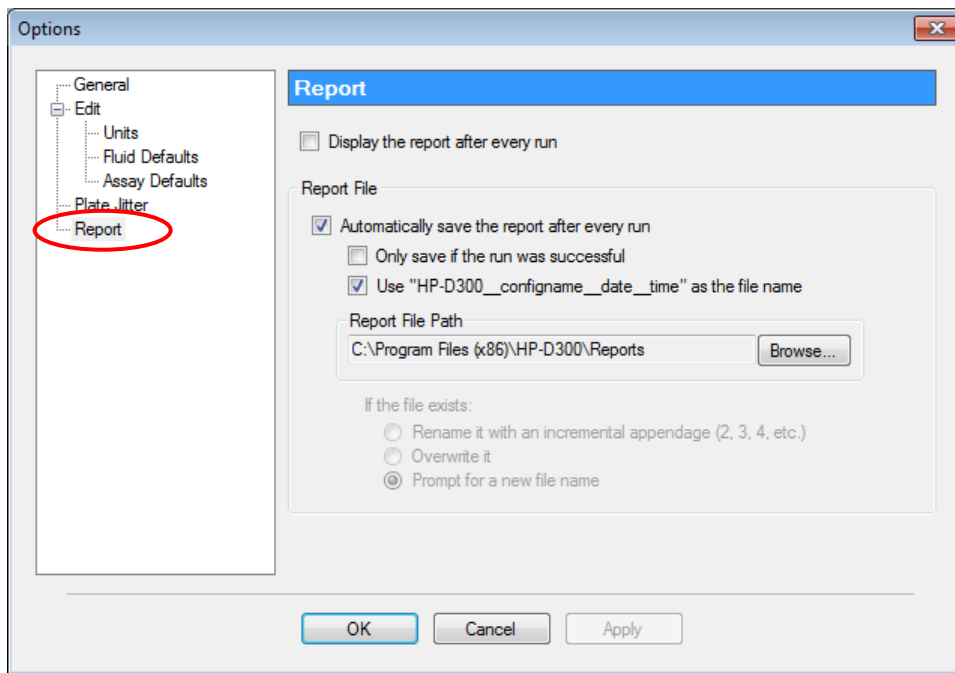
Figure 4-15 Plate Jitter pane of the Options dialog box



4-4-6 Options: Report

In the Options dialog box, click **Report** to open the Report pane of the Options dialog box as shown in [Figure 4-16](#). Use the Report pane to alter options related to the output report that the HP D300 Digital Dispenser software automatically generates after every dispense run (plate). In the Report pane, you can choose to **Display the report after every run**, alternatively you would choose File/View Report or use Excel to view the report after the run. Select **Automatically save the report after every run** to save Report files, and **Only save if the run was successful** to omit reports for aborted runs. You may also choose to use the default report naming convention by selecting **Use “HP-D300_configurationname_date_time” as the file name** and enter the **Report File Path** where these reports should be saved, or if this option is not selected, Report File Path will change to **Report File Name** where you enter your own report file location and naming convention. When this option selected, you can choose to rename each new report with automatic incrementing, overwrite existing reports or to prompt you for a new file name at the completion of each run.

Figure 4-16 Report pane of the Options dialog box

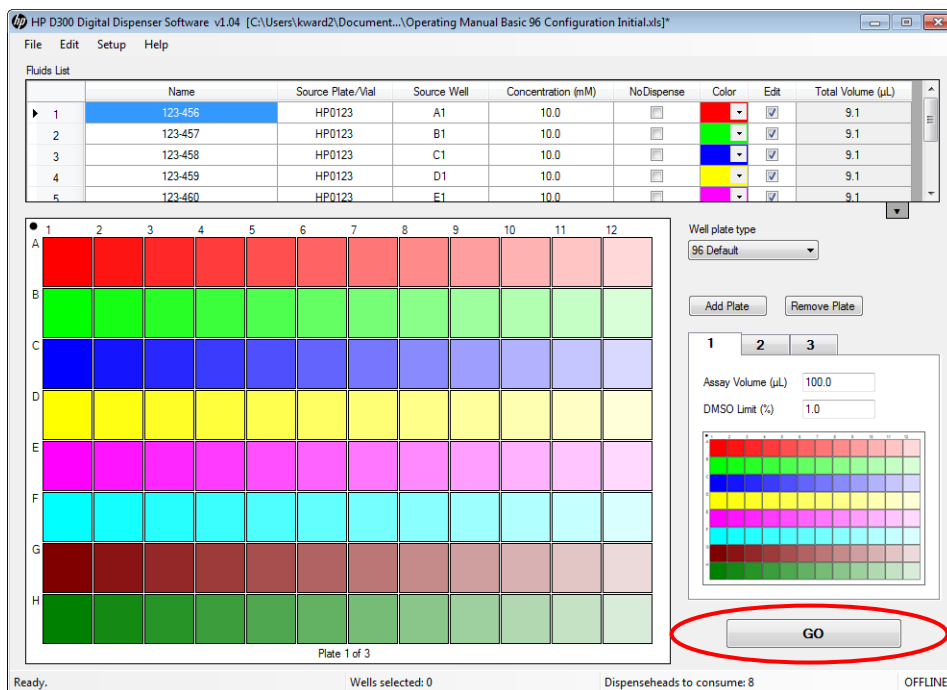


4-5 Plate dispense procedure

4-5-1 Initiate plate dispense

After selecting the desired plate configuration and verifying the options, click the **GO** button in the lower-right corner of the main operating window. See [Figure 4-17](#).

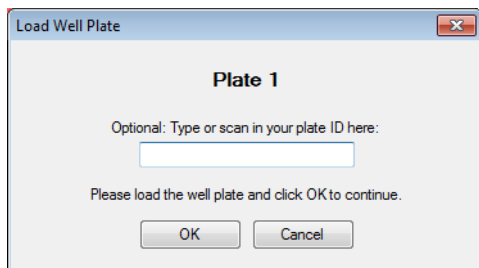
Figure 4-17 Main operating window



4-5-2 Scan and load the well plates

1. Optional. If the destination well plate has an identification code you would like to track, scan the bar code on the well plate using a USB bar code reader attached to your computer, or enter its identification number manually into the box on the Load Well Plate dialog box. See [Figure 4-18](#).

Figure 4-18 Load Well Plate dialog box. Be very careful to load the plate that is requested.



2. Place the well plate on the destination plate holder. See [Figure 4-19](#). Engage the top left corner of the well plate on the pillars first, then snap the well plate fully into the spring clips.
 - Make sure the plate is completely level and down on the destination plate holder.
 - Ensure that the A1 well position is at the top-left corner of the destination plate holder.
3. Click **OK**. The Load Cassette dialog box appears. See [Figure 4-20](#).

Figure 4-19 Destination plate holder on the dispenser

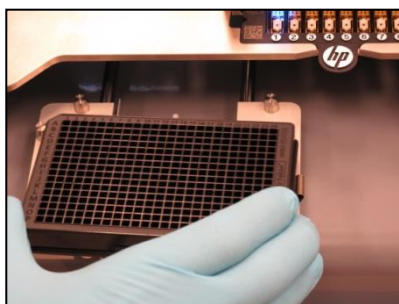
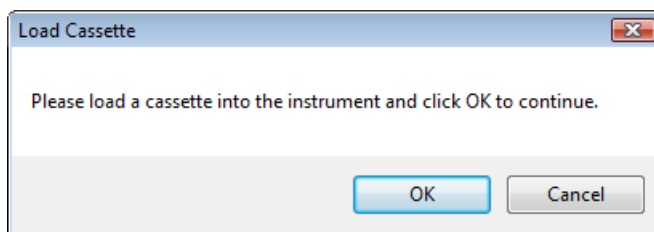


Figure 4-20 Load Cassette dialog box



4-5-3 Load a Dispensehead Cassette

1. If a Dispensehead Cassette is already in place with unused Dispenseheads, click **OK** in the Load Cassette dialog box to continue (see [Figure 4-20](#)). If you are unsure whether there are any unused Dispenseheads on the Cassette, click **OK** and the dispenser will test the Dispenseheads to find which ones are unused.
2. Load a new Dispensehead Cassette if necessary as shown in [Figure 4-21](#):
 - a. If a Cassette is already loaded in the dispenser, lift the tab with the HP logo and pull the Cassette out of the deck.

- b. Lift a new Cassette out of the clamshell container and place it white logo side up into the cutout on the Dispenser deck.
- c. Make sure it is fully inserted so the top surface of the Cassette is the same height as the top of the deck all the way around the cassette.

Figure 4-21 Loading a new Dispensehead Cassette



3. Once the Cassette is fully inserted, click **OK** in the Load Cassette dialog box (see [Figure 4-20](#)). The pogo mechanism will engage and the Dispenseheads will be electrically tested to ensure quality. Then the Load Fluid dialog box appears. See [Figure 4-22](#).

4-5-4 Load fluid into the Dispensehead

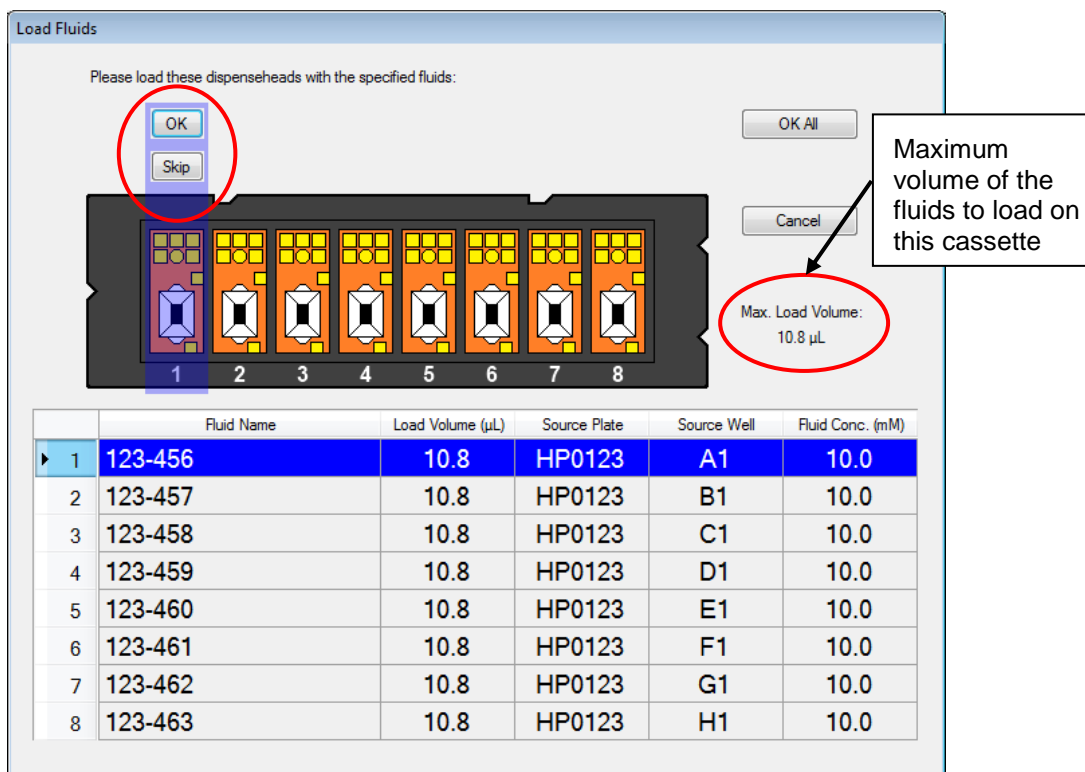
When the Load Fluid dialog box opens, the OK and Skip buttons flash above the first Dispensehead that will be loaded (see [Figure 4-22](#)), and a blue LED illuminates the first Dispensehead on the front of the pogo mechanism.

1. Fill a low volume pipette with at least the amount of fluid that is indicated in the Load Fluid dialog box, or fill a multipipetter with the appropriate number of fluids in the proper order as shown in the Load Fluids dialog box. If a multipipetter is used, use the Max. Load Volume for all Dispenseheads as shown on the right side of the dialog, as circled in [Figure 4-22](#). The source plate and source well location of the fluid to load will also be identified in the Load Fluid dialog box if that information was entered in the Fluids List. Be sure to load the proper compound into the proper Dispensehead.



CAUTION: Using a multichannel pipette enables the loading of multiple Dispenseheads simultaneously. Make sure that you fill the Dispenseheads with the **LARGEST** load volume of the set of Dispenseheads you are loading. It is indicated in the upper right of the dialog box ("Max. Load Volume") as shown in [Figure 4-22](#).

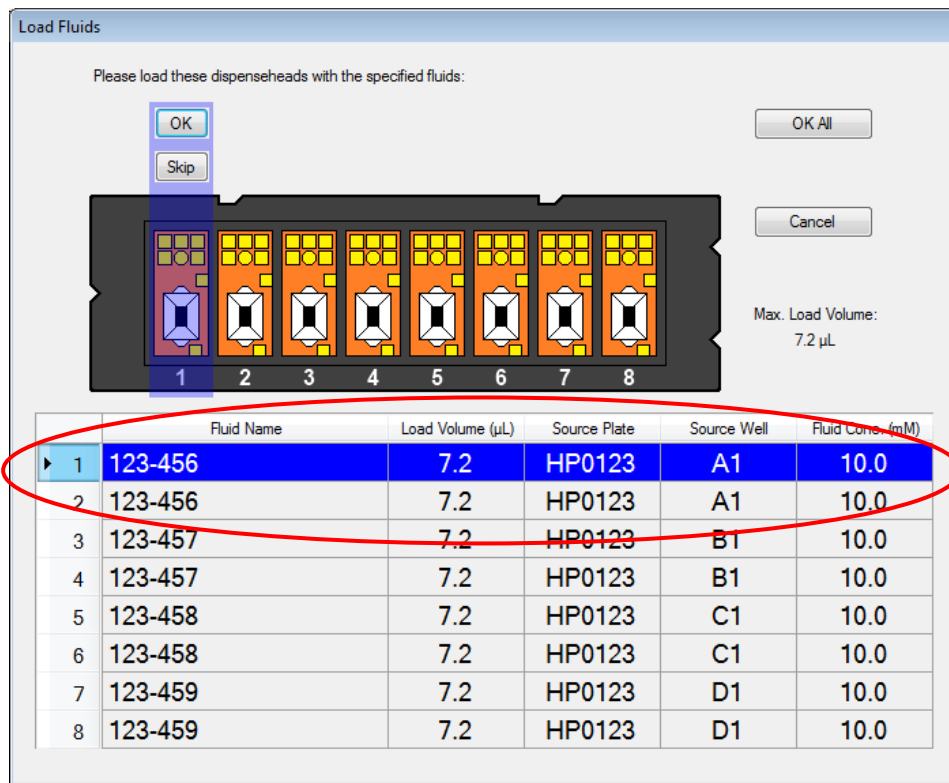
Figure 4-22 Load Fluid dialog box



NOTE: The amount displayed in the Load Volume column in the Load Fluid dialog box (**Figure 4-22**) will always be greater than the amount of fluid that was requested to be dispensed. The volume to be dispensed for each fluid is labelled as the “Total Volume” in the right hand column of Fluids Table on the main operating window. The difference between the dispense volume and the load volume is to account for fluid which is retained in the Dispensehead after dispense, also known as the dead volume. The dead volume is dependent upon the amount of fluid loaded into the Dispensehead, with greater dispense volumes leading to greater dead volumes.

NOTE: The maximum amount of fluid that can be dispensed from a Dispensehead is 10µL. If the total dispense volume of a fluid exceeds 10µL, the volume will be split between two or more Dispenseheads. The split will be identified in the Load Fluid dialog box and in the report file (as “1 of 1” or “1 of 2,” for instance). See **Figure 4-23** for an example of using two Dispenseheads per fluid.

Figure 4-23 Load fluids dialog box with two Dispenseheads per fluid



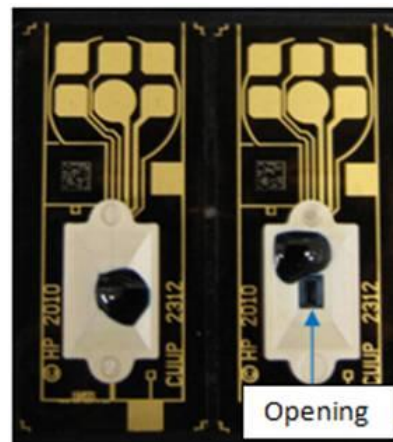
2. If you determine that the Dispensehead in the load position is not good, or if you mistakenly load the Dispensehead incorrectly or with the wrong fluid, press **Skip** instead of **OK**. This will cause the blue LED indicator to move to the next good position, and you will be prompted to load the fluid that was skipped into that next Dispensehead.
3. Load fluid into the Dispensehead position indicated in the Load Fluid dialog box when prompted. This Dispensehead will be indicated on the dispenser by an illuminated blue LED. See [Figure 4-24](#).

WARNING: Make sure to load the fluid into the opening on top of the Dispensehead. Do **NOT** depress the pipette plunger to the second "blow out" stop as this could introduce air into the Dispensehead.

Figure 4-24 A blue LED illuminates the Dispensehead on the Dispensehead Cassette



Figure 4-25 Examples of a good load and a bad load

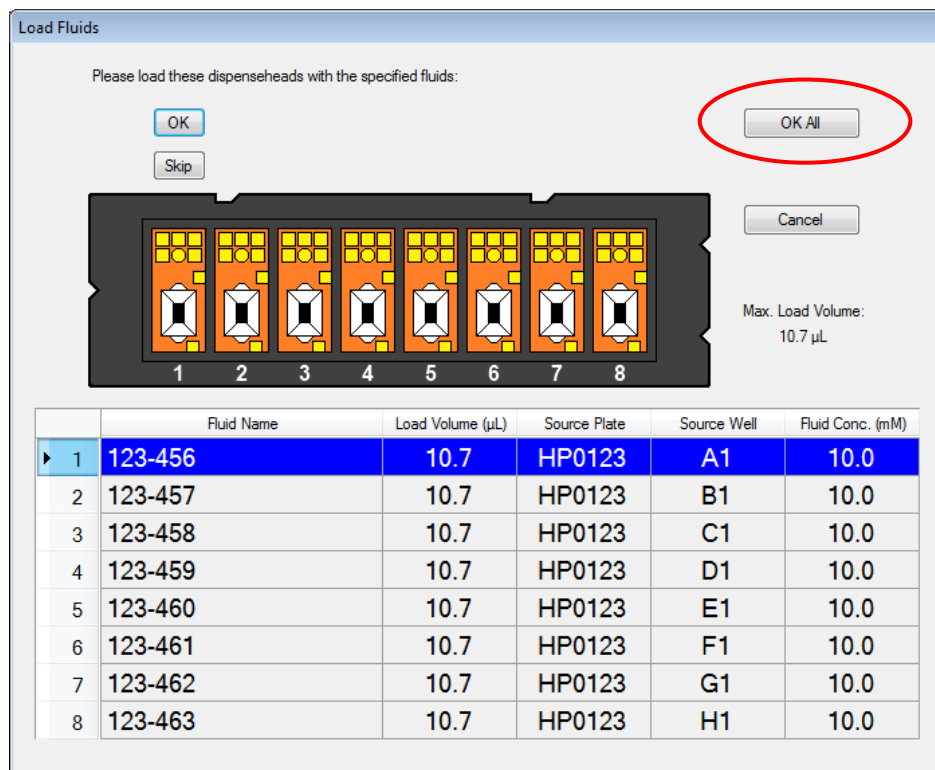


Good load

Bad load

4. Click **OK**. All dispensing for the current fluid will occur. In the main operating window, only the wells for the current fluid are shown, and each well is covered with an X while it is being dispensed and then turns black when the dispensing for that well is complete.
5. Repeat steps 1 through 4 to load and dispense each fluid in the plate configuration. Fluid loading can occur during the dispense operation; you do not have to wait until the current Dispensehead has finished. Just remember to click on **OK** as each fluid is loaded in its Dispensehead, or to click on **OK All** when all Dispenseheads of the current Cassette are loaded, as shown in **Figure 4-26**.
6. If loading all Dispenseheads simultaneously with a multichannel pipette or individually prior to starting the dispense, then after all Dispenseheads of the current Cassette are loaded, click on **OK All** as shown in **Figure 4-26**.

Figure 4-26 Load fluid dialog box showing OK All button



7. After the fluids in the currently loaded cassette are dispensed into plate 1, then depending upon the configuration, you will be asked to either load a new cassette or a new plate. Read the prompt dialog box carefully to determine the next step.

WARNING: For multiplate configurations, the D300 software first minimizes the use of dispenseheads, so that fluids which are used on multiple plates are dispensed into all of those plates as long as the total dispense volume for that fluid does not exceed 10uL. This dispensehead conservation can result in a non-intuitive plate loading order. Be careful to load the correct well plate as prompted by the software! Minimizing the number of dispenseheads used may sometimes result in more plate swaps than expected.

8. If you are prompted to load the next plate in the configuration, you will see a prompt similar to **Figure 4-18**, where you can enter or scan a name for the plate. If the plate has been loaded previously, you will be prompted with the plate name you previously entered or scanned in for the plate, along with the plate number.

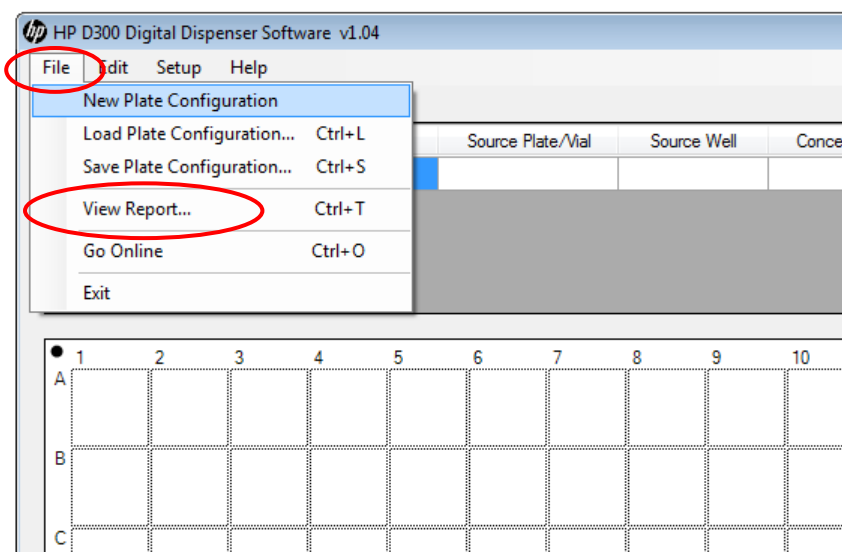
9. After the current set of fluids is dispensed into all appropriate plates, if needed the software will prompt you to load a new cassette using the message shown in [Figure 4-20](#). After a fully used T8 cassette is removed from the D300 dispenser it may be disposed, as it will not be reused.
10. You will then be shown which fluids to place in each dispensehead of the new cassette as shown in [Figure 4-22](#), and steps 7 - 9 above will continue until the configuration is complete.

4-6 View the report

Depending on the options selected in the Report pane, a report may be automatically generated and saved on your computer after the plate dispense operation is completed. To change these report settings, click **Setup**, then **Options**, and then **Report** (see “[Options: Report](#)”).

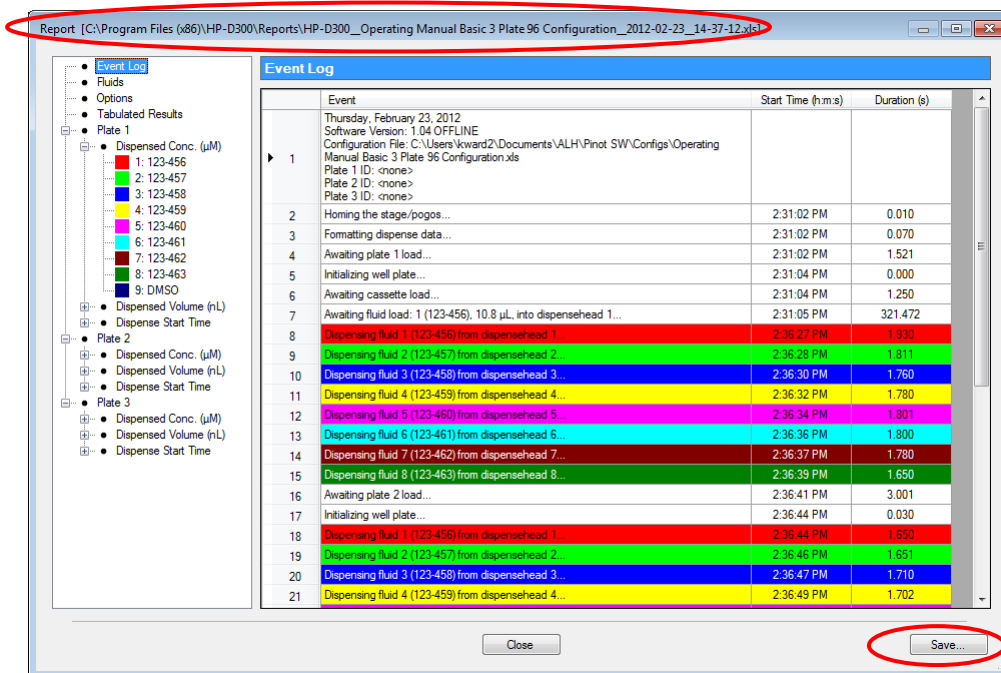
If you have chosen not to display the report after each run, you can still view data from the most recently completed run by clicking **File** and then **View Report**. See [Figure 4-27](#).

Figure 4-27 Open the Report dialog box by clicking **File** and then **View Report**



If you are not sure if a report was automatically saved, look at the title bar of the Report dialog box (see [Figure 4-28](#)). If a file name is displayed in the title bar, then the report was automatically saved. Even if you did not choose to automatically save the report, you can still save the current report by clicking the **Save** button in the lower-right corner of the Report dialog box.

Figure 4-28 Report dialog box



When the Report dialog box opens, the Event Log pane is initially displayed, but information about the fluids and assay volumes can be examined by clicking **Fluids** or **Options** in the menu on the left pane. To view more information about the dispense operation, click the menu options to section of the report you want to view as shown in **Figure 4-29** and **Figure 4-30**.

Figure 4-29 Report Fluids sheet

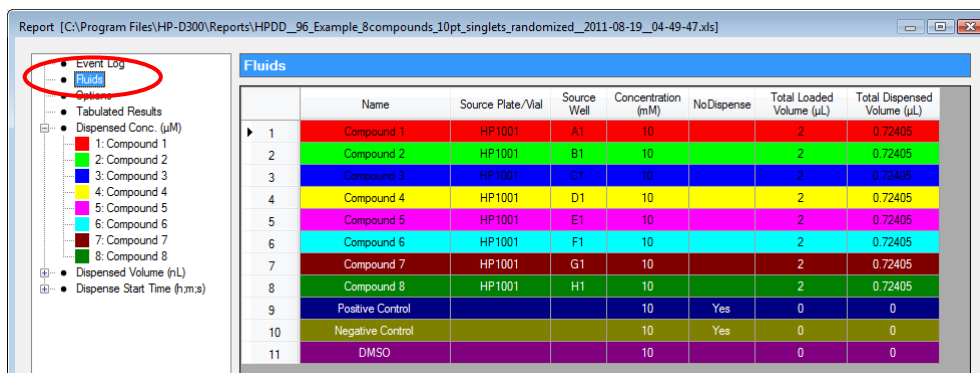
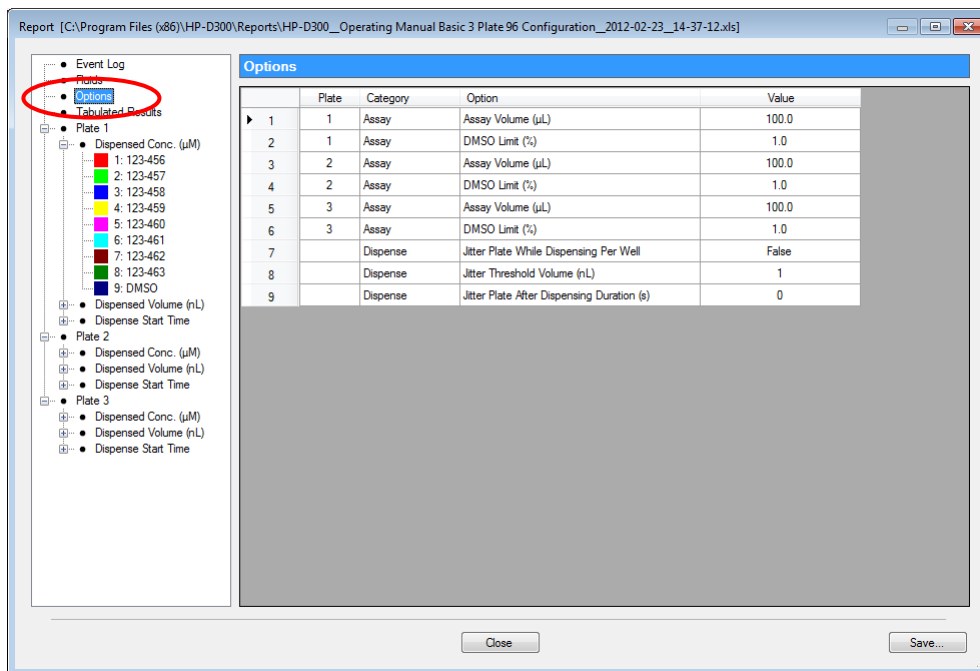


Figure 4-30 Report Options sheet



While most data is displayed in rectangular plate format, the Tabulated Results pane displays concentration data in long skinny columns in order to sort randomized plates. See [Figure 4-31](#). To derandomize your data in order to create IC50 or EC50 plots, use this report file saved in Excel format, and add a column of your plate reader data also in this long skinny format. Next, sort the data by fluid name and concentration in order to process or plot the data by compound and concentration.

Figure 4-31 Tabulated Results pane

Report [C:\Program Files (x86)\HP-D300\Reports\HP-D300_Operating Manual Basic 3 Plate 96 Configuration_2012-02-29_17-08-56.xls]

Tabulated Results

	Well Plate	Well Row	Well Column	Fluid	Dispensed Conc. (μM)	Dispensed Conc. (μM)	Dispensed Conc. (μM)
					<COMPOSITE>	123-456	123-457
0	1	A	1	123-456	100	100	
1	1	A	2	123-456	90.92	90.92	
2	1	A	3	123-456	81.8	81.8	
3	1	A	4	123-456	72.72	72.72	
4	1	A	5	123-456	63.64	63.64	
5	1	A	6	123-456	54.56	54.56	
6	1	A	7	123-456	45.44	45.44	
7	1	A	8	123-456	36.36	36.36	
8	1	A	9	123-456	27.28	27.28	
9	1	A	10	123-456	18.2	18.2	
10	1	A	11	123-456	9.08	9.08	
11	1	A	12	123-456	0.0013	0.0013	
12	1	B	1	123-457	100		100
13	1	B	2	123-457	90.92		90.92
14	1	B	3	123-457	81.8		81.8
15	1	B	4	123-457	72.72		72.72
16	1	B	5	123-457	63.64		63.64
17	1	B	6	123-457	54.56		54.56
18	1	B	7	123-457	45.44		45.44
19	1	B	8	123-457	36.36		36.36
20	1	B	9	123-457	27.28		27.28
21	1	B	10	123-457	18.2		18.2
22	1	B	11	123-457	9.08		9.08

Close Save...

Any previously saved report must be viewed in Excel. If you have trouble finding the reports, the folder that the last report was saved in is listed in the Report pane of the Options dialog box (click **Setup**, then **Options...**, and then **Report**).

4-7 Turn off the HP D300 Digital Dispenser

If you are finished using the dispenser and will not use it again for an extended period of time, turn it off as follows:

1. Close the HP D300 Digital Dispenser software, either by clicking the red X in the upper right corner of the main operating window, or by clicking on **File/Exit**.
2. Turn the Dispenser off using the main power switch on the rear of the dispenser. (Press down the “O” of the rocker switch to turn the dispenser off.)

Figure 4-32 On/off switch in the ON position



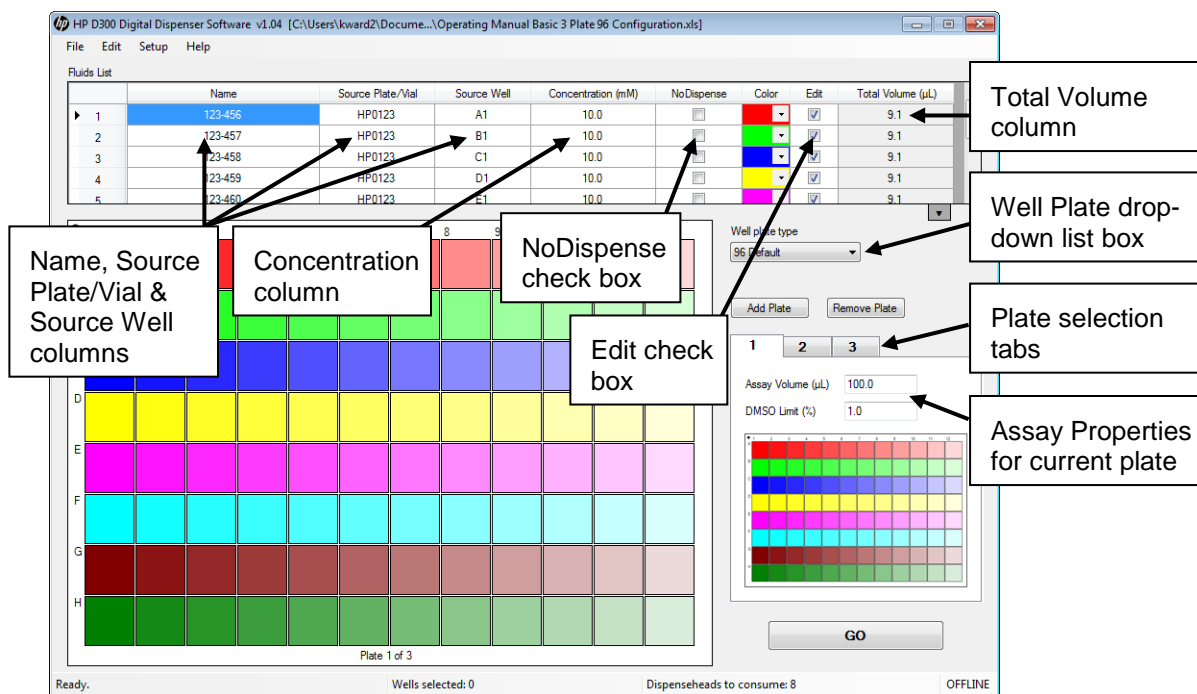
3. Remove any unused Dispensehead Cassettes from the dispenser to avoid contamination from cleaning solutions or the laboratory environment and place them back in the Cassette tray.
4. Clean the dispenser according to the cleaning procedures described in the “**Clean the dispenser**” section of this document.

5 Create plate configurations


5-1 Create the fluids table


1. On the right side of the main operating window, select the type of well plate you will use from the **Well Plate** drop-down list box, either 96 or 384. Up to five plates can be part of a single configuration, but all of them must be of the same type, 96 or 384. The same fluids, or different fluids, can be used in all the plates in a configuration.

Figure 5-1 The HP D300 Digital Dispenser main operating window



2. Enter one or more fluids (compounds to dispense in this set of plates):
 - a. Click in a cell in the Name column of the Fluids List and type the name.
 - b. Optional: click in a cell in the Source Plate/Vial column and type the source plate name.
 - c. Optional: click in a cell in the Source Well column and type the source well location.

 **TIP:** If desired, information can be entered into the Name, Source Plate/Vial columns by scanning a bar code, using a USB bar code reader attached to your computer.

 **TIP:** The Fluids List can also be created by copying and pasting information from an Excel spreadsheet that contains some or all of the fluid data (see "[Get familiar with the software](#)"). Fluid information in the Excel file must have columns in the same order as the Fluids Table.

- d. Enter the source fluid concentration in the **Concentration** column.
- e. Select a color for each of the fluids in the Color column if you do not want to use the default colors.
- f. Repeat steps **a** through **e** for all of the fluids that will be dispensed into this set of well plates. Up to five plates to be run together can be saved in a single configuration file.

NOTE: If no value is entered into the Concentration column, the default values set in the Edit: Fluid Defaults pane of the Options dialog box will be used for those cells.

3. Type the plate assay volume and the DMSO limit for the first plate into the **Assay Properties** box. If more than one plate is in a configuration, they may each have different assay volumes and DMSO limits.

NOTE: If the fluids to be dispensed into a well exceed the DMSO limit, a warning will be generated and the well pop-up data displayed for that well will appear in red; however, the Dispenser will dispense whatever concentration/volume you have specified after you have been warned.

4. In order to program a fluid into wells, the **Edit** check box in the Fluids List for that fluid must be selected. When the **Edit** check box is selected, that fluid can be added or removed from wells, and is visible in the well plate image. Fluids with the **Edit** box not checked cannot be added or removed from wells, and if that fluid was previously programmed into wells, it will remain in the well plate layout and will be dispensed during the **Go** process, but will not be shown in the well plate image. Clear the **Edit** check box if you want to simplify the view or copy and paste only some fluids in wells where multiple fluids are being dispensed. The effect of unchecking an Edit check box is shown in [Figure 5-4](#).
5. To set a fluid or fluids to be non-dispensed, select the **NoDispense** check box in the Fluids List. This may be useful when you want to maintain information about nondispensed fluids in the output report such as for manually or robotically pipetted controls, or for blanks.
6. To program more than one well plate to be run with the same fluids list, select **Add Plate**. To remove the current plate from the current configuration, select **Remove Plate**.

The **Total Volume** column values cannot be changed. The total volume is the sum of the volume of a fluid in all of the wells of the plate, also known as the dispense volume of the fluid. This total volume is less than the volume you will be asked to load in the DispenseHead Cassette due to the amount of fluid retained in the DispenseHead after dispense, also known as the dead volume.

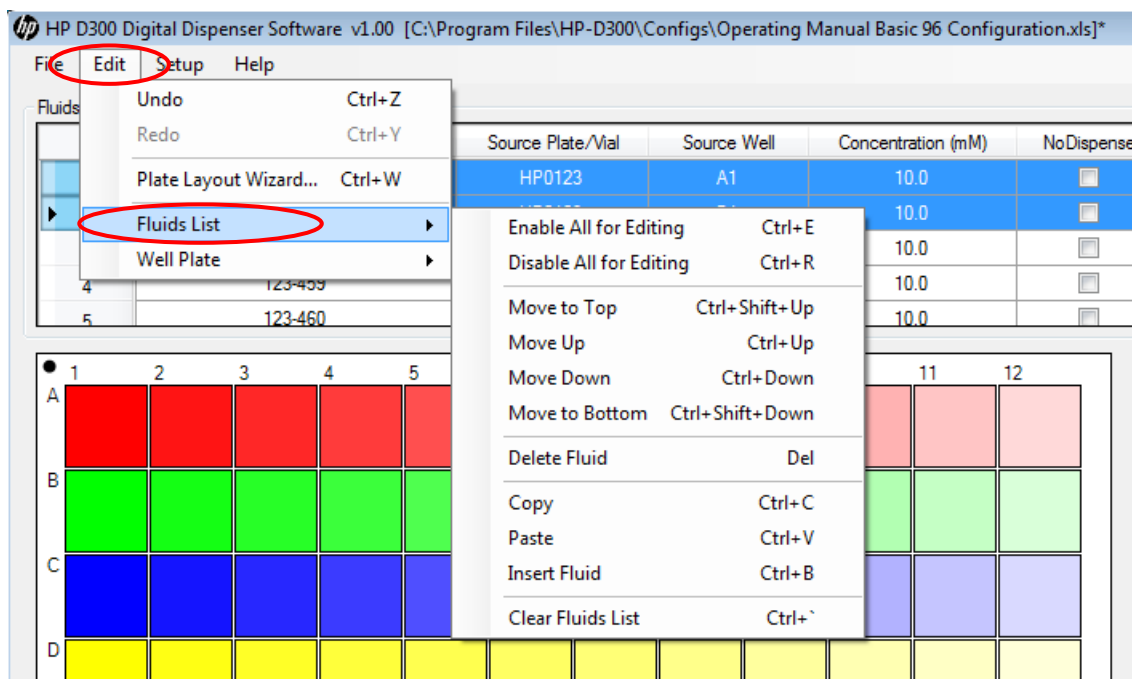
5-2 Edit the Fluids List

In addition to typing directly into the Fluids List table as described in “[Create the fluids table](#)”, the Fluids List can also be edited by clicking **Edit** and then **Fluids List**.

1. Select one or more fluids you would like to edit by clicking on the row number (in the left-most column in the Fluids List). To select multiple fluids, hold down the CTRL key while clicking on the row number.
2. Click **Edit** and then **Fluids List**.
 - Select **Enable All for Editing** or **Disable All for Editing** to change all fluids to be shown or not shown in the well plate image, and able to be added or not to the plate layout.
 - Select **Move to Top**, **Move Up**, **Move Down**, or **Move to Bottom** to change the order in which fluids are dispensed into the well plate.
 - Select **Delete Fluid** to remove the selected fluid from the table.
 - Select **Copy**, **Paste**, or **Insert Fluid** to copy, paste, or insert a fluid into the table.
 - Select **Clear Fluids List** to remove all fluids from the Fluids List.

NOTE: These options are also available by right-clicking on selected fluids in the Fluids List.

Figure 5-2 Edit the Fluids List (or right click within the Fluids Table)



5-3 Edit the well plate

Once the fluid table is set up with at least one compound, Edit Well Plate operations such as adding fluids to the wells may begin. To see the Edit Well Plate menu options, click on Edit/Well Plate or select some wells in the well plate image and right-click. As shown in **Figure 5-3**, the following options are available:

- Select or Deselect All Wells
- Set Titration (see “**Set concentrations or volumes in wells**”, step 3)
- Set Concentration or Set Volume (see “**Set concentrations or volumes in wells**”, Step 1)
- Normalize (see “**Normalize**”)
- Randomize (see “**Randomize**”)
- Remove a single fluid and remove all fluids
- Cut, Copy, Paste, and Paste Special (see “**Use the Paste Special dialog box**”)
- Clear Well Plate
- Plate Layout Wizard (see “**Plate Layout Wizard basics**”)


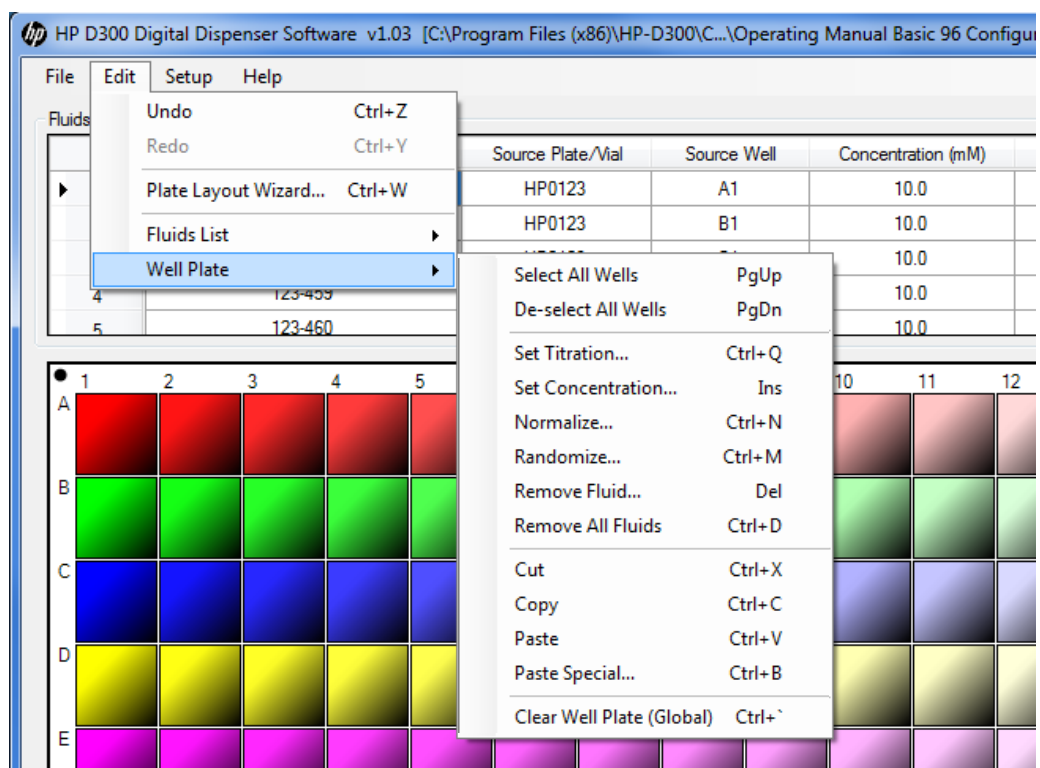
 **TIP:** This menu can also be accessed by clicking **Edit** and then **Well Plate** after selecting a well or wells in the well plate image.

Figure 5-3 Edit Well Plate menu



Each well in the well plate can contain multiple fluids. For example, two fluids of interest in a drug-drug interaction experiment can each be added to the desired wells in the desired sequence of concentrations or volumes, then a third fluid can be added to the same wells to normalize the wells to a constant DMSO level. Each fluid is added to the selected wells in a separate programmatic step.

TIP: All of the fluids are shown in the Global View, whereas only the fluids marked for editing (by selecting the **Edit** check box in the Fluids List) are shown in the well plate image. See [Figure 5-4](#).

TIP: If a well does not contain any fluid, it has a dotted outline. The dotted outline is also used to denote a well that exceeds a dispense condition, such as containing more fluids than the DMSO limit, or having a requested volume of less than one 13pL drop.

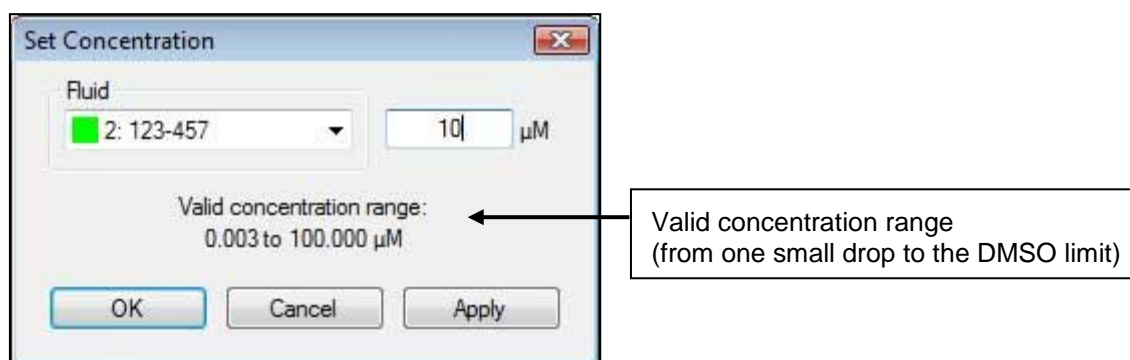
Figure 5-4 The difference between Global View and the well plate image



5-4 Set concentrations or volumes in wells

1. Click and drag in the well plate image to select the wells where a fluid will be dispensed.
2. To add the same single concentration or volume of fluid to the selected wells, right-click anywhere in the well plate image and then click **Set Concentration** or **Set Volume**. The Set Concentration or Set Volume dialog box appears. See [Figure 5-5](#). Whether Set Concentration or Set Volume is displayed is determined by the setting chosen in the Edit: Units pane of the Options dialog box as described in “[Options: Edit: Units](#)”.
 - Select a fluid from the Fluid drop-down list box and then type in a concentration or volume that is within the valid range that is stated in the dialog box. See [Figure 5-5](#), shown for concentration mode.

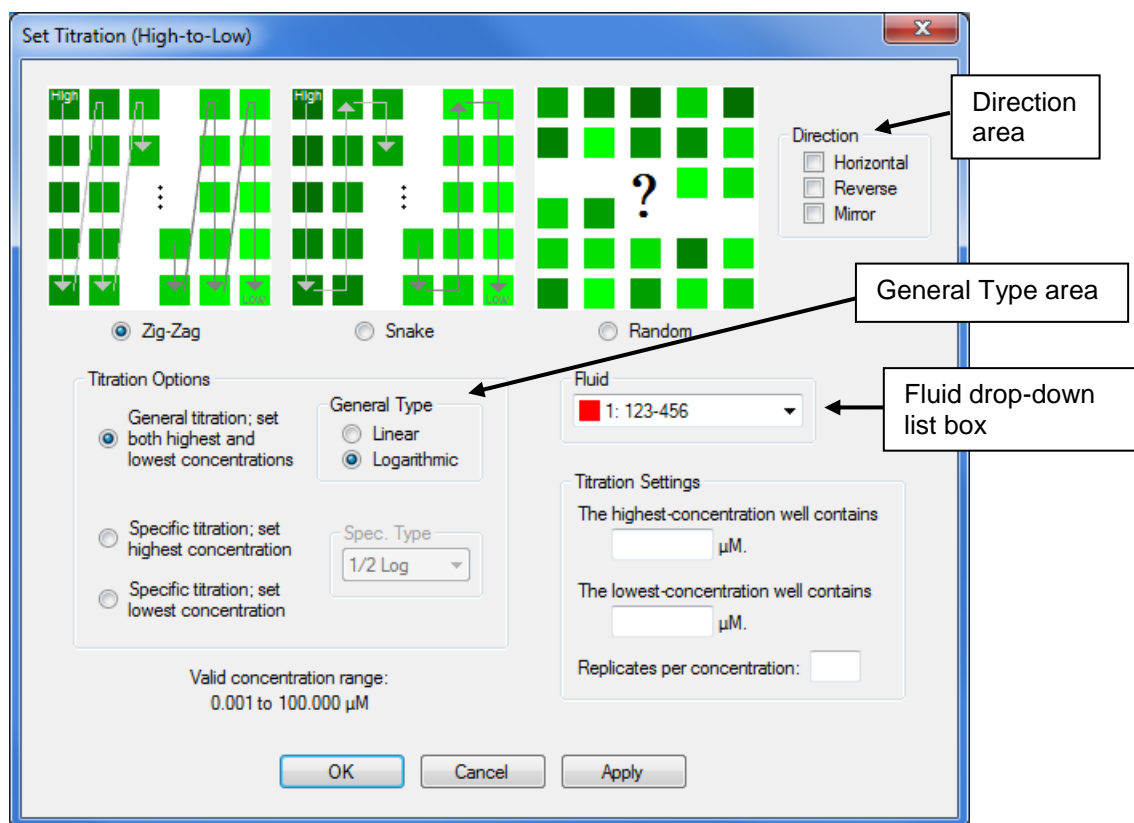
Figure 5-5 Set Concentration dialog box



NOTE: The lowest possible concentration or volume is determined by the single drop volume, the fluid concentration, and the assay volume. The maximum concentration or volume is determined by the DMSO limit and the fluid concentration.

3. To generate a sequence of concentrations or volumes in the selected wells (a titration series, with different concentrations and volumes in each well), right-click anywhere in the well plate image and click **Set Titration**. The Set Titration dialog box opens (see [Figure 5-6](#)). Whether concentration or volume is used in this dialog depends on the setting chosen in the Edit: Units pane of the Options dialog box (see [Figure 4-12](#)).
 - a. Select Zig-Zag, Snake or Random:
 - Zig-Zag builds the titration one row or column at a time (depending on whether Horizontal is selected in the Direction area), then starts the next row or column in a well adjacent to the first well.
 - Snake builds the titration continuously, so that every well of the titration is adjacent to the wells closest in concentration or volume.
 - Random takes the set of concentrations or volumes and distributes them randomly among the selected wells. Randomization of the entire plate can also be accomplished later, using the Randomize function as described in the “[Randomize](#)” section.
 - b. Select the direction that the titration will be built. These choices are not available for random titrations. Note that the arrows and shadings of the graphic in this window change as you make different selections to help you understand the options.
 - Horizontal: This option creates doses from left to right. If this box is not checked, the default is a vertical dose order (top to bottom).
 - Reverse: This option reverses the concentration or volume order from high-to-low to low-to-high. If this box is not checked, the default is a high-to-low titration.
 - Mirror: This option flips the titration either horizontally or vertically depending on whether the **Horizontal** check box is selected. The easiest way to understand this is to select the **Mirror** check box and observe what happens to the displayed graphic.
 - c. Select the fluid to be programmed in the Fluid drop-down list box.
 - d. Select the type of concentration or volume titration in the General Type area of the dialog box. Select either **Linear** or **Logarithmic**.
 - Linear: The titration steps are evenly spaced in concentration or volume units
 - Logarithmic: The titration steps are evenly spaced in log concentration or log volume units

Figure 5-6 Set Titration dialog box

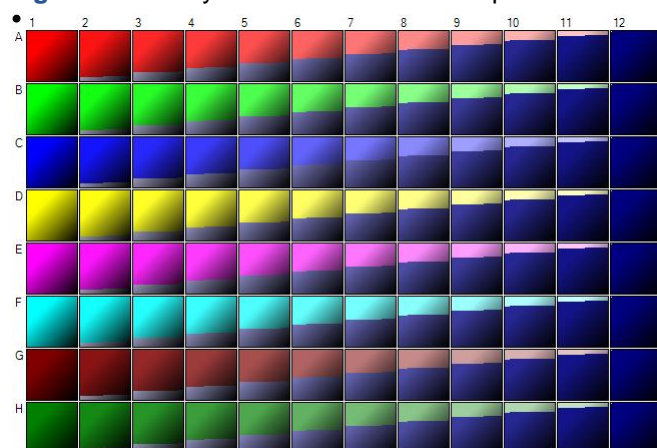


- e. Select the type of titration to use in the Titration Options area of the dialog box. Choose from the following options:
 - General titration: you set both lowest and highest concentrations or volumes and let the middle concentrations or volumes fall along either a linear or log scale (as selected)
 - Specific titration: you set the highest or lowest concentrations or volumes and select one of the following specific types of concentration or volume spacing:
 - 1/2 log
 - 1/3 log
 - 1/4 log
 - 1:1 (50% dilution)
 - 1:2 (33% dilution)
 - 1:3 (25% dilution)
- f. Type in the highest and/or lowest well concentration or volume within the valid concentration range shown in the lower-left portion of the **Set Titration** dialog box.
- g. Type the number of replicates at each well concentration or volume level in the Titration Definition area of the dialog box, for example type “3” for a triplicate titration.

NOTE: The lowest possible well concentration is determined by the minimum drop volume, the fluid concentration, and the assay volume. The maximum well concentration is determined by the DMSO limit and the fluid concentration.

4. Additional fluids can be added to the same wells using the same steps as above. If multiple fluids are in a single well, the well will have two or more colors in relative proportion to represent all the fluids that will be dispensed into the well as shown in [Figure 5-7](#).

Figure 5-7 Array of wells with two fluids per well



5. To easily replicate a plate design, you can select all the wells in a plate you have designed, then right click and select **Copy**, then click on **Add Plate**. Select the A1 well of the new plate, then right click and select either **Paste** or **Paste Special** depending on whether you want to have the same fluids in both plates. For more information about Paste Special, see “[Use the Paste Special dialog box](#)” below.

5-5 Use the Paste Special dialog box

Instead of defining titrations separately for each fluid as described above, titrations can be copied and pasted with new fluids once a starting titration is created. If you would like your plate to contain the same titration for many different fluids, you can use “[Plate Layout Wizard basics](#)” or you can use Paste Special. For Paste Special, follow these steps:

1. Program the initial titration, then ensure the wells you want to copy are selected, then right-click in the well plate image and then click **Copy**.
2. Click in a new well to select the new starting well for the Paste Special, then right-click, and do one of the following:
 - To paste the same fluid into new wells, click **Paste**.
 - To paste a different fluid into new wells, click **Paste Special**. This opens the Paste Special dialog box where you can accept the suggested fluid to paste or use the drop-down list box to select a different new fluid to paste. See [Figure 5-8](#).

In the example shown below, wells A1 to B12 were copied, and fluid 1 will be mapped to fluid 3 and fluid 2 mapped to fluid 4. The result of this Paste Special operation is shown in [Figure 5-9](#).

Figure 5-8 Paste Special dialog box

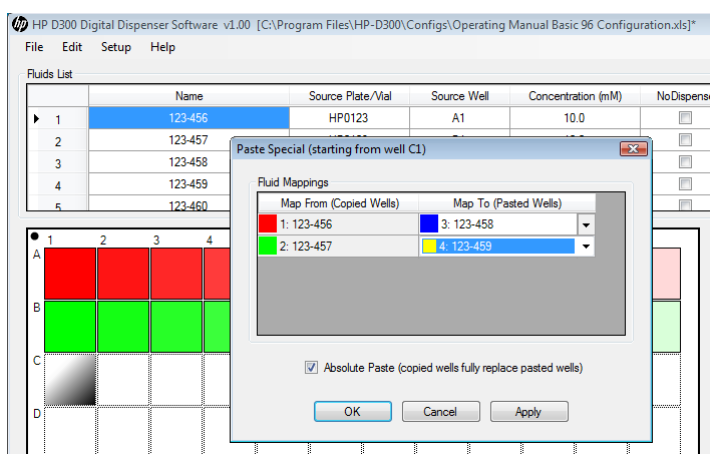
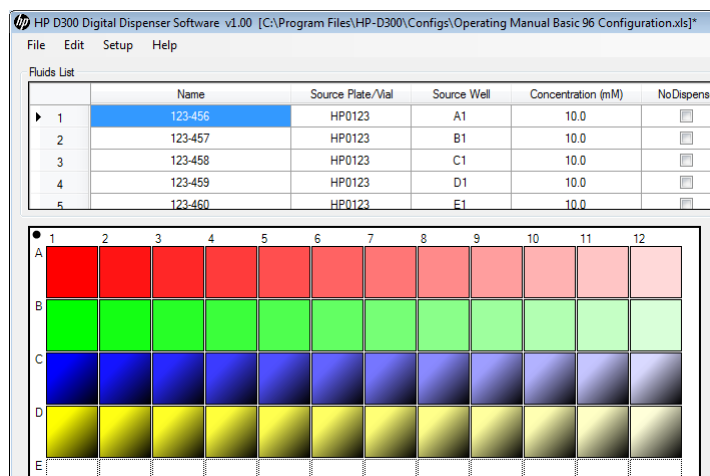


Figure 5-9 Result of Paste Special operation shown in Figure 5-8



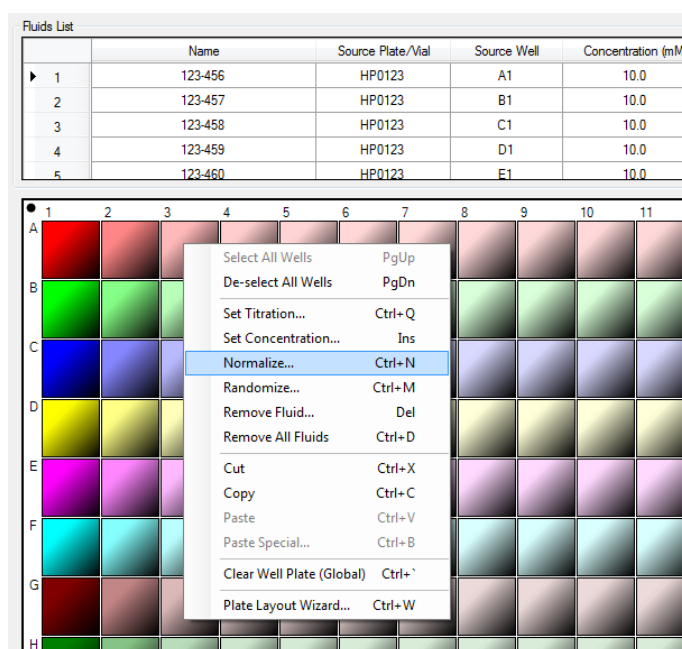
3. Repeat steps 1 and 2 for each fluid until all desired wells on the plate are designed.

5-6 Normalize

Once the plate has been fully configured with the fluids that will be dispensed, a DMSO normalization can be programmed.

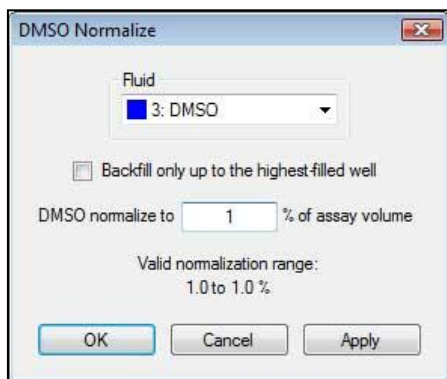
1. Ensure that the **Fluids List** contains the normalization fluid, for example a fluid named DMSO.
2. Click and drag in the well plate image to select wells to normalize (using the control key if you need to select more than one region because some areas of the well plate do not need normalization), then right-click to open the Edit Well Plate menu, and then click **Normalize**. The DMSO Normalize dialog box appears (see [Figure 5-11](#)).

Figure 5-10 Select **Normalize** in the Edit Well Plate menu




3. In the Fluid drop-down list box, select a fluid to use for normalization.

Figure 5-11 DMSO Normalize dialog box



4. Type a value in the box following “**DMSO normalize to**”, or you can choose to normalize up to the highest % DMSO already programmed into any of the selected wells, by selecting the **Backfill only up to the highest-filled well option**.

The maximum normalization range is determined by the DMSO Limit % entered for the selected fluid in the Fluids List. The minimum normalization range is determined by maximum amount of DMSO already present in the selected wells. You can enter a normalization percentage below the minimum valid range; see the note below.

 **TIP:** You **may** enter a normalization percentage that is less than the valid normalization range, however all wells will not be normalized to the same DMSO percentage. For example, entering 0.1% DMSO normalization will only add DMSO to wells which are less than 0.1% DMSO, and any wells which are greater than 0.1% DMSO will have no additional DMSO added to them.


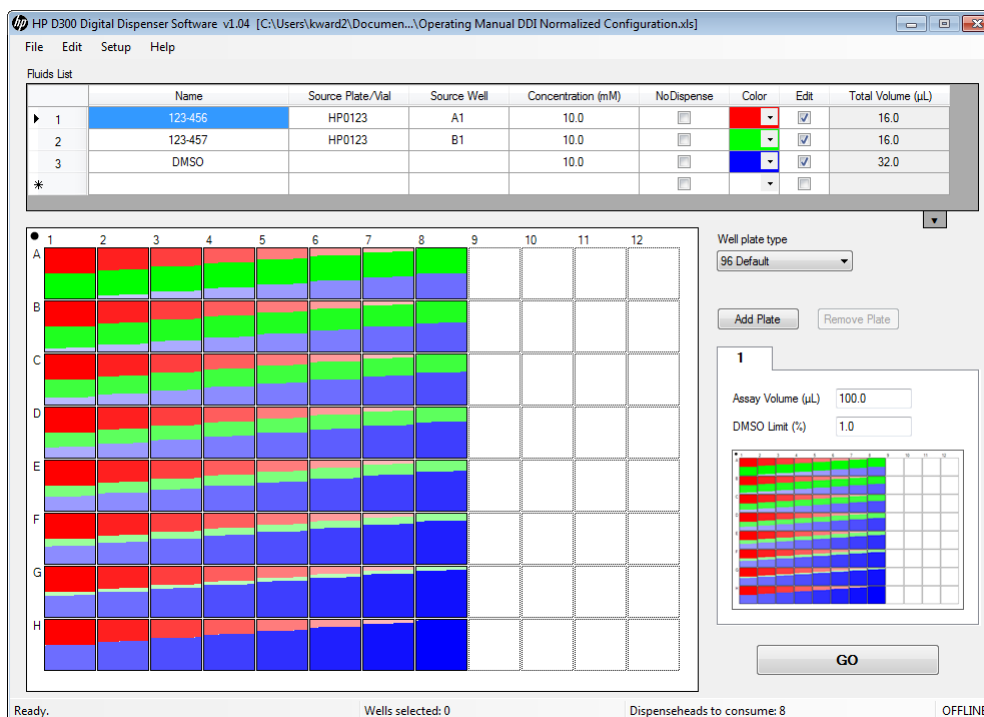
 **TIP:** To ensure proper DMSO normalization volumes, perform the DMSO normalization function after configuring the wells with all fluids. To add another fluid to wells which already had the normalization operation applied to them, delete the DMSO normalization fluid from those wells by selecting the appropriate wells, right click and select **Remove Fluid**, then pick the DMSO from the fluid drop down. After removing normalization, add the new fluid in the appropriate wells before repeating the DMSO normalization step.

Figure 5-12 Result of using fluid 3 for DMSO Normalization




NOTE: When DMSO is added to the Fluids List, the default concentration is often left at 10mM. Since this is not the concentration of neat DMSO, this means that the well pop-up concentrations (as shown in [Figure 4-11](#)) for DMSO in normalized wells will be incorrect. For DMSO, enter the source fluid concentration as 14100 mM, or ignore the concentration part of the well pop-up. Either way, the percentages and volumes for DMSO shown in the well pop-ups are correct.

5-7 Randomize

Once the plate has been fully configured, the wells can be reordered randomly on the plate. This is a useful way to minimize the impact of systematic plate edge effects on the output data.

1. In the well plate image, select the wells that you want to randomize. To omit areas of the plate from randomization, use the CTRL key to select multiple noncontiguous regions. To deselect single wells, hold down the CTRL key while clicking on the well.
2. Right-click to open the Edit Well Plate menu and select **Randomize**. See [Figure 5-13](#).

 **TIP:** To return to the non-randomized, ordered plate, click **Edit** and select **Undo**.

3. Only the selected wells are randomized.

Figure 5-13 Right-click and select **Randomize**

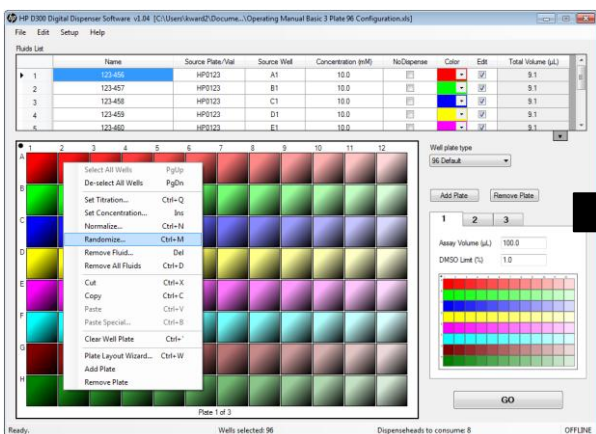
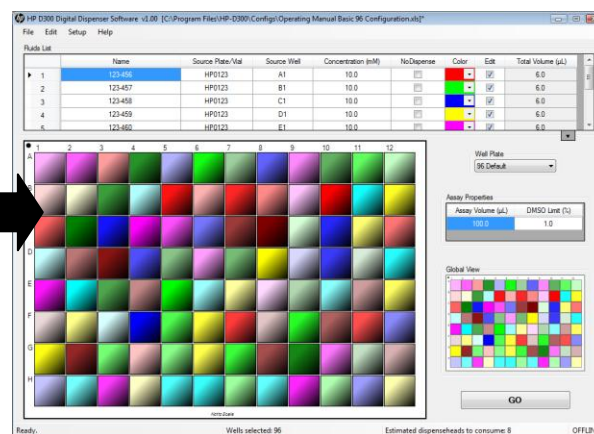


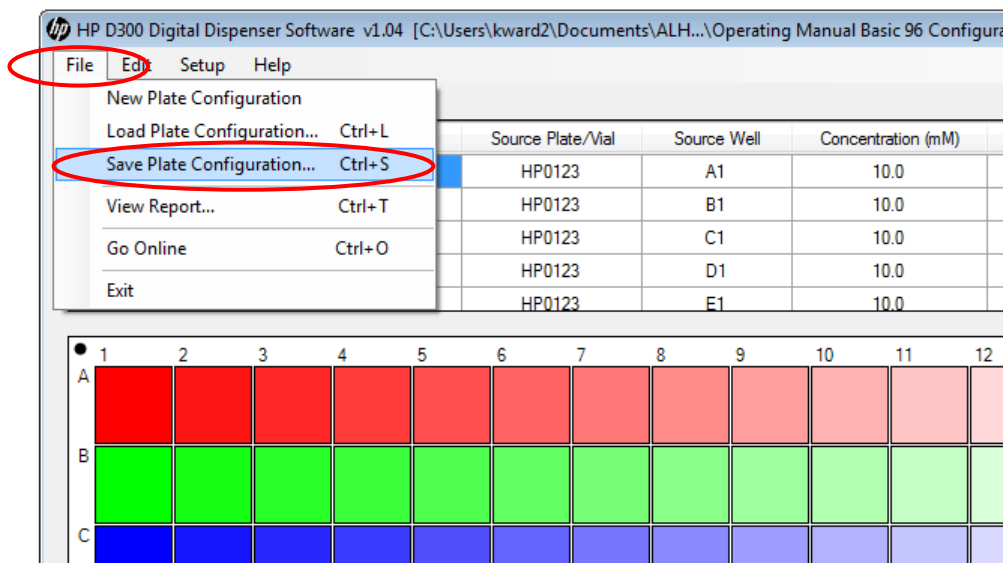
Figure 5-14 Example of a randomized well plate



5-8 Save the plate configuration

1. On the HP D300 Digital Dispenser software main operating window, click **File** and then **Save Plate Configuration**. All plates will be saved in a single file.
2. Name the new plate configuration. The plate configurations are saved as Excel files. They can be edited or even created in Excel, but be extremely careful because the format must exactly match what is expected by the HP D300 Digital Dispenser software.

Figure 5-15 To save the plate configuration, click **File** and **Save Plate Configuration**



5-9 Plate Layout Wizard basics

The Plate Layout Wizard may be used for several purposes, both simple and complex. Complex examples are described in “[Plate Layout Wizard advanced functions](#)”. In the simple case, you can replicate a single titration (a set of different concentrations or volumes) for many or all of a set of fluids. For example if you would like each row of a 384 plate to be an 8 point triplicate, and use those concentrations for all 16 rows of the plate for 16 different compounds, the Wizard can create this configuration much more quickly than a series of copy and paste special commands. This simple case is described here in section 5-9-1, it is a “single zone titration 1” because all the concentrations in the titration are evenly spaced in linear or log scale (known as a “single zone”) and because each well only contains a single compound (known as a “titration 2”).

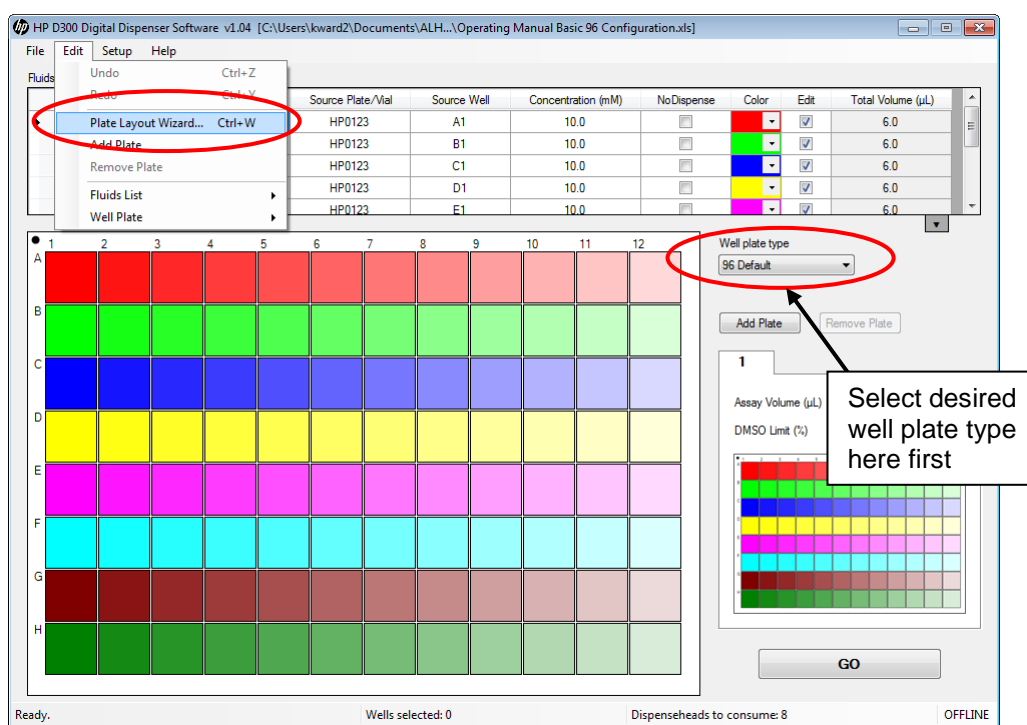
5-9-1 Create single zone titration1 layouts

Ensure that the desired well plate type, either 96 or 384, is selected using the Well Plate drop down menu prior to starting the Plate Layout Wizard.

1. In the HP D300 Digital Dispenser main operating window, click **Edit** and then **Plate Layout Wizard**. The Plate Layout Wizard: Initialization dialog box appears. See [Figure 5-16](#).

The Plate Layout Wizard can also be accessed by right-clicking on any well in the well plate image and then selecting the Wizard option in the Well Plate menu.

Figure 5-16 Click **Edit** and then **Plate Layout Wizard**



2. If there were existing entries in the Fluids List or fluids in any wells in the well plate, the following statement appears in the dialog box:

“Some fluid and/or well data already exists. How would you like to proceed?”

- a. Select one of the three options:
 - To use the wizard to add fluid to empty wells remaining in the current plate configuration only, select **Leave all existing data alone**.
 - To retain the fluids from the Fluids List and design a new plate configuration, select **Clear the well plate only; leave the fluids alone**.

- To start from a new Fluids List to design a new plate configuration, select **Clear all data and start from scratch (no fluids, empty well plate)**.

Figure 5-17 Plate Layout Wizard: Initialization dialog box

Welcome to the Plate Layout Wizard! The following steps will guide you in creating a plate layout that is suitable for your experiment. You will be able to customize single- and double-fluid sequences (sets of volume or concentration points) and position them appropriately within the well plate. After completing this process, if the results aren't quite what you want, you can continue to set up your plate layout using the normal interface.

But first, please answer some initial questions.

Some fluid and/or well data already exists. How would you like to proceed?

- ☐ Leave all existing data alone
- ☐ Clear the well plate only; leave the fluids alone
- ☒ Clear all data and start from scratch (no fluids, empty well plate)

What is the assay volume of each well you will be using?

100.0 μL

What is the DMSO limit, as a percentage of the assay volume, f

1.0 %

How many fluids will you be using, including NoDispenses?

4 fluids and NoDispenses

Enter the name, source, concentration, and water content of each of the fluids, and specify which ones are NoDispenses
 * (You can leave any/all source fields blank; however, the information may be useful later during dispense)

Name	Source Plate/Vial	Source Well	Concentration (mM)	NoDispense
123-456			10.0	<input type="checkbox"/>
123-457			10.0	<input type="checkbox"/>
123-458			10.0	<input type="checkbox"/>
123-459			10.0	<input type="checkbox"/>

10.0 mM → Use this concentration for all fluids

Continue Cancel

Annotations:

- Select one of these three options (points to the radio button options)
- Answer these questions unless you chose to leave all existing data (points to the assay volume and DMSO limit fields)
- Select at least one NoDispense to allow placement of controls (points to the NoDispense checkbox in the table)
- Type fluid information in this area if desired (points to the table)
- Change concentration here if desired (points to the concentration field and button)

- If you selected the first option, the next two questions involving assay volume and the DMSO limit will be unavailable because the previous values are used. If you selected the second or third option, type the assay volume and DMSO limit for the new plate design into the boxes and press the ENTER key on the keyboard.
- The next question involving the number of fluids you are using also changes depending on which option was selected above. Read the question carefully, type an answer into the box, and press the ENTER key.
- Click in the first column of the table to edit the names of the additional fluids, if desired, and then type the source plate/vial, source well, and concentration information into the following columns.
- Select the **NoDispense** check box if the fluid should be a no dispense fluid (fluids are which are non-HP dispensed fluids). There needs to be at least one fluid with **NoDispense** checked in order to reserve rows or columns for controls which are not dispensed by the HP dispenser.
- To apply a concentration other than the default, type the appropriate value in the box at the bottom of the Initialization dialog box and click the **Use this concentration for all fluids** button to apply the new value to all of the new fluids.
- Click **Continue** to move to the next step. The Plate Layout Wizard: Titrations and Positioning dialog box appears. See **Figure 5-18**.

! IMPORTANT: Once titrations and positioning are initiated, changes cannot be made to the fluids table because changes could disrupt the design. To make changes to the fluids table, click **Cancel**, then make changes to the fluids table before launching the Plate Layout Wizard again.

3. Notice the left pane of the Plate Layout Wizard: Titrations and Positioning dialog box.

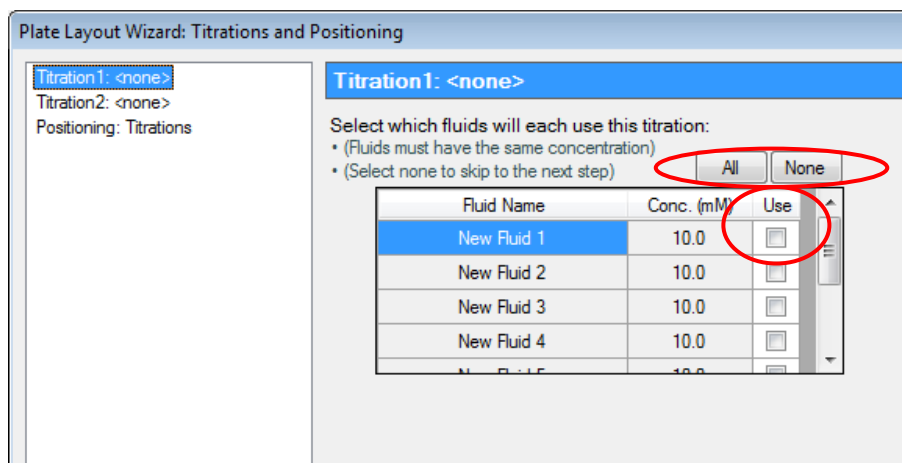
Recall that a titration is simply a set of concentrations or volumes that form a titration. Two types of titrations can be programmed:

- Titration1 is denoted as **Titration1** in the menu in the left pane, and is used for titration series with one fluid per well.
- Titration2 is denoted as **Titration2** and is used for titration series with two fluids per well.

Plate designs can have zero or many titration1s and/or titration2s on a plate. Each titration1 can use from one to all of the fluids in the fluids table. If all fluids in the plate will have the same set of concentrations or volumes, only one titration1 is required. Additional titration1s are only needed in order to use a different set of concentrations or volumes for one or more fluids.

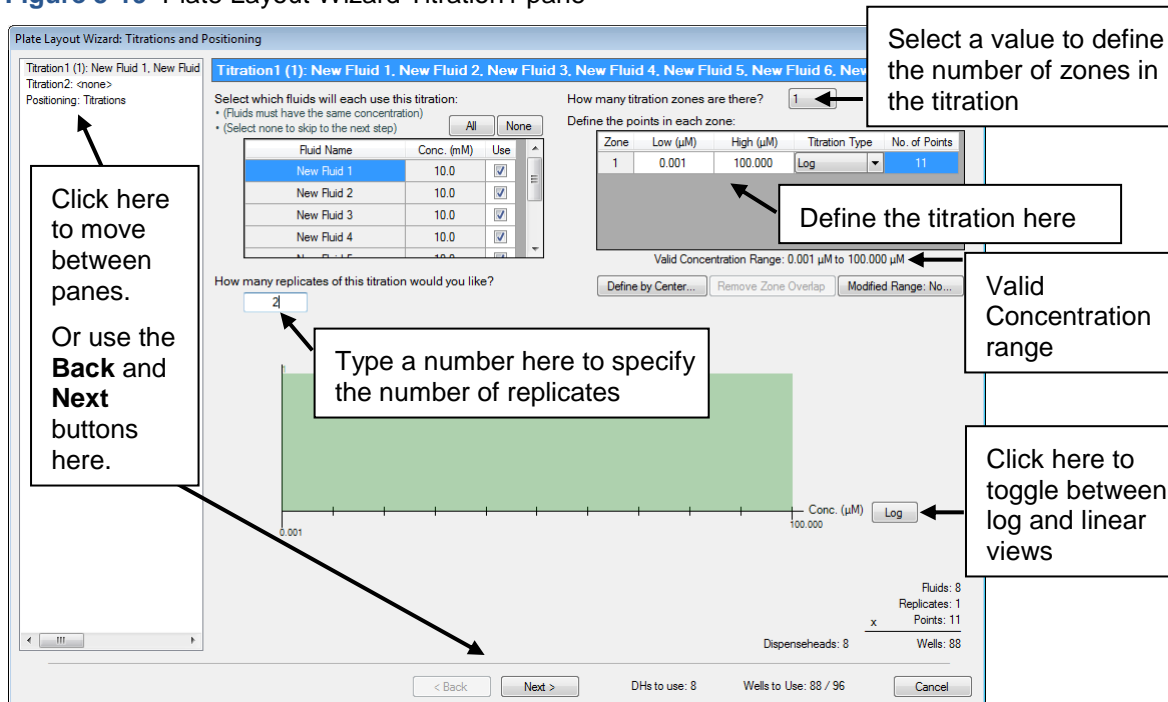
- a. By default, designs start with the definition of any titration1 fluids. If the desired plate design involves only titration2 fluids, click **Titration2: <none>** in the left pane, or click the **Next** button to move down the list to the next item. Click the **Back** and **Next** buttons to go up or down the list as desired.

Figure 5-18 Plate Layout Wizard Titration1 fluid selection pane



- b. Select the **Use** check box next to each fluid that will use this titration to create a titration1 that will be used by one or more fluids. Click on the **All** button to use all fluids in the fluids table with the same titration. Select **None** to uncheck all of use boxes at the same time. To use all fluids except for a few, select **All**, then click on the **Use** check boxes to uncheck just those few fluids to not use. When one or more **Use** checkboxes are selected, the appearance of the dialog box changes as shown below in **Figure 5-19**.

Figure 5-19 Plate Layout Wizard Titration1 pane



c. Define the titration to be used for the selected fluids:

- Type a number in the box to specify how many replicates of each concentration or volume will be used for each fluid as you did in the “**Set concentrations or volumes in wells**” section of this document. For example, if triplicates are desired for each fluid, enter **3**.
- Click the arrow in the drop-down list box and select a value to define how many titration zones the titration will have.

Titration zones are regions (concentration ranges) of the titration which have the same type of spacing between concentrations.

If a titration is desired to have tight spacing around the expected IC50 value, with more sparsely spaced concentrations on either side of the IC50, a three-zone titration could be chosen. However let's practice with a simple one zone titration for eight fluids, four from the existing fluids table, and four new fluids. The eight fluids have already been selected with the Use check boxes.

- A twelve-point singlet will be defined for each of the eight fluids. Click in the Low (μM) and High (μM) columns of the table to enter the concentration endpoints of the titration, then click in the Titration Type column to define how the points are spaced between the low and the high concentration limits, and then type in the number of points. If the titration type is not Linear or Log, then the spacing between concentration points is fixed, so the number of points, the high and low limits are adjusted by the software to make a valid titration series when a fixed spacing is specified.
- The Valid Concentration Range specifies the concentration limits for these. The lowest possible well concentration is determined by the minimum drop volume, the source fluid concentration, and the assay volume. The maximum well concentration is determined by the DMSO limit and the fluid concentration.
- At the bottom of the dialog box, the endpoints and the point spacing are shown graphically. Click the box labeled **Log** to toggle between log and linear views of the layout.

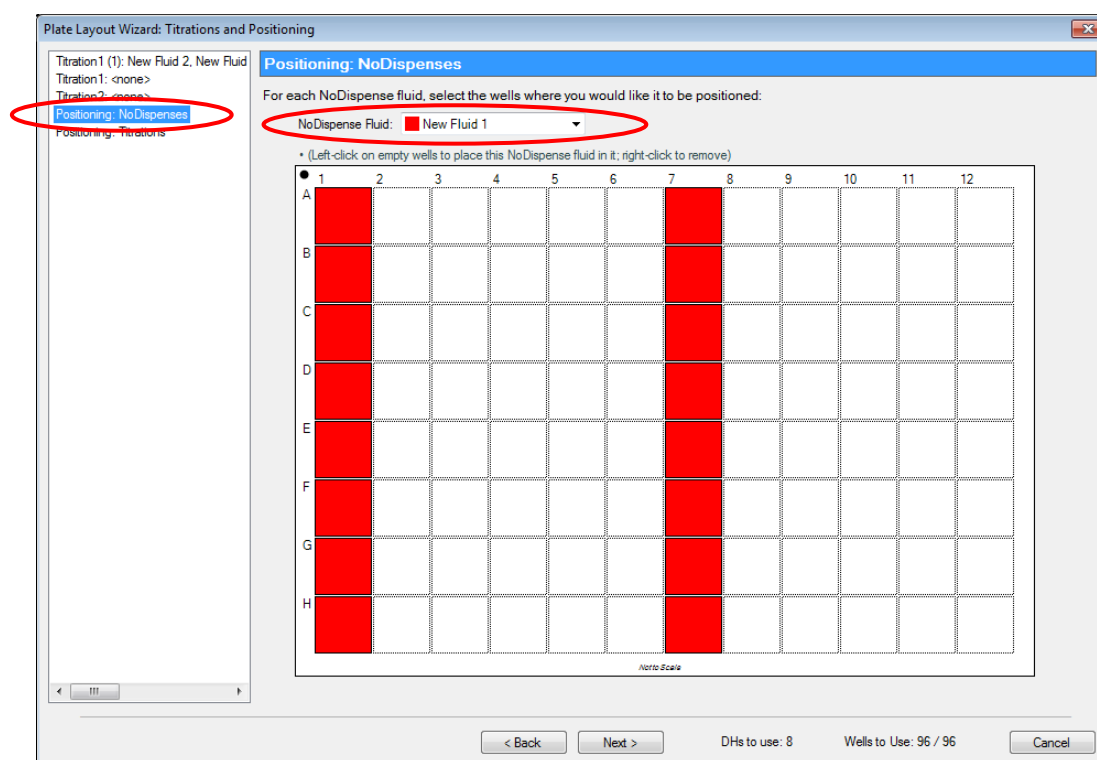
- Click **Next** three times or click **Positioning: Titrations** to open the Positioning: Titrations pane. Only a single titration1 and no titration2's will be used for this example. The twelve point singlet for each of the eight fluids selected fills up the entire 96 wells.

5-9-2 Position NoDispense controls

The use of NoDispense controls or blanks were described above in “**Position NoDispense controls**”.

1. If any fluids selected in the Wizard were designated as “NoDispense” fluids, this option will appear. Click on the “**Positioning: NoDisperses**” in the left-hand menu. The window that will appear is shown in **Figure 5-20**.
2. Select which of the fluids you initially designated as NoDispense fluids you would like to position in the well plate first, using the drop down menu titled “**NoDispense Fluid**”.
3. Left-click and drag to designate wells for this NoDispense fluid. This Wizard window works slightly differently than the main well plate image in that if you select any wells for this NoDispense fluid by mistake, you have to right-click those wells to unselect them.
4. Repeat steps 2 and 3 for each NoDispense fluid you wish to position. Click on Next when finished, or select “**Positioning: Titrations**” from the left-hand menu.

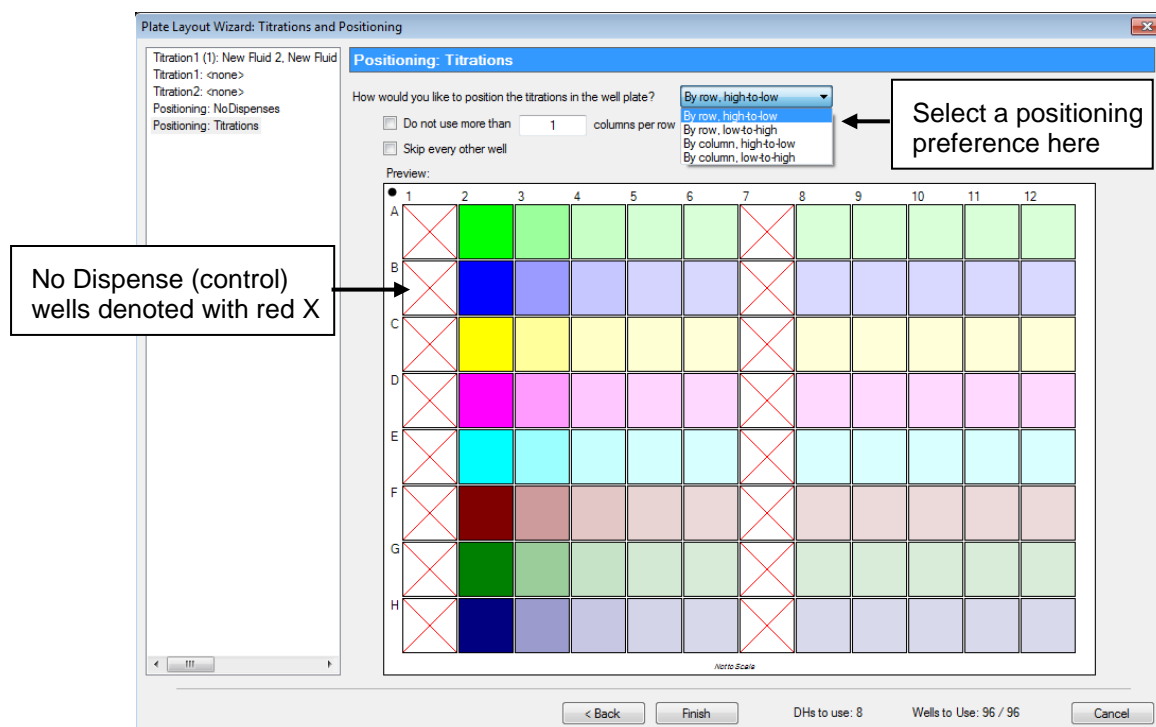
Figure 5-20 Positioning: NoDisperses pane



5-9-3 Position a single zone titration1

1. Click the arrow on the drop-down list box to select one of the four positioning preferences that the 96 points can be placed in. **Figure 5-21** shows the layout by rows with the concentrations in high to low order across the rows.

Figure 5-21 Positioning: Titrations pane



- Select the **Do not use more than ____ columns per row** check box and type in a number to limit titrations which span more than one row or column.
- Select the **Skip every other well** check box to generate an interleaved well plate.
- Once the titrations are positioned properly, click **Finish** to exit the Plate Layout Wizard. More complex designs are described in the following section.

5-10 Plate Layout Wizard advanced functions

In addition to simple full plate layouts using a single titration for all fluids as described above in “[Plate Layout Wizard basics](#)”, the Plate Layout Wizard can be used to create complex plate designs that would be difficult to create using the standard graphical user interface described above.

Three types of advanced configurations are described in the following sections. The first of these is where the titration for a particular compound is not uniformly spaced in log or linear concentrations, which we call a multizone titration 1 layout. Recall that titration 1 refers to a titration with one compound per well. For example, the ends of a particular dose response function may show little change in response over a large range of concentrations, so that these areas do not need to be sampled with as many different doses, while the region near the IC50 or EC50 point where the response is rapidly changing with concentration could be sampled with a dense series of closely spaced doses. This would of course require some knowledge of the IC50 or EC50 prior to setting up the configuration. The process for creating these configurations is described in “[Create multizone titration1 layouts](#)”.

The next type of complex layout is where in some cases the dynamic range of the dispenser (the range between the dose from one 13pL droplet to the DMSO limit) is too small for your region of interest. By specifying your range of interest, the Wizard will help you to determine the fluid pre-dilution required to achieve the desired dynamic range and set up the dispenser to use two stock concentrations to achieve greater dynamic range. This type of configuration is described in “[Create modified range titration1 layouts](#)”.

The final type of advanced designs are drug-drug interaction or synergy experiments where two compounds are placed in the same well for many wells in a set of orthogonal concentrations. We call this a titration 2 layout since there are two compounds in each well, and is described in “[Create titration2 layouts: two fluids per well](#)”. All of these advanced functions take advantage of the digital nature of your HP D300 Digital Dispenser which allows “any dose in any well”.

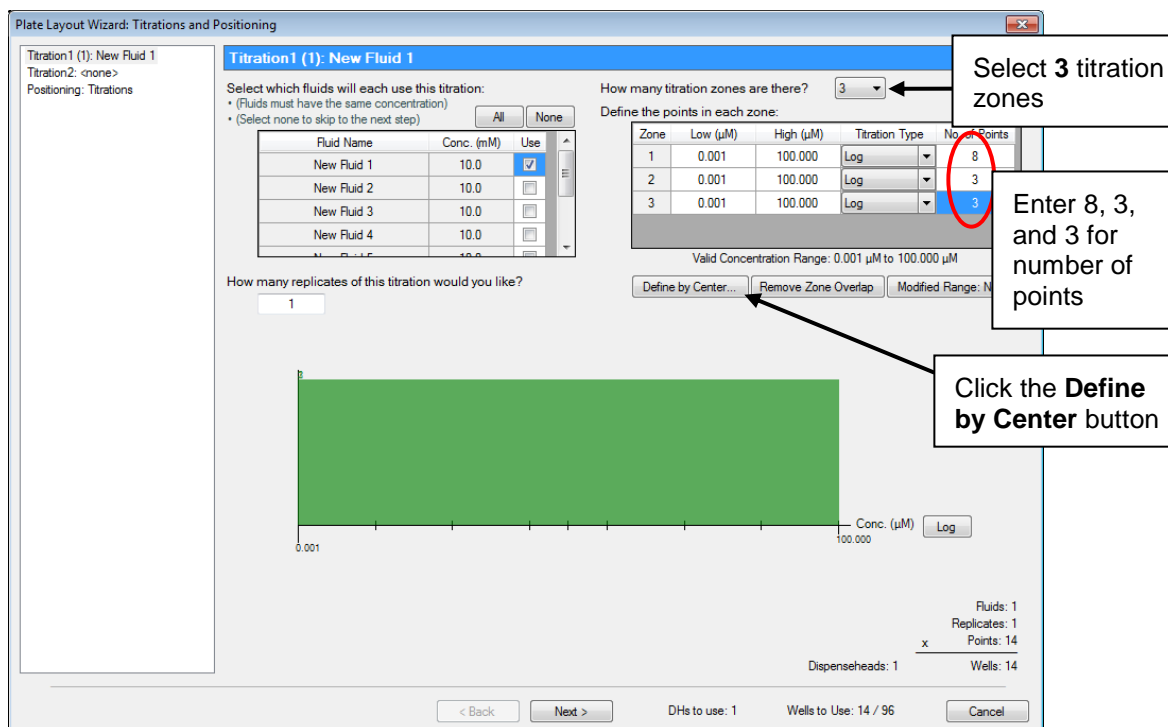
5-10-1 Create multizone titration1 layouts

A multizone titration 1 is a set of concentrations or volumes that is not evenly spaced for the entire fluid. For example, you may wish to create a titration which has more densely spaced points close to a known or targeted IC50 value, with less densely spaced points further away from IC50 where there is little change in the response value.

1. To generate a multi-zone titration1, follow steps 1 and 2 in [5-9-1](#) above, then click **Titration1** in the left pane of the Plate Layout Wizard: Titrations and Positioning dialog box. This opens the Titration1 pane.
2. Select the fluid(s) you would like to use for this multizone titration by clicking in the appropriate **Use** box for each desired fluid. You likely will only program a single fluid at a time using this method since you will center the region of densely spaced doses differently for each compound.
3. Define a three-zone titration with a tightly spaced zone close to the presumed IC50 of the compound, and two sparsely spaced zones on either side of the IC50 (see [Figure 5-22](#)).
 - a. Click the arrow on the **How many titration zones are there?** drop-down list box and select **3**.

Assume that four wells will be used for the sparsely spaced zones, and eight wells will be used for the fine spaced zone close to the IC50 value for fluid 1.
 - b. In the No. of Points column for zone 1, enter **8**, and for zone 2 and zone 3, enter **5**. Below, you will see how this titration which now looks like an 18 point singlet will become a 16 point singlet.

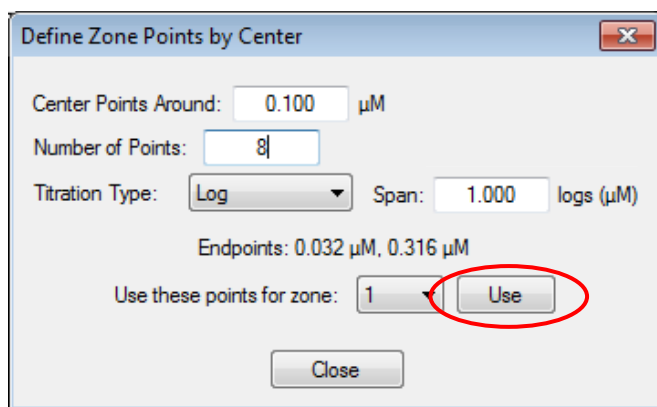
Figure 5-22 Titration1 pane—creating a multizone titration1 layout



- c. Define the densely spaced doses in the center of the titration as zone 1:

Click the **Define by Center** button. The Define Zone Points by Center dialog box appears. Ensure that the Use these points for zone: drop down is set to “1”), as shown in **Figure 5-23**.

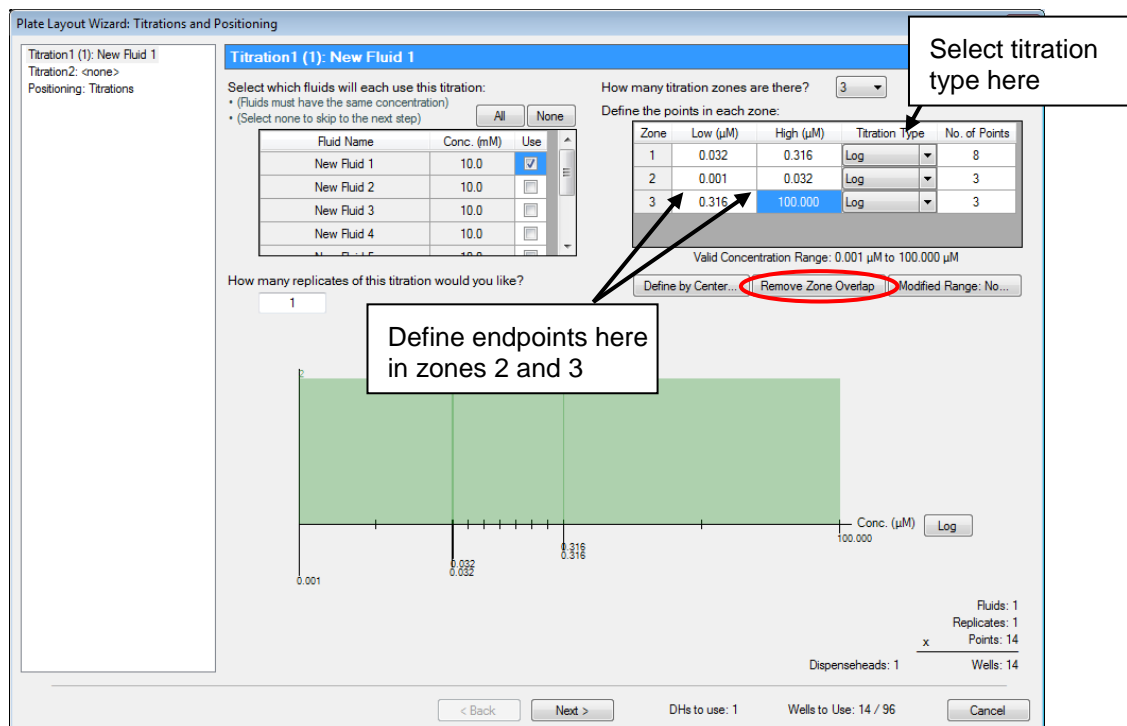
Figure 5-23 Define Zone Points by Center dialog box



- i. Enter the center point concentration for the finely spaced region in the **Center Points Around** box.
For example, the presumed IC50 value might be used. A value of 100 nM has been chosen for this example.
- ii. Enter the number of points for the finely spaced region in the Number of Points box.
- iii. Select the titration type to be used for the set of points by clicking the arrow on the Titration Type drop-down list box.
- iv. Enter a value in the Span box for this zone; if a span of 1 log is entered, the points will span plus and minus a half log around the center point.

- v. Notice that the endpoints for this zone change as values are entered and that zone location within the titration range and point spacings are shown on the green graph on the Titration1 pane.
 - vi. Click the **Use** button as shown in [Figure 5-23](#).
 - vii. Click the red **X** at the top-right corner to exit this window when finished with centered zones.
- d. Now define the sparsely spaced zones 2 and 3.
- i. Use the endpoints of the centered zone 1 and the valid concentration range to define the endpoints for zones 1 and 3. Enter the appropriate values in the Low (μM) and High (μM) columns for zones 1 and 3 as shown in [Figure 5-24](#).
 - ii. Select the Titration Type for zones 1 and 3.

Figure 5-24 Titration1 pane—creating a multizone titration1 layout, part 2



- iii. Click on the **Remove Zone Overlap** button since the three zones have some overlapping concentrations. Removing the zone overlap will delete points from the titration, which is why the number of points for zones 2 and 3 was set to 5, as shown in [Figure 5-24](#). Note that the two zones that have points overlapping with zone 1 are reduced by one point to result in the desired 16 point titration. The result of this operation is shown in [Figure 5-25](#).

Note that the Remove Zone Overlap button is unavailable again because the zone overlap was removed.

Figure 5-25 Titration1 pane—creating a multizone titration1 layout, part 3

Titration1 (1): New Fluid 1

Select which fluids will each use this titration:
 * (Fluids must have the same concentration)
 * (Select none to skip to the next step)

Fluid Name	Conc. (mM)	Use
New Fluid 1	10.0	<input checked="" type="checkbox"/>
New Fluid 2	10.0	<input type="checkbox"/>
New Fluid 3	10.0	<input type="checkbox"/>
New Fluid 4	10.0	<input type="checkbox"/>

How many replicates of this titration would you like?

How many titration zones are there?

Define the points in each zone:

Zone	Low (μM)	High (μM)	Titration Type	No. of Points
1	0.032	0.316	Log	8
2	0.001	0.006	Log	2
3	5.621	100.000	Log	2

Valid Concentration Range: 0.001 μM to 100.000 μM

Buttons: Define by Center..., Remove Zone Overlap, Modified Range: No...

Graph showing concentration (μM) on the x-axis with points at 0.001, 0.006, 0.032, 0.316, 5.621, and 100.000. The graph is divided into three zones.

Summary: Fluids: 1, Replicates: 1, Points: 12, Dispenseheads: 1, Wells: 12

Buttons: < Back, Next >, DIs to use: 1, Wells to Use: 12 / 96, Cancel

4. Click **Next** or on **Titration1: <none>** in the left pane to create another titration for a compound with a different IC50 value to center on.
5. Repeat steps 2 and 3, above, for each compound.
6. Continue with positioning titration as described in “[Position a single zone titration1](#)” above.

5-10-2 Create modified range titration1 layouts

A modified range titration1 is a titration that uses a different stock fluid concentration to expand or shift the dynamic range of the titration titration. For example, by diluting a 10 mM compound 100X, the dynamic range available for titration can be shifted by two log units. Another example would be to use two Dispenseheads, one for the original 10 mM fluid and the other for the diluted 0.1 mM compound, to allow the dynamic range available for the titration to be extended by two log units, effectively increasing the dynamic range. Modified range titrations can only be created with the Wizard if the dispenser is setup to use concentration mode.

1. If not already in Concentration mode, click **Setup/Options.../Units** and select the Concentration radio button as described in “[Options: Edit: Units](#)”.
2. To generate a modified range titration1 with a shifted titration range, follow steps 1 and 2 in [5-9-1](#) above, and then click **Titration1** in the left pane of the Plate Layout Wizard: Titrations and Positioning dialog box. This opens the Titration1 pane. click **Titration1: <none>** in the left pane of the Plate Layout Wizard: Titrations and Positioning dialog box.
3. Select the fluid(s) you would like to use for this modified range titration by clicking in the appropriate **Use** box for each desired fluid, or click the **All** button to use all displayed fluids.
4. For simplicity, set the number of titration zones to 1: click the arrow on the **How many titration zones are there?** drop-down list box and select **1** as shown in [Figure 5-28](#).
5. Type a value that is below the valid concentration range into the Low (μM) column, for example **0.00002** μM. Note that the number in the table turns red to indicate it is outside the valid range.

Figure 5-26 Titration1 pane showing modified range layout

How many titration zones are there? 1

Define the points in each zone:

Zone	Low (µM)	High (µM)	Titration Type	No. of Points
1	2.000e-05	0.002	Log	12

Valid Concentration Range: 0.001 µM to 100.000 µM

Buttons: Define by Center... Remove Zone Overlap Modified Range: No...

Callout 1: Type a value that is below the valid concentration range here. The value will turn red to indicate that it is outside of the valid range.

Callout 2: Valid concentration range

- Click the **Modified Range** button. The Valid Range Modification dialog box appears. See **Figure 5-27**.

Figure 5-27 Valid Range Modification dialog box

Valid Range Modification

You can modify the valid concentration range to better suit the points in this sequence, by changing the fluid concentration using one of the methods below:

Type of Range Modification

☒ Fluid Shift (reduce fluid concentration; span remains the same)

☐ Fluid Extension (create an additional fluid with reduced concentration; span is increased)

Reduce fluid concentration to: 0.1 mM

Optimum: 0.2 mM

Valid: 1.0e-12 mM to 10.0 mM

Resulting Valid Concentration Range: 1.300e-05 µM to 1.000 µM

Buttons: Use Modification Remove Modification Cancel

- Click the **Fluid Shift** option.

Note that a suggestion is made to reduce the fluid concentration to 0.1 mM to enable access to 0.00002 µM concentration with a single drop. Since fluid shift was selected, only one stock concentration will be used for this fluid, and the maximum concentration is shown as 1 µM due to the 1% DMSO limit in a 100 µL assay.

- Click the **Use Modification** button, then note that 100 µM High (µM) column entry is now red, indicating it is outside the new modified range for this fluid. Change the value in the High (µM) column to the upper limit specified by the Valid Concentration Range display in the dialog box (1 µM in this case) and the titration is complete. Make sure to remember to dilute the actual fluid concentration to 0.1 mM when this plate layout is run with the dispenser. The result of this titration is shown in **Figure 5-28**.

Figure 5-28 Titration1 pane showing Modified Range titration1 layout

Plate Layout Wizard: Titrations and Positioning

Titration1 (1): New Fluid 1, New Fluid 2, New Fluid 3, New Fluid 4, New Fluid 5, New Fluid 6, New Fluid 7, N...

Select which fluids will each use this titration:
 * (Fluids must have the same concentration)
 * (Select none to skip to the next step)

Fluid Name	Conc. (mM)	Use
New Fluid 1	0.7	<input checked="" type="checkbox"/>
New Fluid 2	0.7	<input checked="" type="checkbox"/>
New Fluid 3	0.7	<input checked="" type="checkbox"/>
New Fluid 4	0.7	<input checked="" type="checkbox"/>

How many replicates of this titration would you like?
 1

How many titration zones are there?
 1

Define the points in each zone:

Zone	Low (μM)	High (μM)	Titration Type	No. of Points
1	2.000e-05	0.002	Log	12

Valid Concentration Range: 1.300e-05 μM to 1.000 μM

Define by Center... Remove Zone Overlap Modified Range: Yes...

Conc. (μM) Log

2.000e-05 0.002

Fluids: 8
 Replicates: 1
 Points: 12
 Wells: 96

Dispenseheads: 8

< Back Next > DHs to use: 8 Wells to Use: 96 / 96 Cancel

Alternatively, if a 100 μM maximum concentration is desired instead of the 1 μM limit described in step 7, above, the Modified Range function can be used in the Fluid Extension mode.

9. Set the Low (μM) at 0.00002 μM if you have not, and set the High (μM) to 100 μM. Note that the 100 μM text is red, indicating that it is outside the valid concentration range.
10. Click the **Modified Range** button. The Valid Range Modification dialog box appears.
11. Select the **Fluid Extension** option.

Figure 5-29 Valid Range Modification dialog box

Valid Range Modification

You can modify the valid concentration range to better suit the points in this sequence, by changing the fluid concentration using one of the methods below:

Type of Range Modification

☐ Fluid Shift (reduce fluid concentration; span remains the same)

☒ Fluid Extension (create an additional fluid with reduced concentration; span is increased)

Concentration of the additional fluid: 0.1 mM

Optimum: 0.2 mM

Valid: 1.3e-04 mM to 10.0 mM

Resulting Valid Concentration Range: 1.300e-05 μM to 100.000 μM

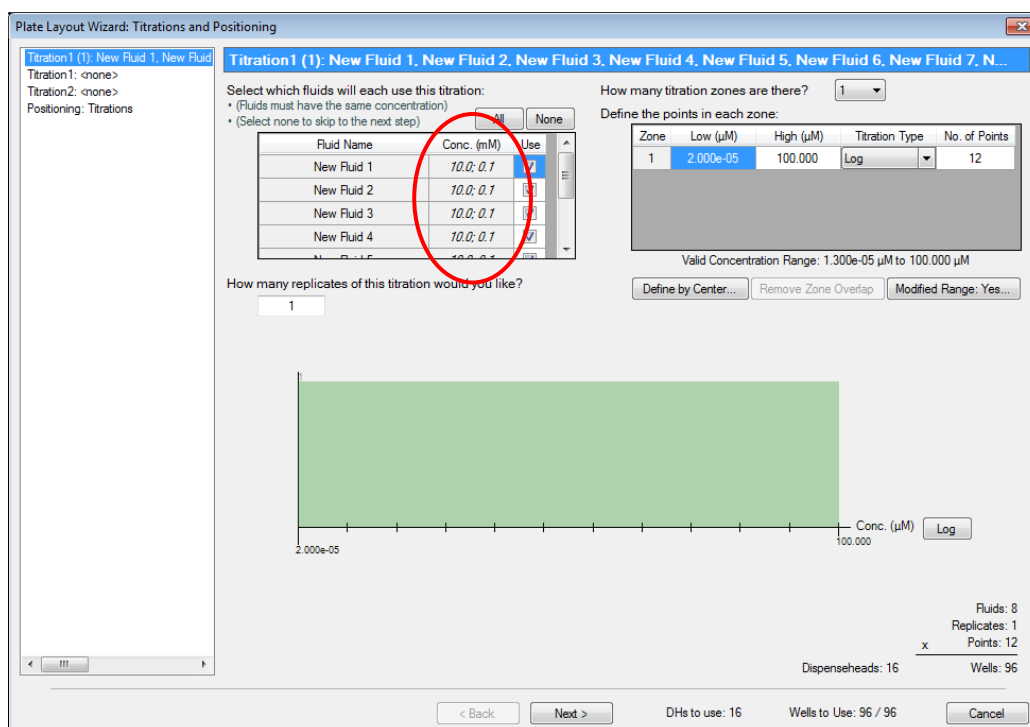
Use Modification Remove Modification Cancel

Note that the wizard suggests using an additional fluid at a concentration of 0.1 mM, which provides a valid concentration range of 0.00002 μM to 100 μM, exactly what is needed.

12. Click the **Use Modification** button.

Note that the fluid table now has two entries in the Conc.(mM) column with two Dispenseheads when this plate layout is run. This is shown in [Figure 5-30](#).

Figure 5-30 Titration1 pane showing two entries in the Conc. (mM) column

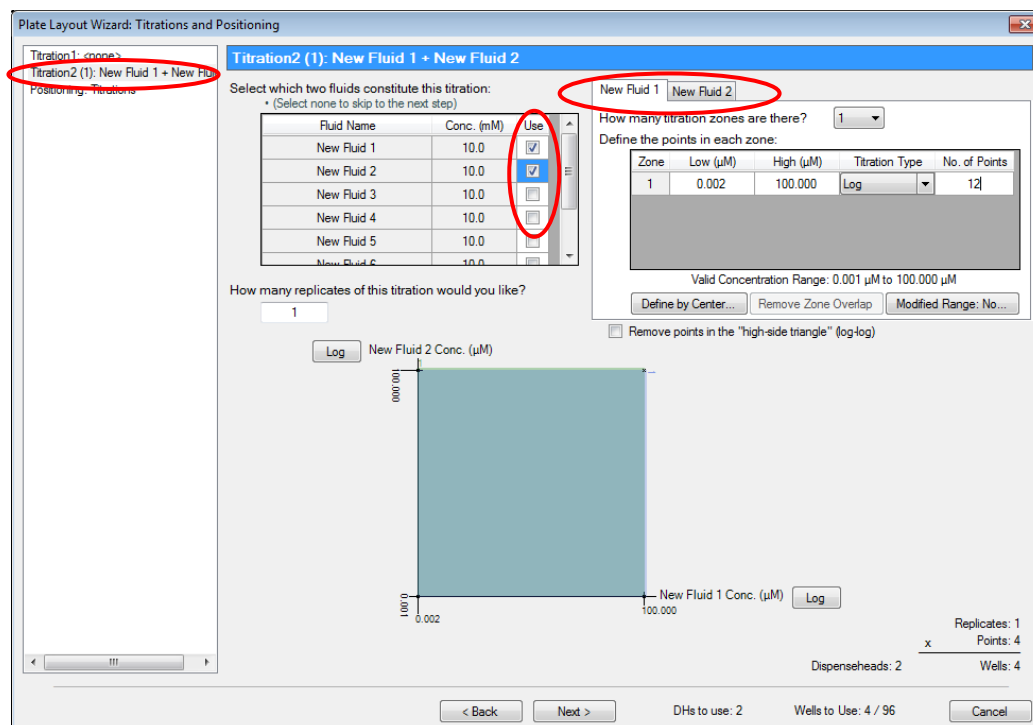


5-10-3 Create titration2 layouts: two fluids per well

A titration2 layout creates a series of wells for each combination of fluid concentrations. For example, if each fluid is designed as a twelve-point singlet, Titration2 will create a 12 × 12 matrix of 144 wells of fluid combinations unless the “Remove points in the high side triangle” option is selected as described below.

1. To set up a titration2 layout with two fluids per well in a systematic design, follow steps 1 and 2 in **5-9-1** above,
2. Click **Titration2** in the left pane of the Plate Layout Wizard: Titrations and Positioning dialog box. This opens the **Titration2: <none>** in the left pane of the Plate Layout Wizard: Titrations and Positioning dialog box. The Titration2 pane is opened as shown in **Figure 5-31**.
3. Select two fluids you would like to use for this dual fluid titration by selecting the **Use** check boxes.

Figure 5-31 Titration2 pane of the Plate Layout Wizard: Titrations and Positioning dialog box



4. Define the titration for the first compound.

Notice that there are two tabs in the titration definition section of the Titration2 pane.

To keep things simple, we will use one replicate and one titration zone titration for both compounds, over the maximum Valid Concentration Range of 2 nM to 100 μM. Of course, more complicated designs can be created using the techniques described in the “[Create multizone titration1 layout](#)” section, and the “[Create modified range titration1 layouts](#)” section.

- Make sure the first tab is selected for the first compound fluid.
 - Select **1** in the **How many titration zones are there?** drop-down list box.
 - Type **0.002** in the Low (μM) column.
 - Type **100** in the High (μM) column.
 - Type **12** in the No. of Points column to create a 12-point singlet.
- Click the second tab for the second compound and use the same settings as you did for fluid 1.
 - If desired, select the **Remove points in the “high-side triangle” (log-log)** check box.
In looking for additive effects one might choose to ignore the high-high drug concentration combinations. This will result in removing all combinations of the two fluids which have a combined concentration exceeding the average of the High (μM) concentrations of the two fluids, the result of which is shown in the graph at the bottom of the dialog box.
 - Note that the design uses 78 of the 96 wells when the “remove points in the high-side triangle” is selected, leaving room for controls on the plate.

Figure 5-32 Titration2 pane with remove points in the high-side triangle selected

Plate Layout Wizard: Titrations and Positioning

Titration1: <none>
 Titration2 (1): New Fluid 1 + New Fluid 2
 Positioning: Titrations

Select which two fluids constitute this titration:
 • (Select none to skip to the next step)

Fluid Name	Conc. (mM)	Use
New Fluid 1	10.0	<input checked="" type="checkbox"/>
New Fluid 2	10.0	<input checked="" type="checkbox"/>
New Fluid 3	10.0	<input type="checkbox"/>
New Fluid 4	10.0	<input type="checkbox"/>
New Fluid 5	10.0	<input type="checkbox"/>
New Fluid 6	10.0	<input type="checkbox"/>

How many replicates of this titration would you like?
 1

Log New Fluid 2 Conc. (μM)

200.000

0.002

New Fluid 1 Conc. (μM)

100.000

Log

Define by Center... Remove Zone Overlap Modified Range: No...

☒ Remove points in the "high-side triangle" (log-log)

Replicates: 1
 Points: 78
 Wells: 78

Dispenseheads

DHs to use: 2 Wells to Use: 78 / 96

< Back Next > Cancel

Remove points in the
 "high-side triangle"
 (log-log) check box

Note that the design
 uses 78 of the 96 wells

6 Troubleshooting and error messages

6-1 Troubleshooting

In general, the HP D300 Digital Dispenser requires proper USB communication between the PC and the dispenser. When unusual events occur, the best first step is to shut down the Digital Dispenser Software, then power off the Digital Dispenser using the rocker switch at the right rear of the Dispenser, wait a few seconds and then turn the Dispenser back on, and finally, restart the software. Then see if the problem encountered can be replicated. If the problem persists and cannot be resolved using the simple procedures described below, call your Tecan Service Help Desk for assistance. The phone number can be found at <http://www.tecan.com/customersupport>.

6-1-1 Problems with software

1. Generally, software problems can be resolved by restarting the software.
 - a. Save the current configuration if desired.
 - b. Close the software using the upper right X or select **File/Exit**.
 - c. Turn the D300 Dispenser off using the rear rocker switch.
 - d. Wait five seconds, then turn the D300 Dispenser back on with the rear rocker switch.
 - e. Restart the software using the desktop icon (the black HP logo).
2. The next step is to reinstall the software, either from the CD enclosed with the D300 Digital Dispenser, or preferably from the *.exe file of the latest version available on the Tecan FTP site: ftp.tecan.com

Username: HP-D300

Password: HP+Tecan

This FTP site can also be accessed directly from the HP D300 Digital Dispenser software by clicking on **Help/About** and then selecting the **Go There Now** link.

3. The software is installed in the C:\Program Files\HP-D300 directory, and the file "HP D300 Digital Dispenser Operating Manual.pdf" needs to be in that same directory in order to be launched from the Help/Operating Manual menu item. Default configurations are in C:\Program Files\HP-D300\Configs and default reports in C:\Program Files\HP-D300\Reports
4. For other software issues contact your local Tecan Service Help Desk. The phone number can be found at <http://www.tecan.com/customersupport>.

6-1-2 Computer issues

1. Hardware minimum configuration is listed in the "**Hardware**" section.
2. Computer issues may relate to hardware configuration or to driver problems. Driver problems generate a "device not found" error and can be fixed by installing the driver as described in "**Insert the CD that was enclosed with the Dispenser**" and be sure to click on the **Install Driver** button in the main Setup window.
3. Ensure that the D300 Digital Dispenser driver was installed correctly from the setup.exe file using the instructions provided in the "**Insert the CD that was enclosed with the Dispenser**" instructions, or preferably from the *.exe file in the latest version available on the Tecan FTP site: ftp.tecan.com

Username: HP-D300

Password: HP+Tecan

This FTP site can also be accessed directly from the HP D300 Digital Dispenser software by clicking on **Help/About** and then selecting the **Go There Now** link.

4. The USB cable between the computer and the Dispenser must be fully connected. Close the software, turn off the Dispenser, remove the USB cable at both ends and reconnect firmly. Restart the Dispenser and then the software. It is worthwhile to try a different USB port on the computer if communication cannot be established. USB cables themselves are rarely at fault.
5. For other computer issues contact your local Tecan Service Help Desk. The phone number can be found at <http://www.tecan.com/customersupport>.

6-1-3 Unable to home stage

Occasionally, the error message “Unable to home stage” may occur upon initiating the HP D300 Digital Dispenser software. This may be due to the stage sensors exceeding their limits. If this message occurs upon startup, close the software and power down the dispenser using the rocker switch on the right rear of the dispenser as shown in “**Turn off the HP D300 Digital Dispenser**”. Once the power is off, it is easy to move the stage by holding the destination plate holder (do not push it with the spring clips, they could break), and moving the stage near the center of its x,y travel. Then power the dispenser back on and restart the software. If this still results in an “Unable to home stage” message, contact your Tecan Service Help Desk. The phone number can be found at <http://www.tecan.com/customersupport>.

6-1-4 Check the A1 location

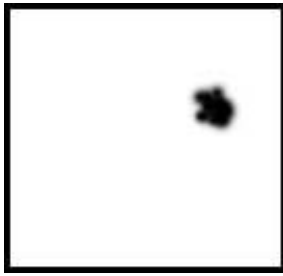
If the dispenser has been treated roughly, during shipment or movement to a new location, the position of the stage assembly relative to the position of the DispenseHead Cassette may change. When you would like to test the dispenser to make sure the alignment is correct, or if you notice poor CVs or fluids dispensed on the side walls or ribs of the destination plate, follow the following procedure to test the alignment and modify it if needed. Please use this section only if you are comfortable with resetting computer parameters for instruments such as the HP D300 Digital Dispenser. If you are unsure about these procedures at any point, call your Tecan Service Help Desk for assistance. The phone number can be found at <http://www.tecan.com/customersupport>.

1. Load the 384 Four Corners Centration configuration by clicking **File** and **Load Plate Configuration**, browsing to the C:\Program Files\HP-D300\Configs directory and then selecting **384_Four_Corners_Config.xls**.
2. Click the **GO** button and when prompted, place a 384 well plate with clear tape over the four corner wells onto the destination plate holder. Use pure DMSO or add some colorant to DMSO to use as the dispense fluid.
3. When the fluid is dispensed in the four corners and the plate moves back to plate load position, examine the fluid location relative to the center of the four corner wells.
4. If the fluid in any one corner is off by more than 1.0 mm in a different direction than the other corners, the stage assembly is not square and you need to contact your Tecan Service Help Desk. The phone number can be found at <http://www.tecan.com/customersupport>. If the fluids in all four corners are offset in the same direction, continue following the **A1 alignment procedure** below.

6-1-5 A1 alignment procedure

1. If the A1 position is not correct, make a little picture as shown in **Figure 6-1** that shows where the drop landed relative to the well outline. This will help you to remember which direction the drop landed relative to the center of the well.

Figure 6-1 Make a picture that shows where the drop landed



2. Measure the approximate offset of the fluid from the center of the well using a metric ruler. Just measure within a quarter millimeter or so, then note the offsets on your drawing.
3. Wipe the dispensed fluid off of the tape that covers the well plate and replace the plate on the destination well plate holder.
4. Click **Setup, Hardware**. The Stage Setup dialog box opens.

Figure 6-2 Stage Setup pane

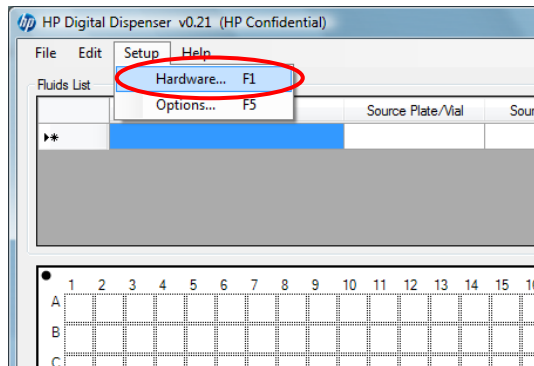
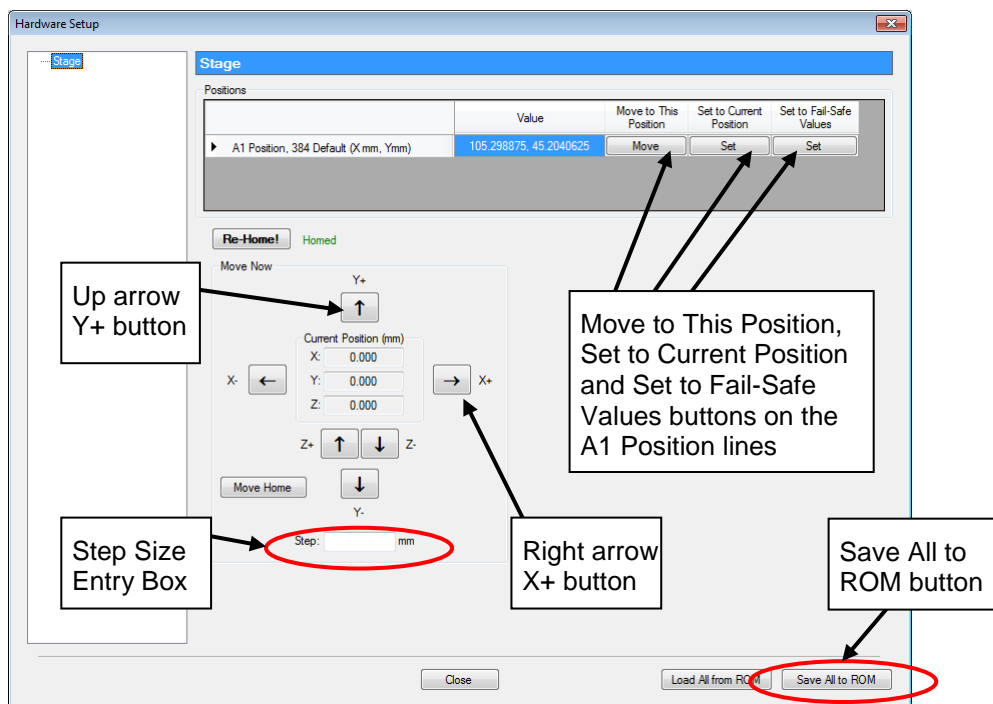


Figure 6-3 Hardware Setup Stage pane



5. Click the **Move** button under **Move to This Position** on the **A1 Position, 384 Default** line. This will move the plate so that the A1 well of the plate is centered under Dispensehead 1 of the Cassette.
6. In the **Step:** box near the bottom of the dialog box, type **0.25**. You may choose a different step size (in mm) if appropriate.
7. In the example of **Figure 6-1** showing the drop location relative to the well walls, the A1 position is off by about 1 mm in the X direction and 0.5mm in Y. Remember that the plate moves under the fixed Dispensehead, so that you are adjusting the location of the well, not the location of the fluid, so in the example shown in **Figure 6-1** the well plate position must be moved to the right and up.
8. Since the step size is 0.25 mm, click the **right arrow X+** button four times, then click the **up arrow Y+** button two times. Click the arrow buttons as appropriate for the A1 offset distances and the step size for your situation.
9. Reset the A1 location to this new position by clicking the **Set** button under **Set to Current Position** in the **A1 Position, 384 Default** line. The A1 position for 96 plates will be automatically adjusted based upon the change selected for the 384 plate, offset from the 384 A1 position by the ANSI standard distance difference.
10. Click the **Save All to ROM** button in the lower right hand corner of the stage setup window, otherwise your changes will not be saved the next time you start the software.
11. Close the **Setup/Hardware/Stage** menu by clicking on OK at the bottom of the window or on the X in the upper right corner.
12. Repeat steps 2-4 of the **Check the A1 Alignment** procedure above and steps 1-12 of this procedure as required until the fluid dispense location is centered in corner wells.
13. At any point, if you lose track of the appropriate A1 position or cannot get the fluid to dispense within the appropriate well, click on the **Set** button under **Set to Fail Safe Values** in the **A1 Position, 384 Default** line. Then you can fine tune this rough position using steps 1-12 above.

6-1-6 Check the fuses and replace if necessary

If the Dispenser does not turn on when the rear rocker switch is depressed at the “1” position as shown in **Figure 6-4**, then check the fuse at the inlet power receptacle.

Figure 6-4 On/off switch on rear of dispenser. Depress “1” to turn on.



The following tools are required for this procedure:

- M2 hex wrench
 - Small slotted screwdriver
1. Use a small flat head screwdriver to flip open and down the cover of the AC power inlet module on the back of the dispenser. See **Figure 6-5**.

Figure 6-5 Pry the cover at the top of the power inlet module and flip down



2. Use the small screwdriver to pry the red fuse holder out of the AC power inlet module. See **Figure 6-6** and **Figure 6-7**.

Figure 6-6 Flip down the black cover and pry out the red fuse holder

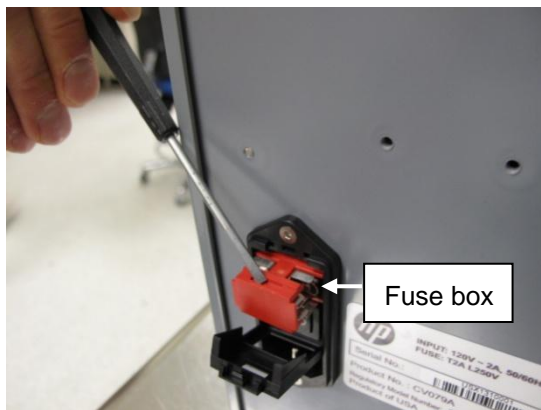


Figure 6-7 Fuse holder

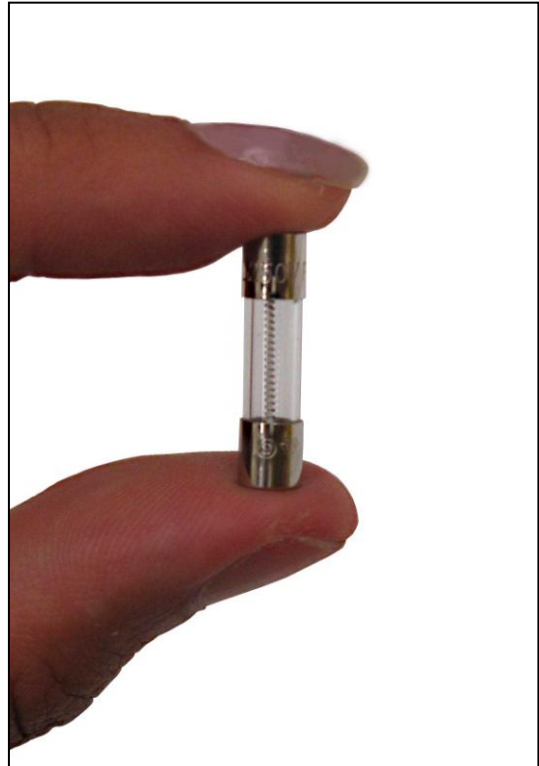


3. Carefully pry the fuse out of the holder with the small screwdriver or other tool and look for visible damage. The fuse is good if the filament is visibly continuous and unbroken from one end of the fuse to the other. See **Figure 6-8** and **Figure 6-9**.

Figure 6-8 Pry the fuse out carefully

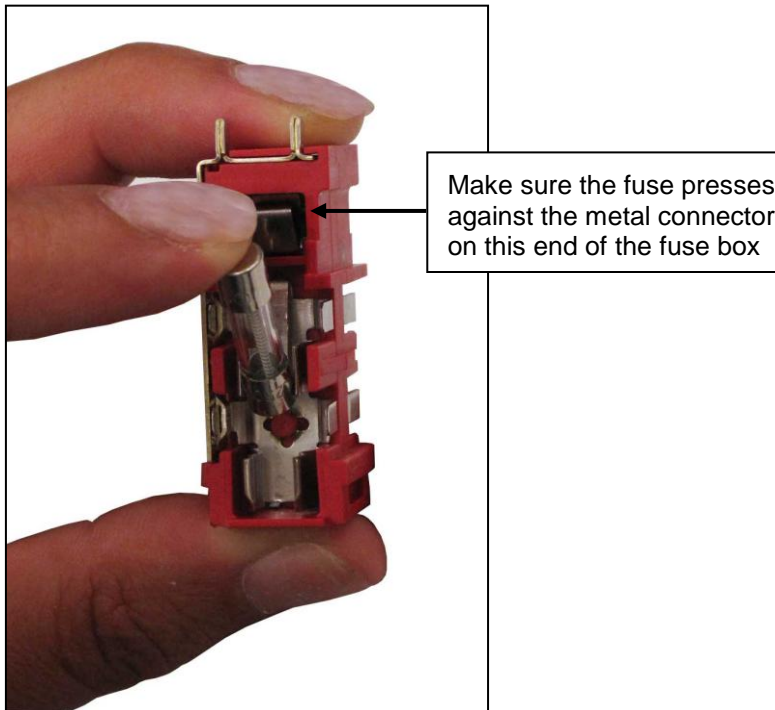


Figure 6-9 An example of a good fuse



4. If the fuse is not damaged, insert it back into the fuse holder, making sure that one end of the fuse presses against the metal connector on the pronged end of the fuse holder. In **Figure 6-10**, the pronged end is up.

Figure 6-10 Insert the fuse so that one end presses against the metal connector on the pronged end of the fuse holder



5. If the fuse is damaged, replace both fuses with a new one. If either fuse is damaged, both fuses should be replaced.

6. Turn the fuse holder over and remove the fuse on the other side of the fuse holder. Check this fuse for damage as well. If either fuse is damaged, both fuses should be replaced.
7. Insert the fuse holder back into the dispenser, flip the black cover up and click it into place.

6-2 Error messages

If you encounter an error message or warning while using the HP D300 Digital Dispenser software, note the title of the error message and the error message text as shown in Figure 6.1, and refer to the alphabetized list of error messages below to determine the appropriate response.

Table 6-1 Error message guide

Error message text	Error message title	Recommended action
ABORT	Dispense Operation Error	Retry GO operation. ABORT is a user-requested interruption, not a dispenser failure.
An unexpected error occurred, forcing premature termination	Dispense Operation Error	Save the configuration, close the software, and turn the dispenser off, Restart the dispenser, restart the software, and retry the configuration.
Could not access the firing system. Ensure driver is installed correctly.	Firing System Error	Close the software, unplug the USB cable that connects the dispenser to your computer, and follow the Insert the CD that was enclosed with the Dispenser instructions.
Could not access the pogos. Ensure driver is installed correctly.	Pogo Error	Close the software, unplug the USB cable that connects the dispenser to your computer, and follow the Insert the CD that was enclosed with the Dispenser instructions.
Could not access the stage. Ensure driver is installed correctly.	Stage Port Error	Close the software, unplug the USB cable that connects the dispenser to your computer, and follow the Insert the CD that was enclosed with the Dispenser instructions.
Could not open the pogo port. Ensure driver is installed correctly.	Pogo Port Error	Close the software, unplug the USB cable that connects the dispenser to your computer, and follow the Insert the CD that was enclosed with the Dispenser instructions.
Could not sense the plate; plate is missing or is too thin.	Stage Motion Error	Ensure that a plate is on the destination plate holder and use a plate with a thickness greater than 12mm.
Could not sense the top of the plate; plate may be too thick to use.	Stage Motion Error	Ensure that a plate is on the destination plate holder and use a plate with thickness less than 18mm.
Excel is missing or is not the correct version. You must have Excel 2003 or later installed to use this function.	Excel Configuration Load Error	Ensure that Excel 2003 or later is installed on your computer and works properly.
Excel is missing or is not the correct version. You must have Excel 2003 or later installed to use this function.	Excel Configuration Save Error	Ensure that Excel 2003 or later is installed on your computer and works properly.

Table 6-1 Error message guide

Error message text	Error message title	Recommended action
Excel is missing or is not the correct version. You must have Excel 2003 or later installed to use this function.	Old or Missing Excel	Ensure that Excel 2003 or later is installed on your computer and works properly.
Firing system communications was lost! The dispense operation has terminated.	Dispense Operation Error	Save the configuration, close the software, and turn the dispenser off. Restart the dispenser, restart the software, and retry the configuration.
No Cassette detected, or no usable Dispenseheads on this Cassette.	Cassette Error	Ensure that a Dispensehead Cassette is loaded properly on the dispenser. If unsure how to load a Dispensehead Cassette, refer to “Load a new Dispensehead Cassette” .
Pogo communications was lost! The dispense operation has terminated.	Dispense Operation Error	Save the configuration, close the software, and turn the dispenser off. Restart the dispenser, restart the software, and retry the configuration.
Stage communications was lost! The dispense operation has terminated.	Dispense Operation Error	Save the configuration, close the software, and turn the dispenser off. Restart the dispenser, restart the software, and retry the configuration.
The Dispensehead-plate spacing is too large; use a thinner plate.	Stage Motion Error	Ensure that a plate is on the destination plate holder and use a plate with thickness less than 18mm.
The following error has occurred. (FIRSYS)	Dispense Error	Save the configuration, close the software, and turn the dispenser off. Restart the dispenser, restart the software, and retry the configuration.
The following error has occurred. (STAGE)	Dispense Error	Save the configuration, close the software, and turn the dispenser off. Restart the dispenser, restart the software, and retry the configuration.
The Microsoft Office Primary Interop Assembly (PIA) for Excel could not be found. Re-install this application, or download and install the Microsoft Office PIAs from Microsoft's website. Would you like to go to this download location now?	Microsoft Office PIAs Not Found	Reinstall Excel, or download and install the Microsoft Office PIAs from Microsoft's website.
The number of drops per nozzle is invalid.	Invalid Drops Per Nozzle	Save the configuration, close the software, and turn the dispenser off. Restart the dispenser, restart the software, and retry the configuration.
There is no known well plate that matches the configuration file's plate dimensions. This configuration cannot be loaded.	Excel Configuration Load Error	Ensure that the file that is opened is a configuration file from the HP D300 Digital Dispenser and that it has not been altered.
This file is not in a supported format.	Excel Configuration Load Error	Ensure that the file that is opened is an .xls configuration file from the HP D300 Digital Dispenser and that it has not been altered.
This file is not in the correct format.	Invalid File	Ensure that the file that is opened is an .xls configuration file from the HP D300 Digital Dispenser and that it has not been altered.

Table 6-1 Error message guide

Error message text	Error message title	Recommended action
Unable to access this file. (Is it currently being used by another program?)	File Access Error	Ensure that the file is not being used by another program.
Unable to communicate with device!	Data Format Error	Use Microsoft Windows Explorer to ensure that the file location is accessible from your computer.
Unable to communicate with the instrument; please ensure that the drivers are installed correctly and all firmware are up-to-date.	Startup Error	Close the software, unplug the USB cable that connects the dispenser to your computer, and follow the "Insert the CD that was enclosed with the Dispenser" instructions. Restart the software and, if requested, follow instructions to update firmware.
Unable to find the instrument; please ensure that the instrument is powered and connected to the PC.	Startup Error	Close the software, check the USB connection between the dispenser and your computer, turn the dispenser on, and restart the software.
Unable to home due to possible obstructions. Ensure there are none and that the Cassette, if present, is seated properly.	Unable to Move	Ensure that nothing is obstructing the motion of the pogo mechanism or the destination plate holder, and that the Dispensehead Cassette is seated properly. Restart the software if retry fails.
Unable to home due to possible obstructions. Ensure there are none and that the well plate, if present, is seated properly, and then re-home.	Unable to Home	Ensure that nothing is obstructing the motion of the destination plate holder and that the Dispensehead Cassette is seated properly. Restart the software if retry fails.
Unable to home the pogos. (Could not move to destination.)	Pogo Home Error	Ensure that nothing is obstructing the motion of the pogo mechanism and that the Dispensehead Cassette is seated properly. Restart the software if retry fails.
Unable to home the pogos. (Could not move to destination.)	Startup Error	Ensure that nothing is obstructing the motion of the pogo mechanism, and restart the software.
Unable to home the pogos. (Lost comms?)	Pogo Home Error	Ensure that nothing is obstructing the motion of the pogo mechanism and that the Dispensehead Cassette is seated properly. Restart the software if retry fails.
Unable to home the pogos. (Lost comms?)	Startup Error	Ensure that nothing is obstructing the motion of the pogo mechanism, and restart the software.
Unable to home the stage. (Could not move to destination.)	Stage Home Error	Ensure that there is nothing obstructing the motion of the destination plate holder and that the Dispensehead Cassette is seated properly. Restart the software if retry fails.

Table 6-1 Error message guide

Error message text	Error message title	Recommended action
Unable to home the stage. (Could not move to destination.)	Startup Error	Ensure that nothing is obstructing the motion of the destination plate holder, and restart the software.
Unable to home the stage. (Lost comms?)	Stage Home Error	Ensure that nothing is obstructing the motion of the destination plate holder and that the Dispensehead Cassette is seated properly. Restart the software if retry fails.
Unable to home the stage. (Lost comms?)	Startup Error	Ensure that nothing is obstructing the motion of the destination plate holder, and restart the software.
Unable to initialize Dispenseheads. (Could not move to destination.)	Cassette Initialization Error	Ensure that nothing is obstructing the motion of the pogo mechanism and that the Dispensehead Cassette is seated properly. Restart the software if retry fails.
Unable to initialize Dispenseheads. (Lost comms?)	Cassette Initialization Error	Ensure that nothing is obstructing the motion of the pogo mechanism, and restart the software.
Unable to initialize the following hardware components: Firing System	Startup Error	Close the software, check the USB connection between the dispenser and your computer, turn the dispenser on, and restart the software.
Unable to initialize the following hardware components: Pogo	Startup Error	Close the software, check the USB connection between the dispenser and your computer, turn the dispenser on, and restart the software.
Unable to initialize the following hardware components: Stage	Startup Error	Close the software, check the USB connection between the dispenser and your computer, turn the dispenser on, and restart the software.
Unable to initialize the well plate. (Could not move to destination.)	Plate Initialization Error	Ensure that nothing is obstructing the motion of the destination plate holder. Restart the software if retry fails.
Unable to initialize the well plate. (Lost comms?)	Plate Initialization Error	Ensure that nothing is obstructing the motion of the destination plate holder and restart the software.
Unable to initialize the well plate. (No plate detected.)	Plate Initialization Error	Ensure that a plate is on the destination plate holder and use a plate with a thickness greater than 12mm.
Unable to initialize the well plate. (Plate is too thick.)	Plate Initialization Error	Ensure that a plate is on the destination plate holder and use a plate with thickness less than 18mm.

Table 6-1 Error message guide

Error message text	Error message title	Recommended action
Unable to load this configuration: no well plate information was found in the file.	Excel Configuration Load Error	Ensure that the file that is opened is an .xls configuration file from the HP D300 Digital Dispenser, and that it has not been altered.
Unable to move to destination due to possible obstructions. Ensure there are none and that the well plate, if present, is seated properly, and then re-home.	Unable to Move	Ensure that nothing is obstructing the motion of the destination plate holder. Restart the software if retry fails.
Unable to move to the static bar position. (Could not move to destination.)	Move to Static Bar Error	Ensure that nothing is obstructing the motion of the destination plate holder. Restart the software if retry fails.
Unable to move to the static bar position. (Lost comms?)	Move to Static Bar Error	Ensure that nothing is obstructing the motion of the destination plate holder and restart the software.
Unable to move to this location. Ensure there are no obstructions and that the well plate is seated properly.	Stage Motion Error	Ensure that nothing is obstructing the motion of the destination plate holder. Restart the software if retry fails.
Unable to open the operating manual 'HP D300 Digital Dispenser Operating Manual.pdf' (Do you have a PDF reader installed?)	No Manual Found	Install Adobe Acrobat Reader if it is not already installed on your computer. Ensure that the HP D300 Digital Dispenser Operating Manual.pdf file is the same directory as the D300 software .exe file. (Right-click on the desktop shortcut and select Properties to determine the location of the .exe file.)
Unable to open this file (is it locked or being used by another program?).	File Access Error	Ensure that the file is not being used by another program.
Unable to open this file. (Is it locked or being used by another program?)	Excel Configuration Load Error	Ensure that the file is not being used by another program.
Unable to read the entire program file.	Program File Read Error	Close the software and reinstall the HP D300 Digital Dispenser software from the installation CD.
Unable to retract the pogos. (Could not move to destination.)	Pogo Retraction Error	Ensure that nothing is obstructing the motion of the pogo mechanism and that the Dispensehead Cassette is seated properly. Restart the software if retry fails.
Unable to retract the pogos. (Lost comms?)	Pogo Retraction Error	Ensure that nothing is obstructing the motion of the pogo mechanism, and restart the software.

Table 6-1 Error message guide

Error message text	Error message title	Recommended action
Unable to send! Some data are not in the correct format.	Data Format Error	Save the configuration, close the software, and turn the dispenser off. Restart the dispenser and then restart the software
Unable to start dispensing this split. (Already dispensing.)	Dispense Error	Wait until the dispenser is finished dispensing and retry if necessary.
Unable to start dispensing this split. (Invalid Dispensehead number.)	Dispense Error	Save the configuration, close the software, and turn the dispenser off. Restart the dispenser, restart the software, and retry the configuration.
Unable to start dispensing this split. (Lost comms?)	Dispense Error	Save the configuration, close the software, and turn the dispenser off. Restart the dispenser, restart the software, and retry the configuration after deleting wells that have already been dispensed.
Unable to start dispensing this split. (Unknown plate type.)	Dispense Error	Save the configuration, close the software, and turn the dispenser off. Restart the dispenser, restart the software, and retry the configuration.

Appendix A

HP D300 Digital Dispenser software basics

Global view

The global view of the plate contains all the fluids that will be dispensed into the plate whether or not the fluids are selected for editing.

The well plate image on the main page of the software displays only the fluids selected for editing, except during the dispense GO operation, when only the current fluid being dispensed is shown.

Plate Layout Wizard

The Plate Layout Wizard is a set of panes designed to assist the user with plate configurations. Plate Layout Wizard is useful for plate designs which use the same set of concentrations for many fluids or for complex plate designs such as targeted, extended dynamic range or two-fluid titrations.

Working with the software

Exiting windows and dialogs: Clicking on the red X in the top-right corner of a window simply closes the window; no modified settings are saved.

OK/Apply: In the software, OK applies and saves your choices and closes the open window. Apply applies and saves your choices but keeps the current window open. Clicking on the red X in the top-right corner of a window simply closes the window and modified settings are not saved.

Right-click: Right-click after selecting a well or wells in the well plate image to open the Edit Well Plate menu. Use this menu to perform fluid-related tasks on the selected wells. Right-click anywhere in the Fluids Table to open the Edit Fluids List menu.

Selecting wells: To select wells for performing operations such as configuring dispense concentrations or volumes, normalizing, or randomizing, click and drag in the well plate image. Drawing a rectangle that includes any portion of a well selects it. If the desired wells are not selected, click and drag again; the previous selection is discarded. Hold down the CONTROL key while clicking on wells to add or remove wells from the previous selection. To select two or more separate areas which each contain multiple wells, left click and drag to select wells in the first area, then hold down the CONTROL key while the other noncontiguous areas are selected. For instance, this is the way to reserve areas of the plate for controls or blanks while setting up titrations in other regions of the well plate.

Shortcut keys: Shortcut keys are available for many common commands and are shown next to menu options.

Undo/Redo: Click **Edit** and then **Undo** or **Redo** to undo or repeat actions after you perform them. The HPDD software permits up to 20 undo/redo levels.

Appendix B

Glossary

Glossary term	Definition
Assay volume	Total volume that will be present in each well of the plate for the purposes of calculating the concentration of each compound. Also known as final assay volume.
Global view	<p>The global view of the plate contains all the fluids that will be dispensed into the plate whether or not the fluids are selected for editing.</p> <p>The well plate image displays only the fluids selected for editing, except during the dispense GO operation, when only the current fluid being dispensed is shown.</p>
Plate	A well plate, either 96 or 384.
Plate configuration	An Excel file which describes the concentrations or volumes that may be dispensed by the HP D300 Digital Dispenser. Click on File, then Load Plate Configuration, followed by GO, to perform the dispense operations described in the plate configuration.
Titration	A series of different concentrations, volumes and doses of a fluid which comprise a titration series, or the x-axis of a dose response analysis. Titrations may be singlets (one well per dose), doublets (2 replicate wells per dose), triplicates, etc. depending on the entry in the number of replicates text box.
Well plate	A microtiter plate; a plastic plate that is composed of numerous small wells. The well plate receives the fluids that are dispensed by the dispenser. The HP D300 Digital Dispenser uses 96-well and 384-well plates.
Well pop-up	The information about the contents of a well that is displayed when you move the mouse over the well plate image in the main operating window. Note that the first three values (assay volume, total volume, and DMSO content) regard the entire contents of the well. Below these values, information is given separately for each fluid in that well.

Appendix C

Quick Start Guide



START HERE

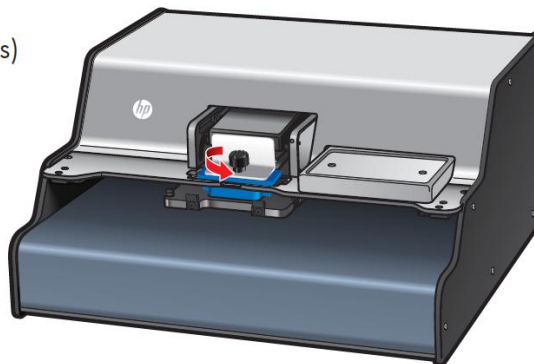
POUR COMMENCER / STARTEN SIE HIER
COMIENCE AQUÍ / はじめにお読みください
시작 / 从这里开始

QUICK SETUP

MANUEL D'INSTALLATION RAPIDE / KURZANLEITUNG / INSTALACIÓN RÁPIDA
クイックセットアップ / 빠른 설치 / 快速安装

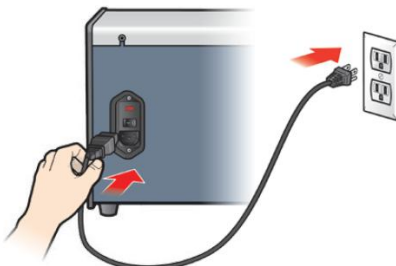
1 Unscrew knob and remove shipping bracket (all 4 pieces)

Dévissez le bouton puis retirez le support d'expédition (l'ensemble des 4 éléments)
Lösen Sie den Drehknopf und entfernen Sie die Versandklammern (4 Stück)
Destornille el tirador y extraiga el soporte de envío (las 4 piezas)
ネジをゆるめてノブを取り外します
出荷用ブラケット (部品4個)
손잡이 나사를 풀고 배송브래킷 (모두 4개) 제거
旋开旋钮并移除运输托架 (4个全部移除)



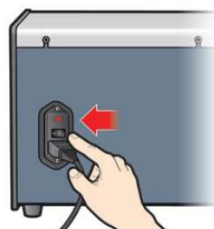
2 Connect power

Connectez à l'alimentation
Stellen Sie eine Verbindung zur Stromversorgung her
Conecte la alimentación
電源を接続します
전원 연결
连接电源



3 Turn on rear power switch

Activez l'interrupteur d'alimentation arrière
Schalten Sie das Gerät an der Rückseite ein
Encienda el interruptor de energía
後部にある電源スイッチをオンにします
후면 전원 스위치 켜기
打开后面的电源



QUICK SETUP

MANUEL D'INSTALLATION RAPIDE / KURZANLEITUNG / INSTALACIÓN RÁPIDA
クイックセットアップ / 빠른 설치 / 快速安装

4 Insert CD. If it does not auto-start, run setup.exe from the CD. Install software and driver.

Insérez le CD. S'il ne se lance pas automatiquement, exécutez setup.exe à partir du CD. Installez le logiciel et son pilote.

Legen Sie die CD-ROM ein. Falls die Setup-Software nicht automatisch startet, führen Sie setup.exe über die CD-ROM aus. Installieren Sie die Software und den Treiber.

Inserte el CD. Si no se inicia automáticamente, ejecute setup.exe desde el CD. Instale el software y el controlador.

CD를挿入します。コンテンツが自動的に起動しない場合は、CDに収録されているsetup.exeのファイルを実行します。ソフトウェアとドライバをインストールします。

CD 넣기, 자동으로 시작되지 않으면 CD에서 setup.exe를 실행합니다. 소프트웨어 및 드라이버를 설치합니다.

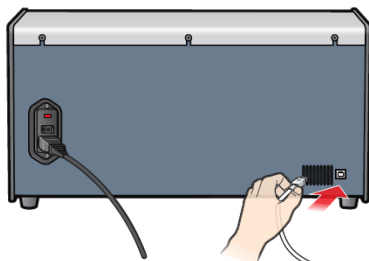
插入光盘。如果没有自动启动，请运行光盘上的安装程序。安装软件和驱动器



5 Plug in USB to dispenser and computer

Branchez le périphérique USB au distributeur et à l'ordinateur
Schließen Sie das USB-Kabel an den Dispenser und an den Computer an

Conecte el USB al dispensador y al equipo
USBをディスペンサとパソコンに接続します
디스펜서 및 컴퓨터에 USB 플러그 꽂기
将 USB 插入分配器和计算机中



6 Start software from desktop icon

Démarrez le logiciel à partir de l'icône du bureau
Starten Sie die Software über das Symbol auf dem Desktop

Inicie el software desde el icono del escritorio
デスクトップにあるアイコンからソフトウェアを起動します

바탕화면 아이콘에서 소프트웨어 시작
通过桌面图标启动软件



POWER SPECS:

CARACTÉRISTIQUES D'ALIMENTATION / TECHNISCHE DATEN ZUR STROMVERSORGUNG:
ESPECIFICACIONES RELATIVAS A LA ALIMENTACIÓN: / 電源仕様: / 전원 사양: / 电源规格:

US CV079A – 120V ~ 2A 50/60Hz
EMEA CV080A – 230V ~ 2A 50/60Hz
Power cable shipped separately.
Contact your authorized HP seller for support and warranty questions.

EE.UU. CV079A – 120 V 2A 50/60 Hz, aproximadamente
EMEA CV080A – 230 V 2A 50/60 Hz, aproximadamente
El cable de alimentación se envía por separado.
Póngase en contacto con su distribuidor HP autorizado para obtener ayuda y respuestas sobre la garantía.

US CV079A – 120V ~ 2A 50/60Hz
EMEA CV080A – 230V ~ 2A 50/60Hz
电源线单独运送。
请联系您的 HP 授权卖家以获取支持和保修信息。

US CV079A – 120V ~ 2A 50/60Hz
EMEA CV080A – 230V ~ 2A 50/60Hz
Cable d'alimentation livré séparément.
Contactez votre revendeur HP pour toutes questions sur l'assistance ou la garantie.

US CV079A – 120V ~ 2A 50/60Hz
EMEA CV080A – 230V ~ 2A 50/60Hz
電源ケーブルは別途発送となります。
サポートや保証についてのご質問は HP 販売代理店までお問い合わせください。

US CV079A – 120V ~ 2A 50/60Hz
EMEA CV080A – 230V ~ 2A 50/60Hz
Das Stromkabel wird separat geliefert.
Kontaktieren Sie Ihren HP-Händler bei Fragen zu Support- und Garantieleistungen.

US CV079A – 120V ~ 2A 50/60Hz
EMEA CV080A – 230V ~ 2A 50/60Hz
전원 케이블 별도 제공.
지원 및 보증에 대한 자세한 내용은 HP 공인 판매업체에 문의해 주십시오.



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Appendix D

Revision History

Table 2

HP Doc. Number: 64A009000de780105924

Edition	Date	Revision
A	December 1, 2011	eCMS 10797. New Document
B	December 5, 2011	eCMS 11125. Edits for SW versions up to 1.03; Quick Start Guide for shipping bracket
C	March 12, 2012	eCMS 14092. Edits for SW version 1.04, primarily multiplate capability