Hydra[®] DT Microdispenser

User Manual

Hydra DT Firmware Version 5.15 or later ControlMate™ Software Version 1.3.33 or later Hydra DT Module Version 1.1.17 or later Manual Version 1.0, October 2006



Matrix Technologies 22 Friars Drive Hudson, NH 03051, USA Telephone: (800) 345-0206

www.matrixtechcorp.com

i



Trademarks and Copyright

© 2006 Matrix Technologies Corporation. All rights reserved.

Hydra, eDrop, TrakMates, ControlMate, DuraFlex, ScreenMates, and the Matrix logo are registered trademarks are trademarks of Matrix Technologies Corporation, Hudson, New Hampshire.

Microsoft, Windows, Windows NT, ActiveX, and Visual Basic are either trademarks or registered trademarks of Microsoft Corporation in the United States and/or other countries.

Third party trademarked products are trademarks or registered trademarks of their respective companies in the United States and/or other countries.



CONTENTS

	Warranty	vi
	Packing List	viii
	Warnings and Cautions	ix
	Stopping the System	ix
	Protecting the Operator	x
	Protecting the System from Damage	x
	How to Use This Manual	xi
1:	Introduction to the Hydra DT Microdispenser	1-1
	Overview of the Hydra DT Microdispenser	1-1
	Hydra DT Base Unit	1-2
	Optional Accessories	
	Hydra DT Microdispenser Model Types	1-4
	Syringe Design and Construction	1-5
	DART Magazine Construction	1-6
	Related Products	1-8
	Contact Us with Questions and Comments	1-9
2:	Setting up the Hardware	2-1
	Prepare the Site	
	Unpack the Microdispenser	
	Moving the Microdispenser	2-2
	Set Up the Stage (Hydra DT with X/Y Plate Stage Only)	2-2
	Change the Voltage Setting for the Microdispenser If Necessary	2-3
	Connect Power Cords and Cables: Turn On the System	2-5
	Configure hardware options	2-7
	Configure XY stage	2-7
	Connect the Computer	
3.	Getting Started: Manual Operation	3-1
0.	Two Operation Methods	3-1
	Steps for Learning Manual Operation of the Hydra DT Microdispenser	3-2
	1 Study the SAFETV Instructions	3_2
	2 Look at flowcharts of Hydra DT interface	3-2
	3. Read the table of file-parameter definitions and values	
	4 Map the microdispenser buttons	
	5. Read the table of microdispenser operations	
	6. Set up a trav table/stage height	
	7. Set file parameters	
	9. Run a procedure	3-11

iii



٨.	Catting Started: Automated Operation	11
4.	Two Operation Matheda	- +- ۱ م
	I wo Operation Methods	4-1
	Steps for Learning Automated Operation of the Microdispenser	4-3
	2. Study the SAFETV instructions	4-4
	3. Bead the overview of the software	4-4 1-1
	4 Check the software configuration	4-13
	5. Calibrate the stage (Hvdra DT with X/Y Plate Stage systems only)	4-21
	6. Create labware	. 4-28
	7. Create and edit sequence files	. 4-39
	8. Run a sequence file	. 4-42
	Command Reference	4-45
	Main Sequence Editor	. 4-46
	Procedure Library	. 4-51
	ControlMate OLE Optional Add-On Software Component	4-69
5:	Service and Maintenance of the Hydra DT Microdispenser	. 5-1
	Overview of Service and Maintenance Tasks	5-2
	Contact Us If You Have Service or Maintenance Questions	5-3
	How to Return the Microdispenser or Microdispenser Parts for Service or Repair	5-3
	Checking the PTFE Tips	5-4
	Replacing PTFE Tips	5-5
	Replacing Syringes	5-7
	Removing the syringe head from the Hydra tower.	5-7
	Syringe Head Disassembly and Syringe Removal	5-9
	Installing a New Syringe	. 5-12
	Attaching the syringe head to the Hydra Tower	. 5-13
	Sending in your DT head	. 5-14
	Checking and Replacing Fuses	5-15
6:	Getting the Most Out of Your Hydra DT Microdispenser	. 6-1
	Basics of Liquid Handling With the Hydra DT Microdispenser	6-1
	Two Aspirate/Dispense Modes	6-1
	Priming Syringes	6-8
	Setting Dispense Heights	6-9
	Using the Microdispenser in Cell Culture Applications	6-15
	Creating Dot Blots and Membrane Arrays With the Vacuum Manifold	6-17
	Using the Wash Operation as a Mix Operation	6-19
	Using the Manually Operated Plate Positioners DT	6-19
	Using the Hydra-96 Plate Positioner for Standard 384-Well Plates	. 6-21
	Using the Hydra-96 Plate Positioner for CyclePlate-384 and 192 Plates	. 6-21
	Dispensing Onto the Sidewalls of Wells in CyclePlate-ET Plates With a Plate Positioner.	. 6-21
	Creating Membrane Arrays With the Vacuum Manifold and a Plate Positioner	. 6-21

iv



Using the Hydra DT 96 With Wide Bore D.A.R.T.s With Resin Suspensions	6-22
General Guidelines for Using the Microdispenser With Resin Suspensions	6-22
Examples of Types of Materials Used in Resin Suspensions on the Hydra DT 96 System With Wide Bore D.A.R.T.s	6-23
Using Viscous Solutions	6-25
Ensuring Compatibility of Chemicals Used With the Hydra DT Microdispenser	6-26
7: Troubleshooting and Tips	7-1
Troubleshooting Topics	7-1
Quick Tips for Using the Hydra DT Microdispenser	7-7

v



Warranty

Please enter the serial numbers and date or dates of installation. Keep this manual near the Hydra DT microdispenser for reference.

	Serial Number	Date Installed
Hydra DT base unit		
2-Position or 3-Position X/Y Plate Stage		
eDrop, Single-Channel Non-Contact Nano Volume Dispenser		

The Hydra DT microdispenser is warranted against defects in material and workmanship for one year from the date of installation. Parts subject to normal wear, such as syringes and PTFE[®] tips, are excluded.

Do not try to service or repair a Hydra DT microdispenser under warranty before first contacting your equipment provider. See page 1-9 for contact information.

If service is required after the warranty period, it is still advisable to consult with your equipment provider before performing the service task, especially when the repair may be technically sophisticated or difficult.

If you need to return the microdispenser or parts of the microdispenser to Matrix Technologies for any reason, you must do the following:

1. Contact your equipment provider and obtain return approval and a Return Authorization Number. A decontamination form can be downloaded from *www.matrixtechcorp.com*.

IMPORTANT: Fill out the IRR Form completely. Describe on the form what applications the instrument has been used for and what materials have been used with the instrument, including any hazardous chemicals, radioisotopes, or biohazard materials. You must also describe how the instrument has been cleaned and inspected prior to its return and list the name and number of the person who performed the cleaning and inspection.



Caution: Ensure that the completed IRR Form is included in the return package. Do NOT return the microdispenser or its parts without this completed form.



- 2. Do not return any of the accessories unless you believe there is a problem with one of them.
- 3. Package the instrument in its original packing materials. All equipment being returned to the factory for any reason must be shipped in the original shipping containers and packaging. If you do not have the original shipping containers and package, you must purchase them and pack the merchandise in it. To order the packaging, see the current price list at the Matrix Technologies website at *http://www.matrixtechcorp.com/*.
- 4. When you pack the unit, ensure that the syringe pistons are in the full down position.
- 5. Ship the returned material by 2-day air service.

The contents of this manual may change as new features and accessories are added. By mailing the self-addressed and postage-free Warranty Card that you received with your Hydra DT microdispenser, you will receive updated information about the microdispenser. Your comments and suggestions for improving the performance and versatility of the Hydra DT microdispenser are always welcome and appreciated.

Warranty vii



Packing List

The **Hydra DT Base Unit Microdispenser** (Item numbers: 109610, 109620, 109630, 109640, 109611, 109621, 109631, 109641, 109625, 109535, 109645, 138410, 138411) is supplied with these items:

- (1) Operator's Manual
- (1) ControlMate Software CD
- (1) Power Cord
- (1) Emergency Shutoff Switch
- (1) RS 232 cable
- (1) Adapter for RS232 to 9 pin serial port
- (1) Adapter for USB to serial port
- (1 ea) Extra Fuses: 0.5 amp slow blow (for motor power supply); 3 amp; 1.6 amp

If your system includes the 3-Position X/Y Plate stage, these items are also included:

- (1) 3-Position X/Y stage with connecting cables (nest and stage) (item #1096-DT-XY3)
- (1) 9/64 Allen wrench
- (1) Phillips screwdriver

If your system includes an eDrop system, these items are also included:

- (1) eDrop TM single-channel dispenser fluidics component with power cord
- (1) eDrop wash/aspirate module (attached to stage)
- (1) Z-positioner for single-channel dispenser
- (1) Tubing set for gas and liquid (includes 5/32-inch fitting and tubing for the air-supply regulator.
- (1) Replacement nano needles, set of 2
- (1) Replacement nano tubing
- (1) Serial cable, 9-pin
- (1) USB adapter
- (1) CD ROM with USB driver

If any of these items are missing when you first unpack your system, contact your equipment provider.

See page 1-9 for contact information.



Warnings and Cautions

Stopping the System



WARNING! To shut down the unit IMMEDIATELY in case of an emergency, use the Emergency Shutoff Switch:

1. Firmly press the red button on the Emergency Shutoff Switch (see Figure i).



Figure i: Emergency Shutoff Switch

- 2. If you are running the system with the software, exit from the software.
- 3. To restore power to the microdispenser: First put an empty reservoir or plate on the tray table or stage under the dispensing head. Then rotate the red button in the clockwise direction, as shown on the arrows on the button. (When you turn the system on, the syringe plungers will descend to their lowest point to empty the tips and the tray table or stage will move down to the zero point.)
- To turn off the system when you are finished using it for the day:
 - 1. Press the power switch on the rear of the Hydra DT microdispenser to **OFF**. See Figure ii.



Figure ii: Power Module

4. If you are running the system with the software, **Save** and **Close** the Sequence file and **File-Exit** to close the ControlMate interface. The computer can now be shut down.

Formatted: Keep with next, Keep lines together

Warnings and Cautions ix



Protecting the Operator

Caution: Before you perform any microdispenser service or maintenance task that does not require you to run the microdispenser, turn the microdispenser off by pressing the power switch to **OFF**. Then unplug the power cord.

Protecting the System from Damage



Caution: In general, do not allow the disposable tips to contact the bottom of any plate, tube, reservoir, tray, or wash basin when you are filling, or emptying or when you are setting tray table heights. The only time the tips need to touch the bottom of the plate is when they are dispensing volumes of 3μ or less onto a dry plate. For dispense volumes greater than 3μ L, the size of the droplet formed on the tip will be large enough to bridge the gap between the tip and the bottom of the plate during a dispense with volume greater than 3μ L, backpressure will build up in the syringes, because the sample cannot exit the tips as fast as the syringe plunger is pushing the sample out. That pressure can damage the syringes. If a syringe is damaged during operation, turn off power to the unit immediately by pressing the **Emergency Shutoff Switch**. See page 6-9 for more information about heights for small-volume dispenses.



Caution: Use a power strip with surge protection with the microdispenser. Although the printed circuit board is protected, noisy AC lines can cause the unit to behave erratically or can blow the fuse. The circuitry within the microdispenser is sensitive to sudden voltage fluctuations that result from a surge in electrical power. Those fluctuations can be common in facilities with older electrical systems. They can also occur frequently during lightning storms.



Caution: Minimize or eliminate the use of appliances on the same electrical line as the microdispenser. Fluorescent light power supplies (such as those used with microscopes), high-current heaters, and cycling electrical motors used on the same electrical line as this unit can cause the unit to behave erratically.



Caution: The printed circuit board might be charged with static electricity. Ensure that you are properly grounded before you touch the printed circuit board.



Caution: Do not allow corrosive or oxidizing agents, such as chlorine bleach, strong acid, etc., to be in continuous contact with the microdispenser. Such agents can be used routinely with the Hydra DT microdispenser but may damage the case, the tray table, and/or the glass syringes if left in contact with the instrument for extended periods of time. If you need to use these reagents in a protocol, ensure that you clean the microdispenser of any residual corrosive agent as soon as you finish the procedure. If you need to use any strong oxidizing or corrosive agent other than those listed in Table 6-9 on page 6-26,



contact your equipment provider for assistance. See page 1-9 for contact information.



Caution: On a Hydra DT system that includes the X/Y Plate Stage, do not allow the stage to strike its X or Y stop points with excessive force. This may damage the stage. Save the shipping plates and all screws. These plates are used to keep the two motorized sections of the positioner immobile during shipment.

How to Use This Manual

This manual is both a user's guide and a reference manual. You can read through the manual from beginning to end to learn about the system and practice using it. You will get an introduction to the Hydra DT microdispenser and then learn how to set up the hardware, program file parameters, check system performance, service and maintain the system, and use the system most effectively.

Chapters 3 and 4 contain "getting started" information. You can use them to practice setting up files and running procedures.

You can also use the tables, diagrams, figures, procedures, and headings (as well as table of contents and index) to access just the piece of information you need.

How to Use This Manual xi



1: Introduction to the Hydra DT Microdispenser

Overview of the Hydra DT Microdispenser	1-1
Hydra DT Base Unit	
Optional Accessories	
Hydra DT Microdispenser Model Types	
Syringe Design and Construction	
DART Magazine Construction	
Related Products	
Contact Us with Questions and Comments	

Overview of the Hydra DT Microdispenser

The Hydra DT microdispenser from Matrix Technologies is an addition to the Hydra line of bench-top microdispensers that provide high-speed aspirating and dispensing into 96-well or 384-well microplates.

The Hydra DT uses DART (Disposable Automation Research Tips) disposable magazines for quick, single-hand, tip loading. The tips in the rigid DART magazines are sealed against a silicone pad on the underside of the pipetting head and locked in place with an easy-grip handle. The pad forms an even and airtight seal that minimizes tip-to-tip height variations and ensures consistent dispensing into all microplate wells.

The Hydra DT base unit has a motorized platform (a "tray table") that moves up and down (Z-axis movement), allowing you to position microplates and other labware under the disposable tips. (If you add the optional X/Y Plate Stage to the base unit, the tray table is replaced with a 3-position stage that moves in the X Y and Z (L,R.F,B, Up) directions.

You can program the Hydra DT microdispenser to perform basic liquid transfer tasks, including aspirate, dispense, mix, and empty, by using the touch pad or the *ControlMate* software that is included with the basic unit. You can program the positioning of the tray table or stage to move labware toward and away from the pipetting head.

When you program the microdispenser using the touch pad, each program is stored in one of 16 selectable files that reside on the microdispenser; no separate computer is required.

If you are planning to use the included *ControlMate* software to create and run programs, you will need to install it on your own computer, with the system running under Microsoft[®] Windows[®] 95 or later operating system.

Introduction to the Hydra DT Microdispenser

1-1



You can also use the communications protocol, provided in Appendix D, to integrate the workings of the Hydra DT microdispenser into an automated laboratory.

Hydra DT Base Unit

The Hydra DT base unit microdispenser appears in Figure 1-1.



Figure 1-1: The Hydra DT Microdispenser



The Hydra DT with single stage and PC in Figure 1-2.



Figure 1-2: Hydra DT system

Optional Accessories

The 3-Position X/Y Plate Stage can be added to any Hydra DT base unit. It provides a programmable stage for positioning microplates and other labware under the pipetting head. See Figure 1-3. The Hydra DT with X/Y Plate Stage requires a computer with *ControlMate* software for operation. Users must calibrate the stage, then program stage positions for their laboratory protocols.



Figure 1-3: The 3-position X/Y Plate Stage

The eDrop single-channel nano-volume dispenser, complete with single-needle wash, can be added to any Hydra DT system. The eDrop dispenser requires the purchase of the 3-position X/Y Plate Stage accessory (1096-DT-XY3). See Appendix E for setting up and using the Hydra eDrop System.

The *ControlMate* OLE ActiveX COM automation interface provides additional functionality to the Hydra DT system. This interface was developed for use with Microsoft Visual Basic® versions 5 and 6.

Introduction to the Hydra DT Microdispenser





Hydra DT Microdispenser Model Types

The Hydra DT microdispenser is available with a 96-well or 384-well pipetting head. The *ControlMate* software is included with every Hydra DT unit.

Optional accessories for the base unit include the 96 to 384 manual plate positioner, X/Y plate stage and eDrop module. An optional software component, *ControlMate* OLE, provides an ActiveX^{®1} COM automation interface.

Table 1-1 lists the base-unit models, and Table 1-2 lists the optional add-ons.

Table 1-1: Hydra DT Microdispenser Base Unit Models

Catalog No. Description		Volume range	Compatible tip types
1096-DT-100	Hydra 96 DT-100 base unit, compatible with 96-channel 30ul and 100ul DARTs	0.5ul to 100ul	5506, 5526, 5586
1096-DT-300	Hydra 96 DT-300 base unit, compatible with 96-channel 300ul DARTs	5.0ul to 300ul	5516, 5536, 5546
1384-DT-100	Hydra 384 DT-100 base unit, compatible with 384-channel 30ul and 100ul DARTs	0.5ul to 100ul	5416, 5316, 5326

Table 1-2: Hydra DT Replacement Pipetting Heads

Catalog No.	Description	Volume range	Compatible tip types
1096-RH-100	Replacement pipetting head for the Hydra 96 DT-100	0.5ul to 100ul	5506, 5526, 5586
1096-RH-300	Replacement pipetting head for the Hydra 96 DT-300	5.0ul to 300ul	5516, 5536, 5546
1384-RH-100	Replacement pipetting head for the Hydra 384 DT-100	0.5ul to 100ul	5416, 5316, 5326

Table 1-3: Optional Add-ons for Hydra DT Systems

Hydra DT Model	Catalog No.	Description
3-Position X/Y Plate Stage	1096-DT-XY3	Programmable stage for positioning microplates and other receptacles the along X, Y, and Z-axes
eDrop, Single-Channel Non-Contact Nano Volume Dispenser	10961N1	Single-channel nano-volume dispenser. Requires purchase of X/Y Plate Stage accessory (10961XY2 or 10961XY3).
ControlMate OLE software component	501-30023	<i>ControlMate</i> OLE ActiveX COM automation interface, to be used with the <i>ControlMate</i> device-control software. Developed for use with Microsoft Visual Basic® versions 5 and 6.

¹ Active X is a registered trademark of Microsoft Corporation.



Syringe Design and Construction

The syringes manufactured for the Hydra DT microdispensers are specially designed to facilitate reproducible, low-volume dispensing. The parts of the syringe setup are:

- Glass syringe barrel: high-quality borosilicate glass, with inner chambers that are precision-machined and highly polished to ensure leak-free performance
- Plunger: stainless steel
- PTFE plunger tip: fitted on the end of the plunger, with ribbing to prevent leaking around the tip under pressure

The removable PTFE tips (for replacement when necessary) on the plungers are included on all syringes.

See Appendix A for a tabular list of the syringe types. See Appendix B for information about how to order replacement syringes and how to get syringes refurbished.

Introduction to the Hydra DT Microdispenser

1-5



DART Magazine Construction

The DART tips are designed to seal tightly against the plate head to ensure accurate and consistent volume delivery. See Figure 1-4: DART Magazine Construction.



Figure 1-4: DART Magazine Construction

Table 1-4: DARTS types by Hydra DT un	DIE 1-4: DAF	{Is typ	es by F	iydra I	וט	uni
---------------------------------------	--------------	---------	---------	---------	----	-----

Catalog number	Description	Туре	Volume range	Compatible Instruments
5526	100ul, 20 disposable magazines of 96 per case	Non-sterile	0.5ul to 100ul	Hydra DT 96/100, PMP, PM2x2
5527	100ul, 20 disposable magazines of 96 per case	Sterile	0.5ul to 100ul	Hydra DT 96/100, PMP, PM2x2
5528	100ul, 20 disposable magazines of 96 per case	Filtered sterile	0.5ul to 100ul	Hydra DT 96/100, PMP, PM2x2
5586	30ul, 20 disposable magazines of 96 per case	Non-sterile	0.5ul to 30ul	Hydra DT 96/100, PMP, PM2x2
5587	30ul, 20 disposable magazines of 96 per case	Sterile	0.5ul to 30ul	Hydra DT 96/100, PMP, PM2x2
5588	30ul, 20 disposable magazines of 96 per case	Filtered sterile	0.5ul to 30ul	Hydra DT 96/100, PMP, PM2x2
5506	30ul Extended Length, 20 disposable magazines of 96 per case	Non-sterile	1.0ul to 30ul	Hydra DT 96/100, PMP, PM2x2
5507	30ul Extended Length, 20 disposable magazines of 96 per case	Sterile	1.0ul to 30ul	Hydra DT 96/100, PMP, PM2x2
5508	30ul Extended Length, 20 disposable magazines of 96 per case	Filtered sterile	1.0ul to 30ul	Hydra DT 96/100, PMP, PM2x2



Catalog number	Description	Туре	Volume range	Compatible Instruments
5517	300ul, 20 disposable magazines of 96 per case	Sterile	5.0ul to 300ul	Hydra DT 96/300, PMP, PM2x2
5518	300ul, 20 disposable magazines of 96 per case	Filtered sterile	5.0ul to 300ul	Hydra DT 96/300, PMP, PM2x2
5536	300ul extended length, 20 disposable magazines of 96 per case	Non-sterile	5.0ul to 300ul	Hydra DT 96/300, PMP, PM2x2
5537	300ul extended length, 20 disposable magazines of 96 per case	Sterile	5.0ul to 300ul	Hydra DT 96/300, PMP, PM2x2
5538	300ul extended length, 20 disposable magazines of 96 per case	Filtered sterile	5.0ul to 300ul	Hydra DT 96/300, PMP, PM2x2
5546	300ul wide bore, extended length, 20 disposable magazines of 96 per case	Non-sterile	5.0ul to 300ul	Hydra DT 96/300, PMP, PM2x2
5547	300ul wide bore, extended length, 20 disposable magazines of 96 per case	Sterile	5.0ul to 300ul	Hydra DT 96/300, PMP, PM2x2
5548	300ul wide bore, extended length, 20 disposable magazines of 96 per case	Filtered sterile	5.0ul to 300ul	Hydra DT 96/300, PMP, PM2x2

Hydra DT for 96-well, 300ul (1096-DT-300)

Hydra DT for 384, 100ul (1384-DT-100)

Catalog number	Description	Туре	Volume range	Compatible Instruments
5416	30ul extended length, 20 disposable magazines of 384 per case	Non-sterile	0.5ul to 30ul	Hydra DT 384/100, PMP, PM2x2
5417	30ul extended length, 20 disposable magazines of 384 per case	Sterile	0.5ul to 30ul	Hydra DT 384/100, PMP, PM2x2
5418	30ul extended length, 20 disposable magazines of 384 per case	Filtered sterile	0.5ul to 30ul	Hydra DT 384/100, PMP, PM2x2
5316	30ul, 20 disposable magazines of 384 per case	Non-sterile	0.5ul to 30ul	Hydra DT 384/100, PMP, PM2x2
5317	30ul, 20 disposable magazines of 384 per case	Sterile	0.5ul to 30ul	Hydra DT 384/100, PMP, PM2x2
5318	30ul, 20 disposable magazines of 384 per case	Filtered sterile	0.5ul to 30ul	Hydra DT 384/100, PMP, PM2x2
5326	100ul, 20 disposable magazines of 384 per case	Non-sterile	1.0ul to 100ul	Hydra DT 384/100, PMP, PM2x2
5327	100ul, 20 disposable magazines of 384 per case	Sterile	1.0ul to 100ul	Hydra DT 384/100, PMP, PM2x2
5328	100ul, 20 disposable magazines of 384 per case	Filtered sterile	1.0ul to 100ul	Hydra DT 384/100, PMP, PM2x2

When a wash sequence using the Wash Pump Module is started, Fill pump 1 fills the Wash Reservoir, and then the tray table or stage rises. On a system without the stage, the tips pass through slots in the tray table and into the reservoir. On a system with the stage, the stage moves away from the front of the microdispenser so that the top

Introduction to the Hydra DT Microdispenser

1-7



Related Products

Matrix Technologies products that you might use with your Hydra DT microdispenser system include:

Table 1-3: Related Products

Product	Description	
TrakMates®	2-D Barcoded Tubes	
V-Groove Automation Reservoirs	96, 96-Deep, 384	
Reservoirs	8 or 12-Channel	
ScreenMates®	96, 384, 1536 Well Plates	
Tube Chiller	1 and 3-Position	

For more information about these and other Matrix Technologies products, contact your equipment provider (see page 1-9 for contact information) or visit *http://www.matrixtechcorp.com*. See Appendix B for more information about Hydra DT microdispenser replacement parts and accessories.



Contact Us with Questions and Comments

Send us the self-addressed, postage-paid Warranty Card included with the Hydra DT microdispenser to receive updated information about your Hydra DT microdispenser system.

Your comments and suggestions for improving the performance and versatility of the Hydra DT microdispenser system and this manual are always welcome and appreciated.

If you have any questions or comments about your Hydra DT instrument, contact your equipment provider. Some of those contacts are listed on this page. If your instrument was provided by a distributor not listed here, contact Matrix Headquarters.

Corporate Headquarters

22 Friars Drive Hudson, NH 03051, USA Toll free: (800) 345 0206 Tel: (603) 595 0505 Fax: (603) 595 0106 Email: info@matrixtechcorp.com

European Headquarters

Lower Meadow Road Brooke Park Handforth, Wilmslow Cheshire SK9 3LP, UK Freefone: 0800 389 4431 Tel: +44 161 486 2110 Fax: +44 161 488 4560 Email: info@matrixtechcorp.eu.com

French Office

15 Rue Des Pas Perdus 95804 Cergy Saint Christophe BP83338 France Tel +33-134-2544-42 Fax +33-134-25-44-00

German Office

Vogelsangstrasse 12 D-61273 Wehrheim, Germany Tel: +49 6081 981878 Fax: +49 6081 981959

Swiss Office

Seedammstrasse 3, Postfach CH-8808, Pfaffikon SZ Tel: +41 55 417 4546 Fax: +41 55 417 4647

Asia-Pacific Office Japan Email: asia@matrixtechcorp.com

Introduction to the Hydra DT Microdispenser





2-1

2: Setting up the Hardware

Prepare the Site	2-1
Unpack the Microdispenser	2-1
Moving the Microdispenser	2-2
Set Up the Stage (Hydra DT with X/Y Plate Stage Only)	2-2
Change the Voltage Setting for the Microdispenser If Necessary	2-3
Connect Power Cords and Cables; Turn On the System	2-5
Configure hardware options	2-7
Configure XY stage	2-7
Connect the Computer	2-8

Prepare the Site

- 1. Clear adequate space on your workbench for the microdispenser and all other system components.
- 2. Ensure that the setup allows the microdispenser to be plugged into a surge suppressor and that electrical noise is reduced or eliminated. Electrical noise generated by appliances such as fluorescent lights, motors, and heaters can cause the microdispenser to behave erratically and can even damage the microdispenser.



Note: If your system includes a Hydra eDrop module, see Appendix E for setup information.

Unpack the Microdispenser

- 1. Make sure that the box or boxes are right side up.
- 2. Carefully open each box and remove any foam packing surrounding the pieces.



Caution: Do NOT grasp the microdispenser by the pipetting head handle. Grasp it by placing one hand firmly on either of its sides, with your fingers in the finger slots, and then slowly lift it straight up out of the box.

- 3. Lift all pieces out of the box or boxes and gently place the pieces on the workbench, with the front of the microdispenser facing you.
- 4. Check the contents of the box or boxes against the packing list. Contact your equipment provider if pieces are missing. See page 1-9 for contact information.

Setting up the Hardware



Moving the Microdispenser



Caution: Always place your fingers in the finger slots, one on each side of the microdispenser, before you lift the instrument to move it. Do **NOT** grasp the microdispenser in any other way before you move it. If your system includes the stage, NEVER lift the system by the stage.



Note: If your system includes the stage and you have already attached and calibrated the stage, you will need to check the calibration and possibly recalibrate the stage after you move the system. (Instructions for calibrating the stage begin on page 4-21.)

Set Up the Stage (Hydra DT with X/Y Plate Stage Only)

Have these items available:

- Allen wrench for hex screws (provided with system)
- Phillips screwdriver
- 1. Place the X/Y Plate Stage so that the two holes in the mounting bracket align with the two pins in the mounting plate on the Hydra DT microdispenser. Secure the stage to the microdispenser with the 4 hex screws using the Allen wrench. See Figure 2-1.



Figure 2-1: Mounting Stage onto Microdispenser

2. Gradually tighten each screw so that uniform tension is applied. Do not over tighten.



3. Remove the metal shipping plates on the front and side of the stage with a Phillips-head screwdriver. (See Figure 2-2.) The stage should now move easily side to side and in and out.



Figure 2-2: Removing Shipping Plates



Caution: Do not allow the stage to strike its X or Y stop points with excessive force. Such strikes might damage the stage. Save both shipping plates and all screws. These plates are used to keep the two motorized sections of the stage immobile during shipment.

4. Change base module to the X Y Stage on the *STOP/PAGE* screen. See "Configure XY stage" on page 2-7.

Change the Voltage Setting for the Microdispenser If Necessary

The microdispenser can operate on either 115-volt power or 230-volt power. You can change the setting, if necessary, as follows:

Have these items available before you begin:

- Flat blade screwdriver
- Two fuses for the voltage you will be using (115V or 230V)
- 1. If the system is already connected and plugged in: Turn off power to the system and then unplug the power cord from the back of the system and from the surge suppressor.
- 2. Put the tip of the screwdriver in the small slot at the top of the power module on the back of the microdispenser and gently pry open the cover on the power module.

Setting up the Hardware





fuse cover



Figure 2-3: Power Module

3. Put the tip of the screwdriver in the slot at the top of the red square (the end of the fuse box) and gently pry the fuse box end away from the microdispenser until you can grasp it with your fingers.



Figure 2-4: Removing the Fuse Box

- 4. Pull the fuse box out and remove the two fuses.
- 5. Insert the new fuses.



Note: If you are inserting 230 fuses, you might need to use the end of the screwdriver to gently push the silver metal tab inside the fuse space toward the end of the fuse box. Sometimes the tab is bent slightly toward the middle of the box, and that tab placement can keep the 230 fuses from being correctly inserted.

6. Re-insert the fuse box in the power module with the number for the voltage you are using (for example, $115\overline{V}$) right side up so that you can read it.



2-5



Figure 2-5: Inserting the Fuse Box With 115V Setting

- 7. Close the power-module cover.
- 8. If the system was already connected and plugged in: Plug the surge suppressor into the new power source, and then plug the power cord into the system and into the surge suppressor.

Connect Power Cords and Cables; Turn On the System

1. Plug one end of the power cord into the power module on the rear of the unit. See Figure 2-6.



Figure 2-6: Back of Microdispenser

2. Plug the other end of the cord into a surge suppressor.

Setting up the Hardware





Caution: You must use a power strip with a surge protector to protect the microprocessors used to control the unit. Install the Emergency Shutoff Switch.



Note: The Emergency Shutoff Switch must be installed for you to operate the unit. The unit will not operate without the switch.

- a. Plug the connector on the cable of the Emergency Shutoff Switch into the receptacle on the back panel of the microdispenser. See Figure 2-7.
- b. Secure the cable by turning the connector clockwise.



Figure 2-7: Attaching E-stop plug



Note: Before you turn the system on, ensure that an empty reservoir or plate is on the tray table/stage directly underneath the syringe head. When the system is powered on, the tray table or stage will move to its lowest (Z-axis down) position, and then the syringe plungers will automatically move to the empty position (plungers down), and the syringes will empty.

c. Test the Emergency Shutoff Switch: Press the red button on the switch to turn it ON. (See Figure 2-8.) Press the power switch on the back of the microdispenser to turn the system ON. Then press the red button on the switch to shut the system OFF. Restart the system by rotating the red button in the clockwise direction, as shown by the arrows on the button.



Figure 2-8: Emergency Shutoff Switch

5. If the stage is part of your system, connect the stage cable between the computer and the STAGE connection on the back of the microdispenser.



Configure hardware options

The XY stage and the wash pump module are both optional accessories; therefore the Hydra base unit must be appropriately configured to work with them. See the following sections.

Configure XY stage

1. Press and hold the **STOP** button, then press the **PAGE** button to display the following screen.



- 2. Press the **EDIT** button to scroll to the XY setting.
- 3. Press the **UP** or **DOWN** ($\blacktriangle \nabla$) buttons to toggle the XY stage option on and off.
- 4. After configuring your Hydra system, press the **PAGE** button to exit the set up screen. You will see the following screen.



5. Press the **ESC** button to return to the main screen.

Setting up the Hardware





Connect the Computer

(If You Added a Computer to Your System Setup)

If you are using your own Windows computer to run the ControlMate software, connect the computer to the back of the microdispenser with the components in Figure 2-9 in the following sequence:

- 1. Computer to USB cable
- 2. USB cable to serial port adapter
- 3. Serial port adapter to Ethernet adapter
- 4. Ethernet adapter to Ethernet cable
- 5. Ethernet cable to Hydra DT



Figure 2-9: Communication cables and adapters Miscellaneous Hardware Tasks

(Optional)

- If you are using *ControlMate* OLE software for your system to install it on your own Windows computer, see "1. Install *ControlMate* software if it is not already installed" on page 4-4.
- If you want to connect your microdispenser to a computer in your workplace to automate some microdispenser functions, see Appendix D, "Integrating the Hydra DT Microdispenser System into an Automated Laboratory."
- If you are interested in finding out whether you can change the syringe size on your Hydra DT microdispenser to another size, contact your equipment provider. See page 1-9 for contact information.



3: Getting Started: Manual Operation

Two Operation Methods	3-1
Steps for Learning Manual Operation of the Hydra DT Microdispenser	
1. Study the SAFETY Instructions	3-2
2. Look at flowcharts of Hydra DT interface	3-2
3. Read the table of file-parameter definitions and values	3-4
4. Map the microdispenser buttons	3-5
5. Read the table of microdispenser operations	3-6
6. Set up a tray table/stage height	3-7
7. Set file parameters	3-9
9. Run a procedure	3-11

Two Operation Methods

The two operation methods for creating and editing files and running procedures on the Hydra DT microdispenser are:

- Manual Operation—Use buttons on the front of the micro-dispenser to edit EPROM files (up to 16) that contain settings for Dispense, Aspirate, Empty, and Wash operations. Then use buttons, also on the front of the microdispenser, to execute those operations. The files are automatically saved on the microdispenser. No computer interface is needed.
- Automated Operation—Use *ControlMate* software installed on the Windows computer to calibrate labware and create, edit, run and save "sequence files" (pipetting programs). If your system includes the 2-Position X/Y Plate Stage, you also use the software to calibrate the stage.

This chapter describes the Manual Operation method. Chapter 4 describes the Automated Operation method.



Note: You can use both methods on a system that includes a Windows computer with *ControlMate* software. If your system setup does NOT include the computer and software, then you will use the Manual Operation method exclusively.

Getting Started: Manual Operation





Steps for Learning Manual Operation of the Hydra DT Microdispenser

The following flowchart shows the steps this chapter will walk you through to learn how to use the Manual Operation method.



1. Study the SAFETY Instructions

Review all Warnings and Cautions for use of the Hydra DT microdispenser, starting on page ix.

2. Look at flowcharts of Hydra DT interface

Diagram 3-1 shows the screens that you will see, in this order, in each of the 16 files stored on the microdispenser. You do not have to "save" the files, because the changes you make to the settings are automatically saved as you make them.



Diagram 3-1: Hydra DT Interface



Getting Started: Manual Operation

3-3



3. Read the table of file-parameter definitions and values

Table 3-1 lists the allowable values for the parameters you will set in your files.

Note: Each file can only contain one instance of a particular parameter. If your procedure requires multiple instances of an operation (Aspirate, for example) with different values for the different instances, you will need to use more than one file to run your procedure.

Screen	Parameter	Possible Values	Description	
Ready Mode	File	1 – 16	File number.	
DISPENSE	Vol	100μL: 0.1 – 110	Dispense Volume. Volume of air or solution to dispense. Increases or decreases by 0.1/0.5/1.0µL (varies with syringe size).	
		290µL: 0.5 – 290		
	Table	0 – 3970	Dispense Height. Height of tray table/stage when solution or air is dispensed. Increases or decreases by 10 units.	
	Speed	1–5	Speed. 5 pre-set speeds for syringe-plunger motor.	
ASPIRATE	Prime	ON/OFF	Prime. A pre-set* (not user-selectable) volume of air or solution will be aspirated and then dispensed back into source plate. Primes syringes before dispense. (*Pre-set volume is 5% of total syringe volume.)	
	Vol	100µL: 0.1 – 110	Aspirate Volume. Volume of air or solution to	
		290µL: 0.5 – 290	aspirate. Increases or decreases by 0.1/0.5/1.0µL (varies with syringe size).	
	Air	100μL: 0 – 110	Air Gap. Volume of air to draw into syringes before	
		290µL: 0 – 290	aspiration. Increases or decreases by 1.0. See page	
			6-5 for information about when to use an air gap.	
	Table	0 – 3970	Aspirate Height. Height of tray table/stage when solution or air is aspirated. Increases or decreases by 10 units.	
	Speed	1–5	Speed. 5 pre-set speeds for syringe-plunger motor.	

Table 3-1: Hydra DT File Parameters



4. Map the microdispenser buttons

The buttons on the microdispenser will become familiar to you quickly. Here's the layout, to get you started:



Figure 3-1: Buttons on Operation Screens (Screens = Empty, Aspirate, Page, and Edit)



Figure 3-2: Buttons on Ready Mode Screen



Figure 3-3: Buttons on Either Side of Syringe Array (STOP and Dispense)

Getting Started: Manual Operation

3-5



5. Read the table of microdispenser operations

Table 3-2 describes how to use the microdispenser buttons.

Operation/ Button	Description
▼ [Down]	Press Down to decrease values for selected parameter (selected parameter appears on top line of screen).
▲ [Up]	Press Up to increase values for selected parameter (selected parameter appears on top line of screen).
Empty	To start operation: <i>Ready Mode</i> screen must be displayed, with current syringe volume appearing in display. [Caution: If "Volume ??" appears on <i>Ready Mode</i> screen, always empty the syringes before proceeding with any other operation.] Press Empty to start Empty operation. Tray table/stage rises and syringes empty. Volume shown in display is updated and tray table/stage returns to home position. To interrupt operation, press STOP . Tray table/stage returns to home position.
Aspr	To start operation: <i>Ready Mode</i> screen must be displayed, with current syringe volume appearing in display. Press Aspr to start Aspirate operation. If Prime is set to ON, a pre-set (default) "overshoot" volume* is aspirated and then dispensed back into the source plate. (*Overshoot volume is 5% of syringe volume). If Air is set, an air gap (volume of air) is aspirated into syringes. Tray table/stage does not move when Air is aspirated. Tray table/stage rises and Vol is aspirated. Volume in display is updated and tray table/ stage returns to home position. To interrupt operation, press STOP . See the discussion beginning on page 6-2 for detailed information about when to use Air.
Page	Press Page to scroll through screens of a file (screens are in a continuous loop).
Edit	Press Edit to move from one parameter in a screen to next parameter in same screen. Selected parameter appears on top line of screen.
STOP (to left of syringe array)	Press STOP to stop current operation.
DISP (to right of syringe array)	To start operation: <i>Ready Mode</i> screen must be displayed, with current syringe volume appearing in display. Press DISP to start Dispense operation. Tray table/stage rises and Vol is dispensed. Volume in display is updated and tray table/ stage returns to home position. To interrupt operation, press STOP .

Table 3-2: Hydra DT Microdispenser Operations

Now that you have learned how to use the buttons, you can practice setting tray table heights. Although setting tray table heights is part of the general practice of setting file parameters, it is introduced separately here so that you can practice this technique before setting up the rest of a file.


6. Set up a tray table/stage height

Most microdispenser operations (Dispense, Aspirate, Empty) include movement of the tray table/stage. Therefore, it is very important that you know how to set the tray table/stage heights for those operations so that neither operator nor instrument is at risk when the microdispenser is operating.



Caution: In general, do not allow the tips to contact the bottom of any plate, tube, reservoir, tray, or washbasin. The only time the tips need to touch the bottom of the plate is when they are dispensing volumes of 3μ L or less onto a dry plate. For dispense volumes greater than 3μ L, the size of the droplet formed on the tip will be large enough to bridge the gap between the tip and the bottom of the plate. In such cases, set the dispense height at 0.2 - 0.3mm back from touching the plate. If the tips touch the plate during a dispense with volume greater than 3μ L, back-pressure will build up in the syringes, because the sample cannot exit the tips as fast as the syringe plunger is pushing the sample out. That backpressure can damage the syringes. If a syringe is damaged during operation, turn off power to the unit immediately by pressing the Emergency Shutoff Switch.

You can find out more about setting optimum dispense heights in Chapter 7, starting on page 6-9.

The following procedure describes how to set the syringes to a "near bottom" position.

Have two microplates or reservoirs available for the procedure.

Procedure 3-1: Set the tray table/stage height



Note: When the system is powered on, the tray table or stage will move to its lowest (Z-axis down) position, and then the syringe plungers will automatically move to the empty position (plungers down) and the syringes will empty.

- 1. Place an empty reservoir or plate on the tray table or stage directly under the dispensing head, for the syringes to empty into.
- 2. Press the power switch on the back of the microdispenser to ON.
- 3. After the syringes have emptied, remove the reservoir or plate from the tray table or stage.
- 4. Place an empty plate ("plate" = plate or reservoir) on the tray table or stage under the dispensing head.

Getting Started: Manual Operation





5. On the *Ready Mode* screen: press **Up** until File 14 appears in the middle of the screen.



Ready Mode screen with File 14 selected

- 6. Press **Page**. The *Dispense* screen appears.
- 7. On the Dispense screen: Press Edit until Table is on the top line of the screen.



Dispense screen with Table parameter selected

- 8. Press **Up** to move the table/stage up until the tips are just touching the bottom of the wells in the plate.
- 9. With your fingertips, gently tilt up the front edge of the plate, off the table/stage and toward the tips. If the plate lifts easily, the tips are not at near bottom. Repeat pressing **Up** (slowly) and testing closeness until you cannot lift the plate.
- 10. Press **Down** 2 or 3 times to bring the table/stage down slightly.

You have now set the Dispense Table (height) to a near-bottom position.

11. Press Page 5 times, to cycle back to the first Ready Mode screen in the file.



7. Set file parameters

Procedure 3-2, following, is an exercise that leads you through setting parameters in a file.

Before you start

- 1. Ensure that the Emergency Shutoff Switch is placed where you can reach it easily.
- 2. Review these Cautions before starting to set file parameters:



If a syringe is damaged, turn off power to the unit immediately by pressing the Emergency Shutoff Switch. See page 6-9 for more information about setting dispense heights for small-volume dispenses.

Procedure 3-2: Set file parameters

- 1. Place one of the empty reservoirs or plates on the tray table or stage directly under the dispensing head, for the syringes to empty into.
- 2. On the rear of the microdispenser, press the power switch to **ON**.



Note: *Firmware Version is shown on the display when you first turn the system on, in the format "V*xxx*" where xxx =Numerals of version number, no decimal point separator; e.g., V515 = Version 5.15.

- 3. After the syringes have emptied, remove the first reservoir from the tray table or stage and place the second empty plate ("plate" = plate or reservoir) on the tray table or stage under the dispensing head.
- 4. On the *Ready Mode* screen: Press **Up** until File 14 appears in the middle of the screen.



Ready Mode screen with File 14 selected

- 5. Press Page. The Dispense screen appears.
- 6. On the *Dispense* screen: If Vol is not on the top line of the screen, press **Edit** until it is. Press **Up** or **Down** to set vol.

Getting Started: Manual Operation







Dispense screen with Vol set to 10.0

- 7. Press **Edit** until *Table* is on the top line. Set Table to 3500 by pressing **Up** or **Down**.
- 8. Press Edit until Speed is on the top line. Press Up or Down to set Speed to 5.
- 9. Press Page. The Aspirate screen appears.
- 10. On the *Aspirate* screen: Press **Edit** until *Prime* is on the top line of the screen. If Prime is set to OFF, press **Up** or **Down** to toggle it to **ON**. (See the discussion beginning on page 6-8 for information about when and how to use Prime.)



Aspirate screen with Prime set to ON

- 11. Press **Edit** until *Vol* is on the top line. Press **Up** or **Down** to set Vol to 50.0/170.0 (varies with syringe size).
- 12. Press **Edit** until *Air* is on the top line. Press **Up** or **Down** to set Air to 0. (See page 6-9 for information about when to use an air gap.)
- 13. Press Edit until Table is on the top line. Press Up or Down to set Table to 2700.
- 14. Press Edit until Speed is on the top line. Press Up or Down to set Speed to 5.
- 15. Press Page. The *Empty* screen appears.
- 16. On the *Empty* screen: If *Table* is not on the top line of the screen, press **Edit** to put it there. Press **Up** or **Down** to set Table to 2500.





Empty screen with Table set to 2500

17. Press Edit until Speed is on the top line. Press Up or Down to set Speed to 5.

9. Run a procedure

Before you start

1. Review these Warnings and Cautions:



WARNING! To shut down the unit IMMEDIATELY in case of an emergency, press the red button on the **Emergency Shutoff Switch.**



Figure 3-4: Emergency Shutoff Switch



Caution: Do not allow corrosive or oxidizing agents, such as chlorine bleach, strong acid, etc., to be in continuous contact with the microdispenser. Such agents can be used routinely with the Hydra DT microdispenser but may damage the case, the tray table, and/or the glass syringes if left in contact with the instrument for extended periods of time. If you need to use these reagents in a protocol, ensure that you clean the microdispenser of any residual corrosive agent as soon as you finish the procedure. If you need to use any strong oxidizing or corrosive agent other than those listed in Table 6-9 on page 6-26, contact your equipment provider. See page 1-9 for contact information.

Getting Started: Manual Operation





Caution: Ensure that the tips are in the down position before you begin any microdispenser operation (Aspirate, Dispense, Wash, or Empty). Having the guide down ensures that each needle is aligned over the center of a well in the microplate on the tray table. If the guide is not down and you start an operation, some of the needles might get bent.



Caution: In general, do not allow the tips to contact the bottom of any plate, tube, reservoir, tray, or wash basin when you are filling, emptying, or washing the syringes or when you are setting tray table heights. The only time the **tips** need to touch the bottom of the plate is when they are dispensing volumes of 3μ L or less onto a dry plate. For dispense volumes greater than 3μ L, the size of the droplet formed on the tip tip will be large enough to bridge the gap between the tip and the bottom of the plate when the dispense height is set at two to three clicks back from touching the plate. If the **tips** touch the plate during a dispense with volume greater than 3μ L, backpressure will build up in the syringes, because the sample cannot exit the **tips** as fast as the syringe plunger is pushing the sample out. That pressure can damage the syringes. If a syringe is damaged during operation, turn off power to the unit immediately by pressing the Emergency Shutoff Switch. See page 6-9 for more information about heights for small-volume dispenses.

- 2. Review Table 3-2, on page 3-6; it describes how to use the microdispenser operation buttons.
- 3. Have the following labware and solutions ready:
 - 1 empty reservoir, to empty the syringes into
 - 5 empty plates or reservoirs (1 is a source plate and 4 are receiving plates)
 - dH₂O if you wish to run the procedure with solution (You can run it without any solution.)
- 4. If the Wash Pump Module is part of your system setup, ensure that the wash containers have water in them if you wish to use solution in the test run.

Procedure 3-3: Run a procedure

- 1. Ensure that the Emergency Shutoff Switch is placed where you can reach it easily.
- 2. If the system is turned off, place an empty reservoir on the tray table or stage under the dispensing head, and then turn the system on by pressing the power switch to **ON**. Remove the reservoir after the syringes have emptied.



Note: *Firmware Version is shown on the display when you first turn the system on, in the format "Vxxx" where xxx = Numerals of version number, no decimal point separator; e.g., V515 = Version 5.15.

3. Access File 14, our exercise file. Press **Page** to cycle through the file-parameter settings for the file to ensure that they are correct. Change settings if necessary.



Table 3-3 shows the settings that you entered in File 14, if you stepped through Procedure 3-2. If you did not do that exercise, you can enter these settings in File 14 now.

Screen	Parameter	Setting
Ready Mode File		14
Dispense	Vol	10.0/40.0/70.0/120.01
	Table	3500
	Speed	5
Aspirate	Prime	ON
	Vol	50.0/170.0/290.0/
	Air	0
	Table	2700
	Speed	5
Empty	Table	2500
	Speed	5
	Speed	5
Pumps3	P1	15
	P2	15

Table 3-3: Parameter Values Entered in File 14

- 4. If the system was already turned on when you began this procedure, now place an empty reservoir on the tray table/stage, under the dispensing head and then, in File 14, press **Empty**.
- 5. Remove the reservoir from the tray table/stage and replace it with the source reservoir or plate, one that either contains dH2O or is empty.
- 6. Press **Aspr**. The syringes aspirate and dispense the Prime amount and then aspirate the Vol amount of dH2O (or air).
- 7. Remove the reservoir/plate from the table/stage.
- 8. Place the first receiving plate onto the table/stage.
- 9. Press Dispense. The Vol of dH2O or air is dispensed into the plate/reservoir.
- 10. Repeat Steps 9 through 11 three times.
- 11. Remove the last receiving plate with the source plate.
- 12. Press Empty.

Getting Started: Manual Operation



4: Getting Started: Automated Operation

Two Operation Methods 4-1
Steps for Learning Automated Operation of the Microdispenser
1. Install ControlMate software if it is not already installed4-4
2. Study the SAFETY instructions
3. Read the overview of the software4-4
4. Check the software configuration4-13
5. Calibrate the stage (Hydra DT with X/Y Plate Stage systems only)
6. Create labware4-28
7. Create and edit sequence files4-39
8. Run a sequence file4-42
Command Reference
Main Sequence Editor4-46
Procedure Library4-51
ControlMate OLE Optional Add-On Software Component

Two Operation Methods

The two operation methods for creating and editing files and running procedures on the Hydra DT microdispenser are:

- Automated Operation—Use *ControlMate* software installed on the Windows computer to calibrate labware and create, edit, run and save "sequence files" (which are programs). For a Hydra DT with X/Y Plate Stage system, you also use the software to calibrate the stage.
- Manual Operation—Use buttons on the front of the micro-dispenser to edit EPROM files (up to 16) that contain settings for Dispense, Aspirate, Empty, and Wash operations. Then use buttons, also on the front of the microdispenser, to execute those operations. The files are automatically saved on the microdispenser. No computer interface is needed.

This chapter describes the Automated Operation method. Chapter 3 describes the Manual Operation method.



Note: You can use both methods on a system that includes a Windows computer with *ControlMate* software. If your system setup does NOT include the computer and software, then you will use the Manual Operation method exclusively.



Note: If your system is an eDrop System, see Appendix E for information on operating that system. It uses eDrop control software.





 ControlMate Online Help contains much of the information contained in this chapter, so you can refer to the help system directly while you are using the software. To access online help, do one of the following (italicized terms are defined in the overview later in this chapter):

 Click the Help icon in the Toolbar.

 Image: Select a help item from the Help menu in the Menu Bar.

 Image: Edit View Bun Tools Addding Window Help

 Click the Help button in any child dialog or command screen you are in.

 Help



Steps for Learning Automated Operation of the Microdispenser

The following flowchart shows the steps this chapter will walk you through to learn how to use the Automated Operation method.



Getting Started: Automated Operation



1. Install ControlMate software if it is not already installed

If you need to install the *ControlMate* software on your own Windows computer, or are installing an upgrade, follow these instructions:

- 1. Set up the computer close to the microdispenser.
- 2. Insert the software CD in your CD drive.
- 3. If the installation does not start automatically, follow these steps:
 - a. From the Windows Start menu, click **Run**.
 - b. In the Open field on the Run dialog, type:

```
<your_CD_drive_letter>:hydra.exe
```

- c. Click OK.
- 4. Follow the installation instructions given on the screen.
- 5. When installation is finished, restart your system.

2. Study the SAFETY instructions

Review all Warnings and Cautions for use of the Hydra DT microdispenser, starting on page ix.

3. Read the overview of the software

ControlMate is a Windows PC application that provides a graphical programming interface for creating and executing device module control programs. The configuration defines the device module in use. The device module described in this manual is the module for the Hydra DT Microdispenser.

The ControlMate overview on the next few pages describes:

- The *Main Application* screen elements:
 - Menu Bar
 - Toolbar
 - Toolbox
 - Status Bar
- The *ControlMate* command groups
- The *ControlMate* "child dialogs," which are the screens that allow you to perform actions within the software

The rest of this chapter, after the overview material, describes how to use the *ControlMate* tools to calibrate the stage (on Hydra DT with X/Y Plate Stage systems), create labware, and create and run programs (programs are called "sequence files" in *ControlMate* software).



The Main Application Screen

When you start the *ControlMate* software, the *Main Application* screen opens. All of the actions you take with the software will be executed with the *Main Application* screen open.



Menu Bar

Select software operations, such as "Open Sequence File," from the Menu Bar at the top of the Main Application window.







Menu	Function	Purpose
File	New Sequence Open Sequence	Create a new sequence. Open (to view, edit, or run) an existing sequence.
Edit	Cut Copy Paste Delete View Clipboard	Cut, copy, paste, and delete commands from sequence files. View the Clipboard to see what is currently on the Clipboard.
View	Toolbox Text Toolbox icon size	View text labels for icons in the Toolbox. Select size for the Toolbox icons.
Run	Validate File Start	Validate command parameters. Open the Execution program to run a procedure.
Tools	Options Connection Commands Limits Positions Plates Motors	Set communications parameters. View individual command settings. View default command-parameter limits. Set positions for stage locations. Create, add, modify, and delete labware. Change the speed of the x, y, and z axes motors.
Add-Ins	Set Device and Pipettor Options	Choose device type (Base model Hydra DT; 3-Position X/Y stage positioner) and syringe size/volume.
Window	Tile Horizontally Tile Vertically Cascade Arrange Icons	Arrange multiple open windows horizontally. Arrange multiple open windows vertically. Arrange multiple open windows in overlapping cascade. N.A.
Help	Contents Search About	Help topics listed in table of contents format. Help topics listed in Index format. Help version.

Table 4-1: Operations in the Menu Bar



Toolbar

Click icons in the Toolbar to start software operations, such as "Open Sequence File." The Toolbar offers easy access to the Menu Bar's most frequently used functions.

<u>F</u> ile	<u>E</u> dit	⊻iew	<u>R</u> un	Tools	Add-Ins	<u>W</u> indow	<u>H</u> elp				
	B			X 🖻		X Add	d Command	- 4		2	

Table 4-2: Operations in the Toolbar

Toolbar icon	Description
	Start a new sequence file.
1	Open an existing sequence file.
	Save the current sequence file.
I	Save all open sequence files.
¥	Cut the highlighted command and place it on the Clipboard.
	Copy the highlighted command onto the Clipboard.
1	Paste the contents of the Clipboard below the current command in the command tree.
2	View the Clipboard contents.
×	Delete the currently highlighted command.
Add Command	Add a new command below the current command in the command tree.
à	Print preview. View/print report of the current sequence file.
5	Validate the current sequence file. Check command values and relationship errors. Highlight an error and then display the command to make corrections.
	Run (execute) the current sequence file.
	View configuration and default options.
2	View Help screens.





Toolbox

Click icons in the Toolbox to insert commands in your sequence files. The Toolbox offers easy access to commands; those commands also appear in the Add Commands dropdown list in the Toolbar.



Table 4-3: Commands in the Toolbox

lcon	Command	Command Type	Description (NOTE: Detailed descriptions begin on page 4-45.)
p de	Call Procedure	Intrinsic (See page 4-10 for definition of command types.)	Calls a procedure from within current sequence file or from another sequence file. Saves time by reusing an existing procedure.
Y	Notepad		Creates a note in the sequence file. Does not execute an operation. Useful for documenting steps and for providing other commentary for user.
6	Group Commands		Groups commands together for a common purpose.
M	Execute External Application		Executes an external application.
B	Procedure		Executes a series of commands when the procedure is called from the main sequence.
	External Procedure File		Imports all procedures contained within an external sequence file into the current file.
Global	Global Value		Allows you to designate a command as "variable"



lcon	Command	Command Type	Description (NOTE: Detailed descriptions begin on page 4-45.)
رو Assign	Assign Global Value		Assigns a Global Value to an individual command in a protocol
export	Export Global Value		Export data to an external program (i.e. Barcode data)
	Aspirate	Module-Specific (See page 4-10 for definitions of	Aspirates a measured volume into syringes. Must be preceded by Move to Position.
	Dispense	command types.)	Dispenses a measured volume or all solution in syringes. Must be preceded by Move to Position.
*	Move to Position		Positions stage or tray table in specific location on the instrument.
	Home Axes		Resets stage or tray table to home position.
٢	Pause		Inserts a pause into a program (sequence file) to stop processing according to pause parameters.
- Colored	Wash Tips		Washes inside and outside of tips. Must be preceded by Move to Automation reservoir vessel
8	Mix		Aspirates and dispenses a measured volume in specified cycles to mix microwell content. Must be preceded by Move to Position.
	Message Display		Allows display of custom message on the device display panel.
	Pump Control		Allows direct control of wash pumps, independent of a wash routine. Must be preceded by a Move to Position command.
Ξ₩	Speed Control		Allows adjustment of piston (syringe-plunger) speeds for Aspirate, Dispense, Empty, and Wash operations.
∏ → 	Change DARTs tip magazine		Reminds the user to change the DART tip magazine during a sequence. Plate stage should be in the down position to allow clearance for moving the tips.

Click and drag the Toolbox to position it anywhere within the open Main Application window. You can "dock" it by positioning it against any side of that window or allow it to float on top of other window dialogs so that the icons are always visible.

You can set the size of the icons to *small* or *large* and view them with or without text labels. Select these options from the View menu.

Getting Started: Automated Operation



Status Bar

Open files (1) 😰 Clipboard active Auto correct: On Current file 'Corning 4 subwell protocol.cms'

The Status Bar provides information about what is currently running in the *ControlMate* window environment.

Open files-Number of currently open sequence files.

Clipboard active—Indicates whether the Clipboard contains some cut or copied material.

Auto correct—Indicates whether automatic correction of command errors is ON or OFF. Toggle this setting on and off in the Tools> Options>Limits. With Auto correct on, the software immediately checks a command for limit errors when you set the command in a sequence file. If it is off, individual commands are not checked for command errors until you validate the entire sequence file.

Current file—Name of the current open sequence file. If there is more than one sequence file open, this panel lists the topmost file.

Command Groups in ControlMate Software

The *ControlMate* software commands are divided into two groups:

- Intrinsic
- Module-specific ("Module" = device; your module is "Hydra DT.")

Intrinsic commands are specific to *ControlMate* rather than to a module. The commands are normally associated with the control of program flow and annotation. The intrinsic commands are:

- Main Sequence*
- Procedure Library*
- Call Procedure
- Notepad
- Group Commands
- Execute External Application
- Procedure
- External Procedure File
- Valve Control*
- Global Values Module
 - 6. Global Value
 - 7. Assign Global Value
 - 8. Export

(**Main Sequence, Procedure Library,* and *Valve Control* do not appear in the Toolbox or Add Commands list. Main Sequence is at the top of the command tree for every sequence file. Procedure Library appears in every command tree. See page 4-56 for information about Valve Control.)



Module-specific commands are specific to the Hydra DT device. They control syringe-head movement, X/Y Stage movement, and liquid-handling parameters. The module-specific commands are:

- Aspirate
- Dispense
- Movements to position for pipetting
- Home Axes
- Pause
- Wash Needles
- Mix
- Message Display
- Pump Control
- Speed Control
- Change DARTs Tip Magazine Command

Examples and discussion of the command screens appear later in this chapter, under "Command Reference" starting on page 4-45.



Note: The commands are listed in the order that they appear in the Toolbox and in the Add Commands drop-down list.

Child Dialogs in ControlMate Software

Actions within *ControlMate* are presented as child dialogs—that is, each of these screens or tools appears within the *Main Application* screen. Each dialog has specific tasks. Table 4-4 lists and describes the dialogs.

Table 4-4: Child Dialogs in ControlMate software

Dialog	Access	Description
Options	Tools>Options or 🗃 on Toolbar	Set default parameters for a number of options within the <i>ControlMate</i> application.
Sequence Editor	File>New Sequence; Open Sequence or	Create or edit a sequence file.
Toolbox	on Main Application screen	Click icons to insert commands in sequence files. See "Toolbox" on page 4-8.
Print Preview	File>Print Preview or 🛕 on Toolbar	View text description of a sequence file.
File Validation	Run>Validate File or 📓 on Toolbar	Check a sequence file for errors.
File Execution	Run>Start or D on Toolbar	Start a sequence-file run.
Clipboard Viewer	Edit>View Clipboard or 🔍 on	View commands that have just been cut or copied from a sequence file command tree.

Getting Started: Automated Operation



Dialog	Access	Description
	Toolbar	
Edit and Run Locks	on Sequence Editor screen	Restrict edit and/or run (execution) privileges for a sequence file.
Password Validation	File>Open Sequence>File Locked or	Enter password to access (unlock) a locked sequence file.
About	Help>About	View information about the software version.

This concludes the overview of the software. The rest of the topics in this chapter show you how to use the software.



4. Check the software configuration

Verify That System Connections Are Correct

1. Click the *ControlMate* icon on the Desktop. The *Login* screen appears.



Then the Main Application window appears.



Getting Started: Automated Operation



2. From the Add-Ins menu, select Set Device and Pipettor Options.



The Add-In: Set Device and Pipettor Options window appears.

🖶 AddIn: Set Device and Pipettor Options
Device
Syringe count 96 VARTs capability VQuery Device
XY stage positioner 3 positions
Pump module
eDrop module 🔽
Selected Hydra DT 96 with 3 stage XY positioner
Pipettor
DARTS magazine DARTs 96 1.0ul - 30ul long tips (5506+5507)
Tip count 96 Tip outside diameter 55 1/100 mm
Tip bottom height 🏧 5325 📑 1/100 mm 🚊 💻
Auto wash module The auto wash module can be used with these tips
XY wash vessel A wash vessel placed on the XY stage can be used with these tips
XY wash depth 100 + 1/100 mm
Volumes
Aspirate/Dispense 1 to 30 Increment 0,1 ul
Wash 10 to 30 Increment 10 ul
Aspirate overstroke 3 🗧 🗧 spitback 1 💼 ul
Max air gap volume 15
Volume Correction
Incremental dispense No volume correction : incremental dispense
Neat dispense No volume correction : neat dispense
Reset To Factory Default Settings
OK Cancel Help
Changed



3. Select the *DARTs capability* option. This option configures the software to enable parameters relating to the removable DARTs tips. If you uncheck this option, the *ControlMate* software returns to the fixed-head hardware configuration. This option can also be set automatically by clicking the **Query Device** button.

Syringe count	96	DARTs capability	V	Query Device
XY stage positioner Pump module eDrop module	None None 2 positions 3 positions			
Selected	Hydra DT 96 with p	ump module		

- 4. Select the **X/Y stage positioner** if it is part of your system; otherwise, select **None**.
- 5. Select the **Pump module** and/or **eDrop module** options if they are part of your system. The **Selected** field is a read-only text fild that confirms the options you have selected.
- 6. Select the DARTs magazine that matches the pipettor head on your Hydra unit.

- Pipettor	
DARTS magazine	DARTs 96 1.0ul - 30ul long tips (5506+5507)
Tip count	DARTs 96 1.0ul - 30ul long tips (5506+5507) DARTs 96 5ul - 300ul long tips (5536+5537)
Tip bottom height 🖷	DARTs 96 5ul - 300ul standard tips (5516+5517) DARTs 96 5ul - 300ul wide bore tips (5546/5547/5548)
Auto wash module	The auto wash module can be used with these tips
XY wash vessel	A wash vessel placed on the XY stage can be used with these tips
XY wash depth	100 1/100 mm

The read-only text fields for the Tip count, Tip outside diameter, Auto wash module and XY wash vessel confirm your selection.

- 7. Determine the **tip bottom height** of the disposable tips.
 - a. Insert a DARTs magazine in the pipettor head and a microplate (similar to the one you will be using) on the single or XY stage.
 - b. Move the pipettor tips down into the wells with the down arrow \checkmark until the tips just touch the bottom. Avoid bending the tips.
 - c. Raise the pipettor tips slightly with the up arrow 1 to keep the tips from forming a seal at the bottom of the well.

Getting Started: Automated Operation



 Pipettor 					
DARTS magazine	DARTs 96 1.0ul - 30ul long tips (5506+5507)				
Tip count	96 Tip outside diameter 55 1/100 mm				
Tip bottom height 🏧	5300 → 1/100 mm				
Auto wash module The auto wash module can be used with these tips					
XY wash vessel	A wash vessel placed on the XY stage can be used with these tips				
XY wash depth	100 ÷ 1/100 mm				

8. If you want to reset Aspirate overstroke, spitback volume, or the maximum air gap volume, you can make those changes now. See the description of the fields on the AddIn Set *Device and Pipettor Options* screen on page 4-23 for details about these options.

Aspirate overstroke	3	÷	spitback	1	ul
Max air gap volume	55	- <u>+</u>			

- 9. Click OK.
- 10. From the Tools menu, select **Options**.

File	<u>E</u> dit	⊻iew	<u>B</u> un	Tools	Add-In:	s
	Ê			<u>O</u> ptic	ons	Į.

The Options dialog appears with the Connection tab on the top layer.



🔁 Addin: Set Devic	e and Pipettor Options 📃 🗖 🗙				
Device					
Syringe count	96 V DARTs capability V Query Device				
XY stage positioner	3 positions				
Pump module					
eDrop module					
Selected	Hydra DT 96 with 3 stage XY positioner				
Pipettor					
DARTS magazine	DARTs 96 1.0ul - 30ul long tips (5506+5507)				
Tip count	96 Tip outside diameter 55 1/100 mm				
Tip bottom height ႃ 🏧	5325 🕂 1/100 mm 🚊 👤				
Auto wash module	The auto wash module can be used with these tips				
XY wash vessel	A wash vessel placed on the XY stage can be used with these tips				
XY wash depth	100 1/100 mm				
Volumes					
Aspirate/Dispense	1 to 30 Increment 0.1 ul				
Wash	10 to 30 Increment 10 ul				
Aspirate overstroke	3 • spitback 1 • ul				
Max air gap volume	15 .				
-Volume Correction					
Incremental dispense	No volume correction : incremental dispense				
Neat dispense	No volume correction : neat dispense				
	Reset To Factory Default Settings				
	OK Cancel Help				
🔲 Changed					

- a) Verify that **Connection** settings are correct for your system. Standard settings are shown in the preceding screen. Make corrections to the settings if necessary.
- b) Click **Test** to test the serial connection between the device (Hydra DT) and the computer. When you click test, the computer communicates with the device by issuing a reset command. A warning message will be displayed if the parameters have not been correctly defined or if the device is not connected and switched on.



Note: If you press the **Run** icon **a** after opening a sequence file (a program) but the system does not execute the commands, the serial-port setting might be





incorrect. If that problem occurs, open the **Connection** tab and click the **Test** button to verify that the correct serial port is in use. If an error message appears, select other ports until clicking **Test** no longer evokes an error message. Then try again to run the file.

View Commands and Limits Tabs

The Commands and Limits tabs contain default parameter values.

Click the **Commands** tab and then select a command from the drop-down Command list to see the default settings for a command:

😭 Options	
Connection	Commands Limits Positions Plates Motors
Command	•
┌─ Default \	ta Aspirate Dispense Home Axes Message Display Mix Move to Position Pause Pump Control ▼
Connection	Commands Limits Positions Plates Motors
Command	Dispense 🔽
Default Va	lues
Ŭ i	/olume IC All I Specific II.100 🔡 ul
	Dwell time 0 📩 1/100 seconds
	Fip touch 🗂 Action 🔚 Side left 💌

When you insert a command in a sequence file ("sequence file" is the *ControlMate* name for a program), parameter values for that command are automatically set to its **Commands** tab default values. Setting default values for commands can save you time in creating sequence files. For example, if you frequently use a particular piece of labware in a particular stage position, you could set defaults for that labware and that stage position in the Move to Position command. When you insert the command in your sequence file, you will not have to edit the command screen.

You do not *have* to use the default values that have been set for a command on the **Commands** tab—you can edit the values for the command as you create your sequence file in the Sequence File Editor.



😭 Options _ 🗆 × Connection Commands Limits Positions Plates Motors Value Min Max Unit Aspirate or Dispense volume u 110 Wash volume ul 55 Air gap volume ul Delay after aspirating 1000 1/100th second Delay after dispensing 1000 1/100th second Pause duration 100 Seconds 10 Iterations Mix cycles Wash cycles 16 Iterations 13000 1/100 mm Z axis height 16090 1/100 mm X axis limits 10450 1/100 mm Y axis limits Auto correct invalid values during command edit 🛛 🔽

Now click the Limits tab to see the Limits screen.

The **Limits** tab establishes minimum and maximum values for some parameter values. If you need to set new limits for your applications, you can edit values that appear in black type on this screen.



Note: We recommend that you set the "Auto correct..." option, at the bottom of the screen, to ON. When this option is turned on, the system automatically checks values you set in a command screen and reports an error if you set a value that is outside the range established as default Limits settings.



IMPORTANT: Limits settings **override** Command settings. See the following description of a "Limits override."

For example, if the **Limits** tab setting for "Delay after dispensing" is 1000:

Value	Min	Max	Unit	
Aspirate or Dispense volume	0.1	110	ul	
Wash volume	10	110	ul	
Air gap volume	0.1	55	ul	
Delay after aspirating	0	1000	1/100th second	
Delay after dispensing	0	1000	1 100th second	
Pause duration	1	100 🗟	Seconds	
Mix cycles	1	10	Iterations	
Wash cycles	1	16	Iterations	
Z axis height	0	13000	1/100 mm	
X axis limits	0	16090	1/100 mm	
Y axis limits	0	10450	1/100 mm	





and you set **Dwell time** to 2000 on the **Commands** tab for Dispense:

Connection	Commands Limits Positions Plates Motors	
Command	Dispense	
- Default V	alues	
Ů	Volume C All O Specific 0.100 - ul	
	Dwell time 2000 + 1/100 seconds	
	Tip touch 📄 Action 🗲 Side left 💌	I

then when you insert a Dispense command in a sequence file, you will see the following error message and reset value for Dwell time:

Dispense	
Volume All Specific 0.100 If ul Dwell time 1000 If 1/100 seconds Tip touch Action If Side left	

View the Motors Tab Settings

Connection Comm	ands Limits Positions Plates Motors	
	X Axis mm per step 0.00762	
	Y Axis mm per step 0.00762	
	Z Axis mm per step 0.0127	

The speeds at which the stage will move in the X and Y axes and at which the stage or tray table will move in the Z axis have been set for you.



Caution: We recommend that you do **NOT** reset these speeds without first consulting your equipment provider. See page 1-9 for contact information. Resetting motor speeds could cause the instrument's stage or tray table to move more quickly than you had intended and perhaps damage the stage or syringes.



5. Calibrate the stage (Hydra DT with X/Y Plate Stage systems only)

Before you begin using your Hydra DT with X/Y Plate Stage system, you need to calibrate the stage.

The values that are set during stage calibration are also used when determining plate definitions. Any change in calibration coordinates will cause a change in all plate definitions.



Note: After calibrating the stage at installation, you will need to recalibrate the stage **ONLY** if it is accidentally bumped and shifted out of its calibrated position or if you move the system.



Caution: Always attach the shipping plates on the stage (see page 2-2) before moving the Hydra DT system. Failure to do this may result in damage to the stage.

Before You Start

When you are ready to start the stage calibration, have these items available:

- A standard 384-well microtiter plate to calibrate the stage for a 96 syringe configuration (use 1536-well plate for 384 syringe configuration)
- Flashlight (optional)



Caution: Do not interfere with the stage when the system is running. If the stage movement is restrained, the position information will be lost and the stepper motors may attempt to drive the stage beyond its allowable limit. If this happens, click the **Emergency Shutoff Switch**.





Procedure 4-1: Calibrate the stage

1. Start the software if it is not already started, and select Add-Ins>Set Device and Pipettor Options. Verify that the settings on the screen are accurate for your system.

🔁 Addin: Set Devic	e and Pipettor Options 📃 🗖 🔀				
Device					
Syringe count	96 VARTs capability V Query Device				
XY stage positioner	3 positions				
Pump module					
eDrop module					
Selected	Hydra DT 96 with 3 stage XY positioner				
Pipettor					
DARTS magazine	DARTs 96 1.0ul - 30ul long tips (5506+5507)				
Tip count	96 Tip outside diameter 55 1/100 mm				
Tip bottom height 🏧	5325 🛨 1/100 mm 🚊 👤				
Auto wash module	The auto wash module can be used with these tips				
XY wash vessel	A wash vessel placed on the XY stage can be used with these tips				
XY wash depth	100 ÷ 1/100 mm				
Volumes					
Aspirate/Dispense	1 to 30 Increment 0.1 ul				
Wash	10 to 30 Increment 10 ul				
Aspirate overstroke	3 ÷ spitback 1 ÷ ul				
Max air gap volume	15 ÷				
Volume Correction					
Incremental dispense	No volume correction : incremental dispense				
Neat dispense	No volume correction : neat dispense				
	Reset To Factory Default Settings				
	OK Cancel Help				
📮 Changed					



Note: The information set up here will be used in any sequence file command, so it is important that the information is accurate. The information loaded must correctly reflect the physical characteristics of the pipettor head and tips currently loaded.

You can test the height settings by selecting the up arrow button. Reset the stage by selecting the down arrow button.



Fields

- *Device*—Specifies the Hydra DT base model device type and attached hardware modules (ie. the X/Y plate stage).
- *Pipettor*—Specifies the tips set currently installed. This is important since all actions have to take account of tip length and outside diameter.
- *Tip count*—Number of tips in the tip set.
- *Tip outside diameter*—The tips' outside diameter. This value is used in positioning movements such as tip touch and well offsets.
- *Tip bottom height*—Used to determine the reference point from which all height movements are calculated when positioning stages at the pipettor head. The value is measured from top surface of a stage at the home position (at the lowest point) to the end of the tips (when the magazine is loaded). Getting this value wrong will cause all height movements to be inaccurate.
- *Volume ranges*—Identifies minimum and maximum values used for checking the piston movement for volume limits.
- *Increment*—Used for determining the increment value associated with the volume increment and decrement buttons in a number of sequence commands.
- *Aspirate Overstroke*—Identifies the amount of overstroke that is applied if Overstroke is selected in the Aspirate command. A good starting point for the overstroke amount is 5% of syringe volume; adjust the volume for your system as necessary. If you choose not to use Overstroke at all, you can set the Overstroke volume (and the spitback volume) to 0 (zero).
- *Aspirate spitback*—Identifies the amount of liquid to be returned immediately after the overstroke volume is aspirated and before the specific volume is aspirated within an Aspirate command.
- *Maximum air gap volume*—Specifies the maximum air gap to be applied if Pre air gap is selected in the Aspirate command. See page 6-9 for recommended air-gap volumes.

Getting Started: Automated Operation



Command Buttons



(Test height)-Communicates with the device to test the associated height.



(Reset from test height)—Resets the device axes and is used to reset the device after testing heights. All axes will return to their home positions.



(*Reset to Factory Default Settings*)—Resets all field values for the current piston/tip configuration to those originally set as the factory default values. Any changes that have been made, via this dialogue, will be overwritten.

- 2. Select Tools>Options.
- 3. Click the **Positions** tab and verify that the settings for the Pipettor:Stage 1 position are as shown in the following screen. These settings are standard for the stage when the source (left) nest is under the dispensing head. If those items do not appear in those fields, select them or type them in. Then leave the screen open.



Note: When you are aligning the needle tips on the intersection of a 384 or 1536 well plate, you might find it helpful to shine the light from a flashlight at a right angle to the needles to enhance visibility.

😭 Options	
Connection Commands Limits Positions Plate	es Motors
Position Pinettor: Stage 1	
- Adjust physical location	
Changing any X or Y home values or using the Manual Movement buttons unil course the priortter board to many	Manual movement Steps (mm)
× home 15400 ➡ 1/100mm Y home 10100 ➡ 1/100mm	
Move in	10
0 1/100mm	- - 100
Detail for :	
X offset 450 to 450 1/100 m	nm Reset To Factory
Y offset 0 = to 450 = 1/100 m	nm Default Settings
Z offset 0 to 60000 1/100 m	nm
OK Cance	Help





Note: If an eDrop device is attached, the window will display an additional field (EMB Device) to position the stage for the eDrop needle. See Appendix E for eDrop information.

Each defined position on the Hydra DT system has been mapped to a specific location: for example, the Pipettor:Stage 1 position places the source (left) nest on the stage directly under the syringe head. Positions for the Hydra DT with X/Y Plate Stage systems can be set along 3 axes. These axes are shown in the following diagram.



You can use the Positions dialog to set the minimum and maximum values that the stage can travel on any one of the three axes at any of the defined stage positions. These values are then used within the Sequence File Editor for each respective command.

Fields

- *Position*—The defined stage position. If you select a different position from the dropdown list, the stage resets to home position before moving to the new X,Y location.
- *Adjust physical location: X and Y home*—Use these fields to directly enter the respective axis value, in 1/100 mm increments, to set the required position.
- *Manual movement*—The manual movement buttons are used to directly control the deck motors to move to the required position. The deck will move in the direction specified by one motor step increment, the value for which is set by the "Steps (mm)" slider. For example, if the slider is set to 1mm steps and the "move to the back" button is pressed, then the deck will move 1mm backwards.
- *Test height*—Height at which the tips will be positioned after the deck movement. This is useful for gauging the tip location. If a new position is selected in the Position field, then the tips will ignore this height and move to the safe travel height first.

Getting Started: Automated Operation



- *EMB Device*—This field allows you to select the associated eDrop (Electro Magnetic Bellows) device. This field will only be available if an eDrop related position is selected in the Position field.
- *X offset*—Specifies the amount of movement the stage can move from left to right (and right to left) in this position. Two fields represent the setting, with the first being the minimum value and the second being the maximum value. A negative value causes the stage to move to the right (therefore moving the tips to the left of a well, for example).
- *Y offset*—Similar to the X offset fields except that the values represent movement of the stage in the selected position from back to front (and front to back).
- *Z offset*—Similar to the previous two fields; represents the up and down movement of the stage in the selected position.

Command Buttons



(*Perform movement*)—Clicking on this button will communicate with the device to test the associated position.



(*Reset from test*)—Resets the device axes and is used to reset the device after testing heights. All axes will return to their home positions.



(*Move in motor steps*) —The deck will move in the direction specified by one motor step increment, the value for which is set by the "Steps (mm)" slider. For example, if the slider is set to 1mm steps and the "move to the back" button is pressed, then the deck will move 1mm backwards.











(*Reset to Factory Default Settings*)—Resets all field values for the current position configuration to those originally set as the factory default values. Any changes that have been made, via this dialogue, will be overwritten.



Troubleshooting Calibration of the Hydra DT with X/Y Plate Stage System



Caution: If you receive error messages when calibrating the stage, the computer may not be communicating with the Hydra DT microdispenser. Follow the steps below to troubleshoot the problem.

- 1. Make sure the cables connecting the serial ports of the computer to the Hydra DT microdispenser are securely attached. Only use the cables shipped with the system. If the cables are loose or not connected, repeat running the calibration procedures. If the stage fails to move again, go to Step 2.
- 2. From the **Tools** menu, select **Options**.

The Options dialog appears, with the **Connection** tab on the top layer.

Ţ	ools	Add-Ins	<u>W</u> indow	<u>H</u> elp			
	Ор	tions	X Ad	d Comma	and 🔄 💽		?
	_						
	P	Options					
	ſ	Connectior	Comma	ands Li	mits Positions Pla	ates Motors	
					Under U		
					Serial port	5	Test
					Firmware		
					Send delay	1 +	1/100 seconds
					Polling retry count	1 .	
					- Transaction log		
					Selected 🔽		Clear

- 3. Click the down arrow next to the Serial port field to get a list of ports, and select another com port. Click **OK**.
- 4. Repeat the calibration procedure.



Caution: If the stage still does not move, repeat Step 2 through Step 3 in this section, selecting different Hydra DT com ports. If the stage will not move after you have tested possible com ports for your system, call your PC service technician.





6. Create labware

You must provide a plate definition for each type of labware you will use with your Hydra DT system. Plate definitions are stored in a database and are used when constructing sequence files (in the *ControlMate* software, "programs" are called "sequence files").

A set of plate definitions is included in the *ControlMate* software for you to use as a starting point for your plate definitions. If you are using industry-standard labware in your applications, you will be able to use some of those provided definitions without having to make many, if any, change to them.

Place plates on the tray table or in the source (left) nest on the stage to define them. On the Hydra DT with X/Y Plate Stage system, precise manufacturing of the twoposition plate holder allows information obtained during plate definition in the source nest to be translated into the correct coordinates for the destination (right) nest.



Note: These limitations on types of labware that can be used with the various Hydra DT system setups:

- A 1536-well plate can only be used on a system that includes the X/Y Plate Stage.
- On Hydra DT 96 systems that do not include the X/Y Plate Stage, you can use 384-well plates using the 96/384 manual plate positioner.


Setting Labware Values: Overview

Use the **Plates** tab under **Tools>Options** to create, edit, and delete labware definitions.

r	Options									
	Connection - Plate Ty) () ()	Commands	Limits Po	ositions Pla	ates Motors				1
	Description	۱	384 Round	l well flat (36	000111/111:	21/1112)		•	Add	Delete
	Detail for :384 Round well flat (36000111/11121/11112)									
	Vessel	:	Туре	Plate	-		•	Height	1020	1/100 mm
	Well	:	Count	384	👻 Shape	Round	-	Diameter	300	÷1/100 mm
			Spacing	450	- 1/100	mm				_
			Depths (1.	/100 mm) :	Well 😝	800	-	Aspirate 📭	600	ㅋ
				т	ip touch 🕇 🛚	200		Dispense া 🔸	1350	a
						1200	<u> </u>	C	1,000	-
	Well qua	dra	Int center	s (1/100 п 96 tips это г	1 m) : 1994	Quadrant	Хаж	kis Ya	axis	+x -x
			C When	384 tips are	used	1	225	-22	5	-y -y
						2	-225	-22	5	+y +y
						3	225	225	5	Quadrant X/Y
						4	-225	225	5	Coordinates
	Sub well	:	Count	0 🔻 S	ub well positi	ons (click to se	et each	sub well pos	sition)	
			Sub well	-		Shape		-		
				Sizes (1/100) mm) :	X size			Y size	
				From well ce	enter (17100 r	nm): × 🔽			Υ	÷
				Depths (1/1	00 mm) : S	ub well 📊			Aspirate 😝	
						_,)ispense 🛃	
L										
					ОК	Cancel	н	lelp		
										/

Description of Parameters on the Plates Screen

Fields

Description—The required vessel type for which parameters are to be changed. *Vessel type*—The collective name for the vessel; for example, "plate" or "reservoir."

Getting Started: Automated Operation



Height—The physical height of the vessel, measured from the bottom of the plate to the top of the plate. The height is important as it is used for all movement commands when calculating the height at which the stage can be safely presented to the tips and also for tip touch positioning, and so on.

Well count—The physical number of wells contained within the vessel. The field is used for determining positional parameters for quadrants, and so on.

Shape—Represents the physical top shape of the well and can either be Square or Round.

Width/Diameter—Represents the physical width/diameter of the well measured at the top of the well. The well width is especially important for determining well centers, quadrant offsets and tip touching.

Well spacing—Used when determining the distance (in 1/100mm) between well centers. The field value is measured from the center of one well to the next and is also known as the "well-to-well" spacing.

Well depths—Defines a pre-set height that defines the bottom of the well. Measure from the well top to the well bottom at the well center.

Aspirate depth—Represents a pre-set height, available within the Move command. This height is useful for setting a default height at which liquid is aspirated. The larger the value, the higher the stage movement (and therefore the further down into the well the tips are positioned).

Dispense depth—Similar to the Aspirate Depth, with the exception that it is used for determining a pre-set height for dispensing liquids.

Tip touch Z offset depth—Sets a value that will be used when positioning the vessel for tip touch off. The value is used to determine how near to the top of the well the tips are positioned; for example, a value of 200 1/100mm will position the vessel so that the tips are approxi-mately 2mm inside the top of the well.

Well quadrant centers—The quadrant grid can be used to enter values that represent the well centers for each quadrant. The X and Y axis offset values are measured from the tip center position when no offsets are applied; for example, the dead center for a 96-well plate.



Applying Offset Values in the Plate Definition

To determine the offset values applied, the direction that the stage has to travel is the opposite of the location of the quadrant; that is, to position tips to the left and back of the plate the stage has to move to the right and towards the front of the microdispenser.



Getting Started: Automated Operation



The Quadrant X/Y coordinates frame in the Plate dialog window shows how to apply the offset values.

r	Options									
ſ	Connection	1.	`ommands	Limits	ositions Pla	tes Motors]				
	⊢ Plate Tv	De 1	,oninanas							
	Description	n	384 Round	d well flat (36	5000111/1112	21/11112)		-		
				,					Add	Delete
	Detail fo	r :S	84 Roun	d well flat	(36000111/	11121/1111	2)-			
	Vessel	:	Туре	Plate			-	Heigh	1020	÷1/100 mm
	Well	:	Count	384	➡ Shape	Round	•	Diameter	300	1/100 mm
			Spacing	450	1/100	mm				
			Depths (1	, /100 mm) :	Well 1.1	800	-	Aspirate	1 600	
				,	Cip touch 📩 (200	-	Dispense M		
						1200	•		_]300	<u> </u>
	Well qua	dra	ant center	rs (17100 r	nm) :	Quadrant	X	axis Y	axis	+x -x
			When	n 96 tips are 2011 tips are	used	1	22!	5 -2	25	<u>-y</u> <u>-y</u>
			wher	1 304 ups are	e useu	2	-22	5 -2	25	+x -x +v +v
						3	22!	5 22	5	Quadrant X/Y
						4	-22	:5 22	:5	coordinates
	Sub well	:	Count	0 - 3	Sub well positi	ons (click to se	et ea	ch sub well po	sition)	
			Sub well			Shape 🗌		-		
				Sizes (1/10	0 mm) :	X size		— <u> </u>	Y size	
				From well or	- enter (17100 r	um): X 🗆			Ý	·
				D J G H				<u> </u>		
				Depths [17]	iuu mmj: s	ud well 💶		·	Aspirate 🖡	!∃
									Dispense	비 크니
_										
					ОК	Cancel		Help		

Setting the Number of Quadrants in a Plate Definition

A different grid with different quadrants is available for each plate. The total number of quadrant groups (that is, 1, 4, or 16) depends on the total number of wells contained in the vessel and the current selection of the tip radio buttons. The radio buttons are used to re-draw the grid based on whether the system is a 96 or 384 model.

For example, setting the radio button to 96 tips for a 384-well plate would display a grid containing 4 quadrants.



Determining Parameters to Be Set for Various Labware Types

Table 4-5 lists required parameters for the different types of labware.

	Labware Ty	/pe
Plate	Reservoir	
Х	Х	
Х	Х	
Х	Х	
Х	Х	
Х	Х	
Х	Х	
Х		
Х		
Х*		
	Plate X	Labware Ty Plate Reservoir X X X X X X X X X X X X X X X X X X X X X X X X X X X X X X X X X X X X

Table 4-5: Labware Types and Required Parameters

Getting Started: Automated Operation



Procedure 4-2: Create, edit, and delete labware

Before You Start

• Have a caliper measuring tool available.

Create a New Plate

- 1. Go to **Tools>Options>Plates**.
- 2. In the **Plate Type** area, click **Add**.

😭 Options									
Connection Commands Limits Positions Pl	ates Motors		1						
Description 384 Round well flat (36000111/11121/1112)									
Detail for :384 Round well flat (36000111/11121/1112)									
Vessel : Type Plate	•	Height 1020	1/100 mm						
Well : Count 384 ▼ Shape	e Round 💌	Diameter 300	→1/100 mm						
Spacing 450 🕂 1/100) mm								
Depths (1/100 mm) : 🛛 Well 📭	J 800	Aspirate 🚺 600							
Tip touch 📩	J 200	Dispense 📩 350							
Well quadrant centers (1/100 mm) :	Quadrant X a	xis Yaxis	+x -x						
When 96 tips are used	1 225	-225	-y -y						
C When 384 tips are used	2 -225	5 -225	+x -x						
	3 225	225	Duadrant X A						
	4 -225	5 225	coordinates						
Sub well : Count 🛛 💌 Sub well posit	tions (click to set eac	h sub well position)							
Sub well 🔍	Shape								
Sizes (1/100 mm) :	X size	Y size	• – – – II						
From well center (1/100	mm): X		,						
Depths (1/100 mm) · · · ·	Sub well 1.1								
		Dispanse M							
		Dispense							
	Cancel	Help							
			1.						



	te i	уре								
Description	٦ļ									1
Similar to	· [None					-		Apply	Cancel
Detail fo	r : N	lew plat	e							
Vessel	:	Туре	Plate			-	Height	0		1/100 mm
Well	:	Count	96	👻 Shap	Round	•	Diameter	0		1/100 mm
		Spacing	0	1/10	0 mm					
		Depths (1	/100 mm) :	Well L	0	÷	Aspirate 📭) 0		
			Ti	p touch 🕇	10		Dispense 🖡	10		-
Well qua	dra	nt cente	rs (1/100 m	m) :	Quadra	nt Xa	nxis Y	axis		+x -x
Well qua	dra	nt center Wher Wher	rs (1/100 m n 96 tips are u n 384 tips are	m) : sed used	Quadra 1	int Xa	ixis Y (O	axis		+x -x -y -y +x -x +y +y Quadrant X/Y coordinates
Well qua Sub w ell	idrai	nt center Wher Wher Count	rs (1/100 m n 96 tips are u n 384 tips are	m) : sed used ub well pos	Quadra 1	int X a 0	ixis Y i	axis sition)	1	+x -x -y -y +x -x +y +y Quadrant X/Y coordinates
Well qua Sub w ell	idrai	nt cente Wher Wher Count Sub well	rs (1/100 m 196 tips are u 1384 tips are	m) : sed used ub well pos	Quadra 1	int X a 0 o set eac	ixis Y (0	axis sition)	1	+x -x -y -y +x -x +y +y Quadrant X/Y coordinates
Well qua Sub w ell	idrai	Int center Wher Wher Count Sub well	rs (1/100 m 96 tips are u 384 tips are 0	m) : sed used ub well pos mm) :	Quadra 1 itions (click to Shape X size	int X a 0	ixis Y (0	axis sition)	1 Y size	+x -x -y -y +x -x +y +y Quadrant X/Y coordinates
Well qua Sub w ell	dra :	nt center Wher Wher Count Sub well	s (1/100 m o 96 tips are u o 384 tips are sizes (1/100 From well cer	m) : sed used .b well pos mm) : nter (1/100	Quadra 1 itions (click to Shape X size	int X a 0	ixis Y i	axis sition)	1 Y size	+x -x -y -y +x -x +y +y Quadrant X/r coordinates
Well qua Sub w ell	dra :	nt center Wher Wher Count Sub well	s (1/100 m o S6 tips are u o 384 tips are Sizes (1/100 From well cer Depths (1/10	m) : sed used ub well pos mm) : nter (1/100 10 mm) :	Quadra 1 itions (click to Shape X size) mm) : X Sub well L		ixis Y . 0	axis sition)	1 Y size ∏ Y ⊑	+x -x -y -y +x -x +y +y +y +y coordinates

The New Plate Type screen appears, with an empty Description field.

3. Type a name for the new plate in the **Description** field.

- New Plate	Туре	
Description	1536 round well	
Similar to	None	_

If the plate is similar to one you have already created, select that similar plate from the **Similar to...** list.

Getting Started: Automated Operation



The *New Plate Type* screen shows values for the **Similar to ...** plate that you selected.

Options	A	
Connection New Plate Description	Commands Limits Positions P Type 1536 round well	lates Motors
Similar to	1536 Square well 'Matrix' (4511B-4	517B) Apply Cancel
Vessel	Type Plate	Height 1200 - 1/100 mm
Well	Count 1536 V Shap	e Square Vidth 170 1/100 mm
	Depths (1/100 mm) : Well La	↓ 450 - Aspirate ↓ 350 -
'₩ell quad	a ant centers (1/100 mm) :	Quadrant X axis Y axis +x -x 1 338 -338 -y -y -y 2 113 -338 +x -x +y +y 3 -113 -338 Quadrant X/Y coordinates 4 -338 -338 ✓
Sub well	Count 0 Sub well positive Sub well Sizes (1/100 mm) : From well center (1/100 Depths (1/100 mm) :	itions (click to set each sub well position) Shape Y size Y size Y J mm): X Y Y Y Sub well Y Aspirate Y Y Dispense Y
🗐 Changed	OK	Cancel Help

- 4. Use your calipers to determine values for all required parameters for this type of labware.
- 5. Enter and/or change plate values in the **Detail for: New plate** area of the screen, as follows:
- 6. If the plate is not similar to any defined plates, enter values in all the required parameter fields.
- 7. If you selected a "similar to" plate, make any necessary changes to the parameter values that already appear on the screen. In the following example, we changed the value for "Shape" (of the top of the well) to **Round**. (Note that the parameter name "Width" changed to "Diameter" when we made that change in shape selection.)

Well	:	Count	1536	✓ Shape	Round	-	Diameter	170	1/100 mm
		Spacing	225	1/100					

8. When you are finished entering and changing values for the plate, click **Apply** and then click **OK**.





Edit a Plate



Caution: This function changes an existing plate definition. Before you edit a plate's definition, determine whether the CURRENT plate definition might continue to be needed in existing sequence files. If this plate type is still needed, exit this function and follow the steps to add a new plate type. Select the existing plate type in the field "Similar to..." and then make your changes.

1. Go to Tools>Options>Plates. The *Plate Type* screen appears.

Plate Type			15	_	
escription 1536 Squ	are well 'Matrix' (4	4511B-45176	3) 🔄	·	Add Delete
Detail for :1536 Squ /essel type Plate Vell count 1536 Vell spacing 225	iare well 'Matri Height Shape	x' (4511B - 1200 Square	4517B)	m h [1	70 1/100 mr
1					
Depths (1/100 mm	<u> </u>				
Depths (1/100 mm Well 1 450) Aspirate	₺ 350	📑 Tip tou	ich Z off	iset 📩 200 🛨
Depths (1/100 mm Well 1 450) Aspirate Dispense	▶ 350▶ 250	금 Tip tou [극	ich Z off	iset 📩 200 🛨
Depths (1/100 mm Well L± 450	Aspirate Dispense	 ⊎ 350 ₾ 250 	는 Tip tou 근	ich Z off	iset 📩 200 📑
Depths (1/100 mm Well L± 450	Ascirate Dispense	 ➡ 350 ➡ 250 m) 	Tip tou	ich Z off	iset 亡 200 😐
Depths (1/100 mm Well L±J 450 Well quadrant cen Applicabale to tip magazine that bas	Aspirate Dispense tres (1/100 mm	 ▶ 350 ▶ 250 n) × axis 	Tip tou	ich Z off	iset 📩 200 📑
Vell quadrant cen Applicabale to tip magazine that has	Aspirate Dispense tres (1/100 mm	 ↓ 350 ↓ 250 n) × axis 338 	Tip tou	ich Z off	iset 📩 200 🕂
Well quadrant cen Applicabale to tip magazine that has © 36 tps G 384 tips	Aspirate Dispense tres (1/100 mm Quadrant 1 2	 ↓ 350 ↓ 250 ∧) × axis 338 113 	Tip tou	ich Z off	Note: Quadrant X/Y positions
Well Left Well Left 450 Well quadrant cen Applicabale to tip magazine that has • 96 tps • 384 tips	Aspirate Dispense tres (1/100 mm Quadrant 1 2 3	 ↓ 350 ↓ 250 ∧ axis 338 113 .113 	Tip tou	ich Z off	Note: Quadrant X/Y positions +x -x -y -y -y -y -y
Well Image: Constraint of the second secon	Aspirate Dispense tres (1/100 mm Quadiant 1 2 3 4	 ▲ 350 ▲ 250 ► 338 113 -113 -338 	Tip tou	ich Z off	Note: Quadrant X/Y positions +x -x -y -y +x -x +y -y

2. In the *Plate Type* area, click the arrow for the *Description* dropdown list and select the name of the plate you want to edit.

Description	1536 Square well 'Malrix' (4511B-4517B)	Add	Delete
	384 Square well flat (4310-4324)		
	96 deep well 1.0 ml (4211/4212)		
Detail for :	96 deep well 2.0 ml (4221/4222)		
Vessel type	96 latch rack 1.4ml tubes (4157/4158)		
Well count	96 well 800ul block (8800-96)	170 -	1/100 mn
	96 well flat (4915/4916/4922-29)		

- 3. Make any changes you need to make to the parameter values.
- 4. When you are finished making changes: If you want to save these changes, click **OK** at the bottom of the **Plate Type** window. If you do not want to save them, click **Cancel**.

Getting Started: Automated Operation



Delete a Plate

1. Go to **Tools>Options>Plates**. The *Plate Type* screen appears.

N 22.445						0.2	
escription	1536 Squa	are well 'Matrix' (4	4511B-4517E	3) 🔄	·	Add	Delete
Detail for :	1536 Sau	are well 'Matri	x' (4511B-	4517B)			
/essel type	Plate	✓ Height	1200	± 1/100 m	m		
Vell count	1536	- Shape	Square	🚽 🛛 Widtł	n 1	70 + 1	/100 m
Well spacing	225		ກ	_			
	1						
Denths (1/100 mm)						
Depths (1/100 mm)			Tin tou	ich Z off	set 🐮 🕻 🗔 🖂	
Depths (Well	1/100 mm)	Aspirate	350	Tip tou	ich Z off	set 📩 200	÷
- Depths (Well	1 /100 mm) لغا (450	Aspirate Dispense	➡ 350➡ 250	Tip tou	ich Z off	set 📩 200	÷
-Depths (* Well -Well qua	1/100 mm) 1450 drant cent	Aspirate Dispense	 ➡ 350 ➡ 250 n) 	는 Tip tou -	ich Z off	set 📩 200	÷
Depths (* Well Well qua Applicabale	drant cent	Aspirate Dispense res (1/100 mn Quadrant	➡ 350 ➡ 250 n) X axis	Tip tou	ich Z off	set 📩 200 Note: Quadrant X	_ ⇒ ∧
• Depths (* Well • Well qua Applicabale magazine t	drant cent a to tip hat has	Aspirate Dispense res (1/100 mm Quadrant 1	 ➡ 350 ➡ 250 n) X axis 338 	Tip tou	ich Z off	set t 200 Note: Quadrant× positions	
• Depths (* Well • Well qua Applicabale magazine t • 96 tips • 384 tip:	1/100 mm) 450 drant cent e to tip hat has s	Aspirate Dispense res (1/100 mm Quadrant 1 2	 ➡ 350 ➡ 250 ► 250 ► 338 ■ 113 	Tip tou	ich Z off	Note: Quadrant X positions	// //
Well qua Applicabale magazine t 96 tips C 384 tip:	1/100 mm) 450 drant cent a to tip hat has s	Aspirate Dispense res (1/100 mm Quadrant 1 2 3	 ▶ 350 ▶ 250 ▶ 250 ▶ 338 113 -113 	Tip tou	ich Z off	Note: Quadrant X positions +x +x	→
Well Well Applicabale magazine t © 96 tips © 384 tip:	1/100 mm) 450 drant cent e to tip hat has s	Aspirate Dispense res (1/100 mm Quadrant 1 2 3 4		Tip tou	ich Z off	set 200 Note: Quadrant X positions +x - -y - +x - +y -	// -y -y +y

2. Select the plate type you want to delete from the dropdown list for the Description field and then click the Delete button. The Delete Plate message appears.

Plate Type						
Description	384 Roun	d well flat (360001	111/11121/111	12) 💌	Add	Delete
				-	-	
	Delete p	late type			×	
	⚠	Delete : 384 Ro	ound well flat (36	000111/1112	1/1112)?	
		C	<u>Y</u> es <u>N</u> o			



Caution: Any sequence files that contain this plate definition will not be executable if the plate is deleted.

3. Click Yes if you want to delete the plate. Click No if you do not want to delete it.



7. Create and edit sequence files

After you have calibrated the stage (if it is included in the system) and the labware, you can create sequence files. *Sequence files* are the programs that you will run to execute your procedures.

Follow these rules when you create sequence files:

- A Move command must precede (not necessarily directly) each Aspirate, Dispense, Mix, and Pump Control command.
- If dispensing specific volumes, the total volume dispensed cannot exceed the total volume aspirated.
- Typically aspiration must precede a dispense action (although the Aspirate command does not have to appear in the position in the sequence *immediately* preceding the Dispense command).

The following two procedures, Procedure 4-3 and Procedure 4-4, provide the basic steps for creating and editing sequence files.

Detailed information about using the Sequence File Editor to insert commands into a sequence file and then how to add, cut, copy and paste commands, as well as detailed descriptions of the commands, appears under "Command Reference," starting on page 4-45. You can study that information now or you can begin the procedures and refer to the reference information when you need to.



Note: A sample sequence file appears in Appendix C. It shows examples of entering commands and of grouping commands.

Procedure 4-3: Create a sequence file



Note: *Firmware Version is shown on the display when you first turn the system on, in the format "V*xxx*" where xxx = Numerals of version number, no decimal point separator; e.g., V515 = Version 5.15.

- 1. Start the *ControlMate* software if it is not already started.
- 2. Click the New Sequence File icon 🛄 in the Toolbar.
- 3. Type a name for the sequence in the Title field at the top of the Sequence File Editor screen.
- 4. Enter commands in the sequence by clicking command icons in the Toolbox and then selecting or entering values and options for those commands. Use copy and paste functions, as well as the Group Commands command and the Procedure command, to speed up file creation.
- 5. Click the Validate File icon 🖻 when you are finished adding commands.
- 6. Correct any errors that appear in the Validate Sequence File screen.

Getting Started: Automated Operation



- 7. Save the file:
- 8. Select Save As from the File menu:



10. Type the name of the file in the File name field on the Save Sequence File window and click the Save button.

Save Sequence File		? ×
Save jn: 🔄 ControlMate	- E 🖻	*
AspirateTransfer.cms	DryDisp1uL.cms	Mix1.cms
AutoWash.cms	Exer4-1Prog.cms	폐 MultipleDisp
AutoWashAW.cms	🔊 group.cms	🔊 MultipleDisp
demoProgram.cms	🖻 ManualWash.cms	폐 NeatDisper
Disp2Times.cms	🖻 ManualWashforXYorAW.cms	폐 NeatDisper
DispWetPlate.cms	🖻 ManuaWashXY.cms	🔊 Rearray(4)S
•		Þ
File <u>n</u> ame: NeatDispense		Save 🔪
Save as type: ControlMate sequ	ence files	Cancel

Procedure 4-4: Edit a sequence file

- 1. Click the **Open Sequence File** icon *in the Toolbar*.
- 2. Select the file you want to open from the *Open Sequence File* window and click **Open**.

] hasProc	Disp2Times.cms	Mix1.cms
] preAG	DispWetPlate.cms	MultipleDispense.cms
AspirateTransfer.cms	DryDisp1uL.cms	MultipleDispense1.cm
AutoWash.cms	Exer4-1Prog.cms	NeatDispense .cms
aw wash.cms	ManualWash.cms	NeatDispense.cms
demoProgram.cms	📓 ManualWashXY.cms	📓 Wash2.cms
		<u></u>
a name: NeatDisn	anse cms	

3. Click the **plus** sign ***** next to *Main Sequence* (and the plus sign next to **Procedure Library**, if there is a plus sign there) in the command tree so that you can see all the commands.



- 4. Choose one of the following to begin making your changes:
- 5. Click on the first command/procedure that you want to edit.
- 6. Right-click a command/procedure if you want to add a command below it, delete it, or copy and paste it, and then select the operation you want from the dropdown menu.



- 7. Edit the command as desired or select the values and options for the new command or paste the copied material where you want to.
- 8. Make any other changes you wish to make to the file.
- 9. Click the **Validate File** icon **C** in the Toolbar when you have finished making changes to the file.
- 10. If errors are listed in the Validate File Sequence screen, correct them.
- 11. Click the **Save** icon **III** in the Toolbar to save the changes to the file.

Getting Started: Automated Operation



8. Run a sequence file

Procedure 4-5, starting on this page, provides the basic steps for running a sequence file.



Note: We recommend that you test run each new sequence file without using solutions, so that you can be certain that the stage/tray table heights and other parameters are correct.

Procedure 4-5: Run a sequence file

Before You Start

1. Review these Warnings and Cautions:



WARNING! To shut down the unit IMMEDIATELY in case of an emergency, firmly press the red button on the Emergency Shutoff Switch.



Figure 4-1: Emergency Shutoff Switch



Caution: Do not allow corrosive or oxidizing agents, such as chlorine bleach, strong acid, etc., to be in continuous contact with the microdispenser. Such agents can be used routinely with the Hydra DT microdispense but may damage the case, and the tray table if left in contact with the instrument for extended periods of time. If you need to use these reagents in a protocol, ensure that you clean the microdispenser of any residual corrosive agent as soon as you finish the procedure. If you need to use any strong oxidizing or corrosive agent other than those listed in Table 6-9 on page 6-26, contact your equipment provider. See page 1-9 for contact information.



Note: *Firmware Version is shown on the display when you first turn the system on, in the format "V*xxx*" where xxx = Numerals of version number, no decimal point separator; e.g., V515 = Version 5.15.

In the ControlMate software: If the sequence you want to run is not open, select Open Sequence File from the Toolbar, select the file you want to run, and open the file.



3. Click the **plus** sign next to Main Sequence in the command tree (and the plus sign next to Procedure Library if there is a plus sign) to display the commands and procedures in the file.

曜 Rearray(4)96to(1)384.cms	
Title Rearray (4) 96-well to (1) 384-well	🔲 Edit lock 🔚 Run lock.
Main Sequence	This is the main command sequence. The commands in this section will be executed sequentially from the first command.

- 4. Review the commands/procedures to be sure that this is the file you wanted to run.
- 5. Verify that checkmarks appear next to all of the commands and procedures you want to execute during this run. Select (check) unmarked commands/procedures if necessary.
- 6. Click the **Run** icon on the Toolbar. The software automatically runs a file-validation procedure.
- 7. If there are any errors in the file, correct the errors and save the changes to the file. Then click **Run** again.

The Execute Sequence File screen appears.

Sequence file	Rearray(4)96to(1)384.cms
Last run date	
Last run duration	N/A
Estimated time remaining	9
Progress	0%
	-
100 C 100	

8. Click the **Start** button to start running the file. The stage (or tray table) moves to home position for all applicable axes.

As the sequence file runs, the percentage of the run that is complete appears in the Progress bar on the screen.

9. If the syringes are not down at their lowest (home) position, you will be prompted to place a vessel in the stage 1 position. Put an empty reservoir or plate in the position and then click **OK**. The syringes will empty, and then you will be prompted to remove the reservoir or plate.



Caution: The syringes must be emptied before the system executes a sequence file. Running a file without emptying the syringes might cause damage to the system.

Getting Started: Automated Operation



- 10. If you receive a prompt to place labware in a position, place the labware in the nest and then click **OK** to restart the run.
- 11. When the run is finished, click the close box on the *Execute Sequence File* window and then click the close box on the *Sequence File Editor* screen.

Stopping or Pausing a Sequence File Run

To IMMEDIATELY STOP a sequence file run: Press the red button on the **Emergency Shutoff Switch**.



To pause or stop the sequence file run from within the software: Click the **Stop** button on the *Execute Sequence File* screen. On the *Pause* or *Abort run execution* screen, click **OK** to restart the run or **Cancel** to end the run.



Command Reference

Pages 4-45 through 4-68 provide reference information for the Main Sequence Editor functions and the commands.

The Main Sequence Editor, with its Sequence File Editor window, is described first. You will use the Main Sequence Editor to create and edit all your sequence files. The parts of the *Sequence File Editor* screen and the editor's functions—including add, delete, move, cut, copy and paste commands— are described.

Descriptions of the commands follow the Main Sequence Editor information. The command descriptions are listed in the order that they appear in the Toolbox and in the Add Command dropdown list.

Call Procedure Notepad Group Commands **Execute External Application** Procedure External Procedure File Valve Control Aspirate Dispense Move to Position Movements to position for pipetting Home Axes Pause Wash Needles Mix Message Display Pump Control Speed Control Change DARTs Tip Magazine Command



Note: *Main Sequence Editor, Procedure Library, and Valve Control do NOT appear in the Add Command list or in the Toolbox. See the individual descriptions of these commands for more information.

Getting Started: Automated Operation



Main Sequence Editor

R <new></new>		_ 🗆 ×
Title	📕 Edit lock 📕 Run lock	
Main Sequence Procedure Library	When Sequence This is the main command sequence. The commands in this section will be executed sequentially from the first command.	
	Set Undo Test mode Help	
Hydra II ? Sequence u	unchecked 🗸 Command: OK	11.

The Main Sequence represents the main command sequence. It is called once during file execution and is processed from the top down.

Note: You must select the Main Sequence checkbox in the command tree to execute the entire file.

Elements of the Sequence File Editor Window

Title Bar

🙀 DryDisp1uL.cms	
Title Dry dispense 1uL	🔽 Edit lock 🗖 Run lock

If the open sequence file is one that has already been saved, the title of the file (also known as "file header") appears in the bar at the top of the Sequence Editor window. If the file has not been saved, then that bar contains <New>.

Title—A text entry used to label the file for captions and reports.

Edit and Run Locks—The edit and run lock dialogs are used to control access to sequence file editing and execution. Once a lock has been set, the correct password is required to enable access to all file functions. This means that it is possible to create sequence files and then set an edit lock to prevent a file from being changed. This is useful when creating file templates where the new files are created based on existing ones. An edit locked program can be opened "read-only" without a password and then saved under a new file name using the "save as" function.



Setting the run lock will prompt the user for a password and then for confirmation of the password. If the two password entries do not match then the lock is not set.



Note: Passwords are case-sensitive.

Status Bar

Open files (1)	🔁 Clipboard active	Auto correct: On	Current file 'demoProgram.cms'	
----------------	--------------------	------------------	--------------------------------	--

The status bar is located at the bottom of the *Sequence Editor* screen and provides information relating to the sequence file that is currently open.

Module—The first panel of the status bar displays the instrument name (the name of the device) to which the commands relate.

Changed Status—The second panel shows the current edit status of the file and displays whether or not any changes have been made since the sequence was last saved.

Sequence Validation—The third panel indicates whether the entire sequence has been checked for validation and whether the validation has passed or failed. The sequence validation looks at the command relationships rather than the individual limit values of commands. This type of validation is useful for determining total volume levels, and so on, throughout the sequence. You run a sequence validation by selecting either Validate File or Run from the Run menu.

Command Validation—The fourth panel provides status on the validation of individual commands—for example, if you entered an incorrect height value or a volume value that exceeds the volume limit of the syringes. If any commands contain such errors, this panel displays the text "command errors." You can identify commands within the main sequence that have been detected as invalid by a large red cross that appears over the command icon in the command tree. If no errors are present, the panel reads "Commands OK."

Last Run Information—The duration and date of the last run (if applicable) for the respective file is displayed in this last panel. Determining run time is useful for workload planning.

Command Buttons

Located at the bottom of the edit field screen for each command are the following command buttons:

Set—Refreshes the screen and updates the tree structure with the changes made to the command being edited. The changes are not saved to the file until a save file option has been selected. Values are checked for validation against the rules (which can be set in Tools> Options) before they are written to the tree structure. Any validation

Getting Started: Automated Operation



errors are displayed in a frame within the command edit screen. The icon in the tree structure is also overlaid with a red cross to indicate invalidity.

Undo-Changes the settings back to the original values prior to the current edit.

Test mode—(Available for Move commands.) Allows the values for the command to be tested on the device by restricting the editor to the current command only. Any changes made are then sent directly to the device. This is useful, for example, for the testing of heights and positions. To return from this mode select the button again (it will now show *Edit Mode* as a caption).

Help—Displays help for the respective command.

Command Tree

The command tree represents the program sequence that you create by inserting commands below the "Main Sequence" title. Sequence files are executed from the top to the bottom of this tree structure. Commands can be manipulated in the tree structure in a number of different ways.

Adding, Deleting, Moving, Cutting, Copying and Pasting Commands

Add a command:

- 1. Click (highlight) the command below which you want to add a command.
- 2. Do **ONE** of the following:
 - Right-click the highlighted command. Select **Add** from the dropdown menu and select the command you want to add:





• OR: Click the icon for the command you want in the Toolbox.



• OR: Select the command from the Add Command dropdown menu on the Toolbar.



As each new command is added to the tree on the left side of the *Sequence File Editor* screen, the edit fields for that highlighted command are available on the right side of the screen. You can expand or reduce either side of the screen by clicking and dragging the bar that divides the sides.

Delete a command:

- 1. Click (highlight) the command you want to delete.
- 2. Do ONE of the following:
 - Right-click the highlighted command and select **Delete** from the dropdown list.
 - OR: Press the **Delete** key on the keyboard.
 - OR: Click the **Delete** icon X on the Toolbar.

Move a command:

- 1. Click (highlight) the command you want to move and, while keeping the left mouse button pressed, drag it to the new position.
- 2. Release the mouse button.

You can also use Cut, Copy and Paste buttons to move commands.

Getting Started: Automated Operation



Cut, Copy, and Paste commands:

You can cut, copy and paste commands in the command tree to or from the Clipboard. These features allow you to save time by copying and editing existing commands rather than recreating them manually. For example, if you would like to dispense the same volume into all four quadrants of a 384-well plate using a 96-syringe head, create a group command for the first quadrant. Then copy the group command, paste it three times, and edit only the quadrant information within each new pasted group command.

If the current command is a group command such as Main Sequence, Group Commands or Procedure, then all child commands will follow the same action as their parent.

Cut a command:

- 1. Click (highlight) the command you want to cut.
- 2. Do ONE of the following:
 - Right-click the highlighted command and select Cut from the dropdown list.
 - OR: Click the **Cut** icon 🐰 on the Toolbar.

When you cut a command, it is saved on the Clipboard until you cut or copy another item.

Copy a command (or commands):

- 1. Click (highlight) the command you want to cut.
- 2. Do ONE of the following:
 - Right-click the highlighted command and select Copy from the dropdown list.
 - OR: Click the **Copy** icon **a** on the Toolbar.

Paste a command (or commands):

- 1. Click (highlight) the command after which the new command is to be pasted.
- 2. Do ONE of the following:
 - Right-click the highlighted command and select **Paste** from the dropdown list.
 - OR: Click the **Paste** icon 🔀 on the Toolbar.



Select Commands to be Included or Excluded From Execution

You can exclude or include individual commands or command groups from execution. To prevent a command from executing when the sequence file runs, uncheck the box beside the icon of that command in the tree. NOTE: Checking (or unchecking) a group command will cause the child commands of that command to also be checked (or unchecked). The following example shows an unchecked group command:



Procedure Library



The procedure section contains all of the procedures that exist within the current sequence file. The procedure section is not executed during file execution. Procedures contained within this section can be called either within the current file main sequence structure (internal call) or from another file (external call).

Getting Started: Automated Operation



Call Procedure

 In this fi 	ile mant file		
Exte	nal File ——		
Filena	ime		Browse
Path		 	
	1		Load is

This command allows the inclusion of a selected procedure within the main sequence file, thus allowing creation of standard procedures. For example, a procedure for aspirating an entire plate could be called from another program without having to redefine parameters. Using this command reduces time needed to create sequence files. The Call Procedure command allows procedures to be included either from the library of procedures within the current sequence file or from another sequence file.

Fields

In this file—Populates the *Procedure* field (after "Local procedures" is selected from *Groups*) with the procedures within the current sequence file.

In a different file—Populates the *Procedure* field (after the external sequence file is selected and Load List is clicked) with procedures from that external sequence file.

Filename—Displays the selected file name (if calling a procedure from a different file).

Path—Displays the file path (if calling a procedure from a different file).

Groups—If In this file is selected, includes the list item "Local procedures.

Procedure— If *In this file* is selected and "Local procedures" has been selected from *Groups*, lists procedures within this sequence file. If *In a different file* is selected and Load List has been clicked, lists procedures within the external file. You select the procedure you want to call from this list.



Notepad



Use this space to provide comments about the sequence.

This command is used to create a note entry within the sequence file. The Notepad command is useful for documenting the reason for command values and positioning and for reminders of device configurations for specific sequence files, etc. The content of the notepad has no effect on the sequence file during file execution.

Group Commands

7	Group Commands	
Title		
Loop	1 iterations	

A command group represents a collection of commands. This command allows you to group two or more commands that are used together to perform a task. For example, a group command could represent the copying of samples from a block; another group command could represent the dilution of a block.

Fields

Title—Allows the entry of a text value that represents a name for the group.

Loop—Allows the setting of a value that represents the number of iterations that the group is to be processed. Command execution proceeds to the command following the group command after the loop has been processed the number of times specified by this value.

Getting Started: Automated Operation





Execute External Application

-ilename				Select
Parameters				
Wait until compl	ete 🔽			
utindow				
Window C Visible				
Size (Normal C Mi	nimized (Maximized	
o 1044				
Hidden				
Massage h	n display			

This command allows an external application to be called during sequence file execution. This command might be used, for example, to execute an application that controls an alarm, sends an E-mail message to an administrator, or starts the Microsoft Excel program.

The fields allow you to specify whether the software is to wait for an application to finish (that is, until the application's window is closed) before continuing with the sequence file execution or to continue immediately. You can also set the way in which the applications window is displayed or even choose not to display the window.

Fields

Filename—The full path and file name of the application to execute. Use the Select button to choose the file from a file selection dialog window.

Parameters—Additional parameters that are passed to the application when it is loaded. For example, if the application being called is "C:\Windows\Notepad.exe," then the parameters field might contain the name of a file to open when Notepad starts.

Wait until complete—If checked, specifies that sequence file execution is paused until the application finishes executing or until its dialog window is closed. If unchecked, causes the program to load the specified application and then immediately continue with sequence file execution, leaving the application running.

Visible—Specifies that the application is to display a dialog.

Size: Normal, Minimized or *Maximized*—These fields are available for selection if *Visible* is selected. Only one field of the three can be selected. It specifies the initial window size of the application window.



Hidden—Selecting this field deselects the *Visible* option. If *Hidden* is selected, the application is loaded without showing any window dialog. This option is useful, for example, if the application requires no user interaction; it reduces screen clutter.

Message to display—Selecting the *Hidden* field enables this field. It is optionally used to display a message window to inform a user that the selected application is running or to provide the user with other relevant information.

Procedure

S	Procedure	
Title	Untitled	

A procedure contains a group of commands that are executed whenever the procedure is called from a main sequence structure. The call can be made from the current sequence file (internal call) or from another sequence file (external call). During file execution, any commands contained within a called procedure will be executed as if they existed within the main program structure.

This command allows the creation of standard procedures. For example, commands for aspirating or dispensing an entire plate could be called from other files without having to redefine the parameters, thereby reducing the time needed to create sequence files.

Fields

Title—A text field that can be used to represent the name of the procedure. If the text entered is the same as the text for another procedure in the procedure collection, an index value is appended to the text; for example:



Getting Started: Automated Operation



External Procedure File

🧏 External Pro	ocedure File	
Filename		Select
Path		
File date	File time	
Check file date/time		

A procedure contains a group of commands that are executed whenever the procedure is called from a Main Sequence structure. Using this command will import all procedures contained within an external sequence file into the current file; those imported procedures will be grouped under this command heading. The procedures can then be called from the current sequence file (internal call). During file execution, any commands contained within a called procedure will be executed as if they existed within the main program structure.

This command allows the creation of standard procedures. For example, commands for aspirating or dispensing an entire plate could be called from other files without having to redefine the parameters, thereby reducing the time needed to create sequence files.

Fields

Filename—The name of the file containing the required procedures.

Path—The file path for the file to be included.

File date/File time—The last modified date and time stamp of the required file.

Check file date/time—A message is displayed when the current file date and time is different from the file date and time when the file was last referenced in this sequence file. If this checkbox is checked, the command also displays a validation error message and prevents the sequence file from being executed unless this external procedure file is re-imported.

Valve Control

Allows a VALCO multi position actuator to be directly controlled within a file sequence. The file sequence pauses whilst the actuator selects the required valve position before continuing with execution.

The Valve Control command is not installed during a standard *ControlMate* installation. To have the Valve Control command made available a custom installation must be selected during the initial installation. Alternatively it is possible to install this feature by reinstalling *ControlMate* and selecting to install an additional module.



Fields

Type—The VALCO actuator model type.

Serial Port—The RS232 serial port to which the actuator is attached.

Switch ID—The specific actuator device id. This may not have been set. If a single actuator is used then VICI recommends that the id is not set; in this case choose 'None'. If a device id has been set and a number of actuators are attached to the RS232 serial port then it is possible to set the position for all actuators simultaneously by selecting the 'All' value.

Valve position—Used to determine which valve position to make current.

Direction—The movement from the current position to the desired position requires the actuator rotating the valves. Use this field to determine the direction of rotation 'Clockwise' or 'Counter clockwise'. It is possible to allow the actuator itself to determine the quickest direction automatically by selecting the 'Quickest' value.

Aspirate

🍟 As	pirate
Overstroke	Volume 3 ul
Pre air gap	□ Volume 0,100 🔛 ul
Volume	0.100 🚽 ul
Dwell time	0 1/100 seconds
Tip touch	Action 🗲 Side left

This command aspirates a volume. The command causes the piston motor to move the plungers upward in the syringe barrels, thereby aspirating the given volume. The aspiration is immediate and therefore requires the use of a preceding Move command to ensure that a vessel is positioned below the dispensing head. Checks exist to ensure that the piston does not over-aspirate and therefore stays within the maximum volume limit defined for the current piston head.

Fields

Overstroke—Check this box if the aspirate is the first aspirate prior to multiple dispenses. The selection of this option ensures that the piston motor is primed and improves volumetric accuracy throughout all subsequent dispense aliquots. The Overstroke volume is set on the *Set Device and Pipettor Options* screen under the Add-Ins menu. You can set the Overstroke value and its accompanying spitback volume to 0 (zero) on that screen if you wish to disable the Overstroke option.

Getting Started: Automated Operation



Pre air gap—Aspirates an air gap at the top of the syringe barrel. Aids in single transfers of small volumes. See detailed information about the use of air gaps on page 6-5. (Air-gap maximum volume is set on the Set Device and Pipettor Options screen under the Add-Ins menu.)

Volume—Liquid volume to be aspirated. Use the increment and decrement buttons to change the volume value.

Dwell time—Specifies a period of time over which to leave the needle tips in the sample immediately after the aspirate step.

Tip touch—Causes the tips to be touched against the top of the well after aspiration to remove liquid, which may have adhered to the side or bottom of the tips. Note: Tip touch is executed near to the chosen vessel's well top, the height of which can be adjusted via the Tip Touch Z offset value in Tools>Options>Plates.

Tips for using the Aspirate command:

- Aspirating with Overstroke on the first volume ensures that the piston motor is primed. This is especially useful when aspirating small volumes and works to ensure consistency and accuracy throughout a series of incremental dispenses. Aspirating with Overstroke is required when smaller aliquots are to be dispensed after a large volume aspiration.
- If a small amount of liquid is to be withdrawn and delivered, we recommend that you insert a pre air gap to ensure that all of the liquid is expelled from the needles. See page 6-9 for information about recommended air-gap volumes.
- If you are pooling samples from a 384-well (on a Hydra DT 96 system only) or a 1536-well plate (on a Hydra DT 384 system only), different volumes can be aspirated from the different quadrants. You have to use a different Move to/Aspirate command combination for each quadrant.



Caution: The Aspirate volume must be sufficient to dispense the amount called for in following Dispense commands.



Dispense

Dis 🔰	pense				
Volume	 All Specific 	:	<u>n 100</u>	ul 🔄	
Dwell time	0 ÷	1/100 se	econds		
Tip touch		← Side	left		*

This command dispenses either a measured volume or all of the liquid in the syringes (as specified in the command). The command causes the piston motor to move the plungers down in the syringe barrels to dispense the given volume. The action is immediate and therefore requires the use of a preceding Move command to ensure that a vessel is positioned below the dispensing head.

Fields

All—If selected, dispenses all liquid in the syringes. This does not perform a piston reset at the end of the action.

Specific—If selected, dispenses the volume specified in the adjacent field. Use the increment and decrement buttons to change the volume value. Note: Choose this option when dispensing small aliquots after an aspiration with Overstroke.

Dwell time—Specifies a period of time over which to leave the needle tips in the sample immediately after the dispense step.

Tip touch—Causes the tips to touch one or more designated points on the sides of the wells, to dispense liquid at those points. Note: Tip touch is executed near to the chosen vessel's well top, the height of which can be adjusted via the Tip Touch Z offset value in Tools>Options>Plates.

Tips for using the Dispense command:

- For a 384-well (on a Hydra DT 96 system only) or a 1536-well plate (on a Hydra DT 384 system only), different volumes can be dispensed into the different quadrants. You have to use a different Move to/Dispense command combination for each quadrant.
- For best performance in low-volume dispensing, dispense some liquid in the syringes back into the source plate before dispensing into the destination plate. To do this, use the Overstroke option in the Aspirate command that precedes the dispense.

Getting Started: Automated Operation



Move to Position

Position	Pipettor: Stage 1		
Vessel type	96 well flat (4915/4916/4922-29)		
Check vessel h	nolder sensor 🔽		
Height	Preset Well top Specific 3340 1/100 mm		
Quadranl	1		
Tie Augli affect	Redefiend		

This command is used to position either of the stage nests at a pre-set position. The nests are identified by the position number, with source nest (stage position 1) being on the left (as viewed from the front of the device) and destination nest (stage position 2) on the right. The movements are grouped into two position types:

- Position a nest under the dispensing head
- Position a nest at a place forward of the dispensing head—for example, when filling plates with solutions

The Move command is a very important command, because all actions require the positioning of a nest or the tray table (the latter is positioned along a Z axis only; its X and Y axes are static). It is therefore possible to test the actions of a Move command when editing a sequence file by clicking the **Test** mode button on the Sequence File Editor dialog window.

Movements to position for pipetting

Fields

Position—Sets the stage position. Other field parameter availability is dependent on the position selected.

Vessel type—Specifies the vessel type ("vessel" = "labware") that is situated at the target location. Different vessel types have different settings for the other fields within this command. The values for this dropdown list are created in the Tools>Options>Plates dialog.



Note: The labware choices that populate the dropdown Vessel type list are those pieces of labware that your system configuration (referring to device type and syringe count) allows. For example, if your system has 96 syringes and does NOT include either the X/Y Plate Stage, then the labware list will include only labware pieces that contain either 96 wells or one large well (reservoir) that accommodates all 96 tips.



Height—Specifies the height to which the stage or tray table should be raised. Heights can be set either by choosing one of the preset heights (which are determined by the vessel type parameters) or by specifying a specific height value. Movement is measured from the position at the base of the stage or tray table from its zero position upwards.

Quadrant—[on Hydra DT with X/Y Plate Stage only] If the combination of vessel well count and syringe count allows for movement to different quadrants within a vessel, then this field is available. Quadrant offset values are entered in the Tools>Options>Plates dialog for the respective vessel.

Tip/well offset—[on Hydra DT with X/Y Plate Stage only] It may be necessary to position the tips away from the center of each well. For example, when using 384-well plates with low volumes, the greatest accuracy might be provided by positioning the needle tips to touch off in one of the well corners. Checking this field will allow the setting of tip positioning either by choosing one of the pre-set positions or by entering X- and Y-axis offset values. See the Tools>Options>Positions dialog for more information on entering offset values.

Home Axes



This command can be used to reset the X, Y, and Z axes to their home position during file execution.

Fields

Piston—Resets the piston (syringe plunger) motor.



Caution: This reset will cause any liquid in the tips at the time to be dispensed immediately. It is therefore advisable to position a vessel (such as a reservoir) under the tips before executing this command.

Stage Z—Resets the Z axis.

Stage Z, X and Y—Resets the X, Y and Z axes.

Getting Started: Automated Operation



Pause

٢	Pause	
۹	Duration	seconds
c	Wait for user	

Inserting a pause command into a program causes the device to stop processing according to the pause parameters. After the continue condition has been reached, the program continues at the next step within the sequence.

There are two types of pauses. These are:

- Time based
- Infinite—requiring a user to specify when to resume

Fields

Duration—Causes the device to pause for a set timed interval.

Seconds (duration)—If checked, specifies the time interval over which to pause if *Duration* has been selected.

Sound alarm when pause complete—Sounds an alarm on the internal speaker on the PC 5 times after the pause period has expired.

Wait for user—Pauses the device indefinitely and requires the user to select, via a window dialog, when to continue.



Wash Needles

🖁 Wash Needles				
Wash Location				
C Auto wash module				
 Use XY stage positioner based wash vessel 				
Position Pipettor: Stage 1				
Vessel type XY stage top wash vessel				
Cycles 2 ··· Volume 30 ··· ul				
First Pump 1 Pump 2				
Fill cycles 1				
Fill time 10 ÷ seconds				
Second 🔲 💿 Pump 1 🔿 Pump 2				
Fill cycles 1				
Fill time 1 seconds				
Drain time 14 seconds				

The Wash Needles command facilitates the washing of both the inside and outside of each tip loaded in the tip magazine. The command can use a wash vessel on the stage or, via a wash vessel placed on the XY stage positioner.

The command requires a preceding Move command to position the tips at the wash vessel position and also for ensuring that a wash vessel type has been selected.

Fields

The fields in the command are grouped into 3 sections; Wash Location: which allow the user to specify which wash vessel and wash location should be used; Wash which allows the number of wash cycles to follow; Pump usage sequence which allows the pump usage order to be specified.

Fields relating to Wash Location

Auto wash module

Selecting this field will use the inbuilt wash sequence within the firmware. The wash vessel location used is that which is underneath the XY stage positioner. This field is not available if the device does not have an auto wash module installed. During file execution, the Hydra DT device controls the XY stage and piston movements, using the parameters entered in the fields within this command.

Use XY stage positioner based wash vessel

Selecting this option will us a wash vessel to be placed on top of the XY stage positioner. This field is not available if the currently selected needles or DARTs tips cannot be washed using an XY stage positioner based vessel. During file execution,

Getting Started: Automated Operation



ControlMate will directly control all XY stage and piston movements, using the parameters entered in the fields within this command.

Fields Relating to Wash Cycle

Cycles

This field is used for defining the number of wash cycles required.

Volume

The liquid volume to be aspirated and dispensed during each wash cycle. The increment and decrement buttons will change the volume value accordingly. The increment for these actions can be set via the <u>Set Device or Pipettor options Add-In</u>

Fields Relating to Pump Usage Sequence-

Please note: These functions are not available for the Hydra DT

Pump usage sequence

The *Pump usage sequence* fields are used to specify the pump usage during the each wash cycle

First/Second

These fields specify, within each cycle, whether to use one or both pumps. The *First* pump set of fields is mandatory since the cycle has to at least run one set of pumps. The *Second* is therefore optional and is used if, for example, two different liquids are required for washing during each wash cycle.

Pump1/Pump2

Specified which pump within the wash module to use.

Fill cycles

Specifies the number of times the pump is to be used within the current cycle. This is used in conjunction with the drain pump usage.

Fill time

Specifies how long, in seconds, the fill pump is to run before the drain pump is switched on.

Drain time

Specifies how long, in seconds, the drain pump is to run.

- 1. Fill pump 1 activates and fills the Wash Reservoir with wash solution.
- 2. The syringes aspirate and dispense 10µL from—and back into—the Wash Reservoir 3 times.
- 3. The drain pump empties the reservoir.


- 4. Steps 1 through 3 are repeated 2 more times (because Fill cycles for Pump 1 is set to 3).
 - Fill time—Specifies how long, in seconds, the fill pump is to run.
 - Drain time—Specifies how long, in seconds, the drain pump is to run.



Note: This command cannot be used with Hydra DT base-unit only systems, because they do not include the Wash Pump Module. For those systems, use a Mix command to execute a Manual Wash operation. See more about Manual Wash in the "Tips" section of this command description.

Tips for using the Wash Needles command:

The syringes can be washed manually and automatically.



Note: Use the Mix command to do a Manual Wash. A **Manual Wash** takes place using a reservoir (single-well piece of labware) in the source nest or on the tray table. During the wash procedure, the reservoir is placed under the dispensing head and the tips are cleaned with the number of up/ down strokes (mix cycles) specified. Fill the reservoir with the wash solution before you begin the wash procedure.

See also: Mix command, page 4-65; Pump Control command, page 4-67.

8.4	:
IVI	IX

ycles	1	-	
'olume	0.100	u	
Aspirate h	eight	📩 Well top	-
)ispense	height	📩 Well top	•

This command can be used to aspirate and then dispense liquid in a vessel to resuspend material in the vessel so that a homogenous solution can be created prior to a subsequent aspiration.

The Mix command can be used to aspirate/dispense liquid in a vessel to re-suspend material in the vessel so that a homogenous solution can be created prior to aspiration.

The Mix command can also be used to wash the D.A.R.T.s on Hydra DT systems that do not include the Wash Pump Module or in instances where the standard wash routine is not required. See page 4-65 for details about using the Mix command for these purposes.

Getting Started: Automated Operation



This command requires a preceding Move command to ensure that the vessel is positioned under the dispensing head.

Fields

Cycles—Specifies the number of mix cycles. (One *cycle* is one aspirate plus an immediately following dispense.)

Volume— Specifies liquid volume to be aspirated and dispensed during each mix cycle. Use the increment and decrement buttons to change the volume value.

Aspirate height—Specifies the tray table/stage height at which aspiration occurs. Choose one of four pre-set heights (heights are set during labware calibration).

Dispense height—Specifies the tray table/stage height at which dispense occurs. Choose one of four pre-set heights (heights are set during labware calibration).

Message Display



This command allows a custom message to be displayed on the Hydra DT display panel at the top front of the microdispenser.

Fields

Normal display—Returns display to its original display, the one that appeared on the LCD display before the Message Display command was executed.

Custom message—Allows user to type message into screen area that will then be displayed on the display panel. The character limit of the message is the number of characters that fit on the four text lines.



Pump Control



Note: This command is not available for use with the Hydra DT

This command allows you to control the pumps directly, independent of a wash routine. You might want to use this command, for example, if you wanted to fill the Wash Reservoir with a buffer solution but did not want any wash cycles to be performed.

The pumps can be used to fill or empty wash or other appropriate vessels for a user specified duration (in seconds).

A Move command must precede a Pump Control command to ensure that a reservoir vessel is positioned at the dispensing location. The Move command must have the Wash position specified. The software prevents the file from executing if this Move command is omitted.

Fields

Pump—Pump1/Pump2/Drain—Selects the associated pump to use. ("Drain" is Pump 3, or the "Empty" pump.)

Duration—A specified duration (in seconds) after which time the pump will automatically deactivate. Default is set to 10 seconds.

Speed Control

Annianta	E.	Claur	2			2		с. На
Aspirate	1.	SIUW	1	63	Е	-		га
Dispense	V	Slow	-					Fai
Empty	v	Slow		20	30			Fa
			1	60	10	- {	1	

The piston (syringe-plunger) speeds for Aspirate, Dispense, Empty, and Wash operations can be adjusted using the Speed Control command.

To set a speed, click the check box for the respective action and then use the slider to set the required speed.

Getting Started: Automated Operation



A speed setting used in a sequence file is used for that action type until another speed control command is encountered in the sequence during file execution.

Tips for using the Speed Control command:

Reducing piston speed is recommended for improving accuracy and precision for high-viscosity liquids.

Change DARTs Tip Magazine Command

¶→¶ Change DARTs Tip Magazine			
DARTs type	DARTs 96 1.0ul - 30ul long tips (5506+5507)	-	
Track usage	✓ Change after 10 📩 uses		

This command is used to pause the sequence file execution in order for the DARTs tip magazine to be replaced. The command has the ability to check and issue the instruction to pause and replace tips each time it is called within a program sequence or by an incremental counter to check the usage. To use the incremental counter, the command must exist in a command Group that uses iterations.

During File Execution, a window dialogue prompt is displayed on the screen prompting the user to replace the DARTs tip magazine. The user is then required to click the OK button to continue with the rest of the protocol sequence.

Fields

DARTs type

This field is used to specify the DARTs tip type to use. By default, the one that is specified in the Add-In : Set Device and Pipettor Options is highlighted. During File Execution, once the user has changed the DARTs tip magazine and clicked the OK button to continue, the Add-In : Set Device and Pipettor Options field value is changed to the one specified in this command.

Track usage

This sets an internal counter that determines when tips should be changed within a loop. The counter is set to 1 at the start of every file execution.

Change after (n) uses

This determines the internal tip change counter. This field is only displayed if the Track usage check box has been checked.



ControlMate OLE Optional Add-On Software Component

ControlMate OLE is an optional, add-on software component. It provides an application developer with an ActiveX^{®2} COM automation interface for the *ControlMate* device-control application. The component enables an application to instantiate an object that can load, modify, and execute *ControlMate* sequence files.

The calls made to the *ControlMate* components do not require loading the *ControlMate* application into memory.

A powerful feature of *ControlMate* OLE is the ability it provides to dynamically change and set command field values within a sequence file. This ability allows the integration of information such as source values (for example, aspiration volumes) that have been extrapolated from databases.

The *ControlMate* OLE component has been developed for use with Microsoft[®] Visual Basic[®] versions 5 and 6.

For information about ordering the *ControlMate* OLE component (cat. no. 501-30023), go to the Matrix Technologies web site at <u>http://www.matrixtechcorp.com/</u> and access the price list.

Getting Started: Automated Operation

² ActiveX is a registered trademark of Microsoft Corporation.



5: Service and Maintenance of the Hydra DT Microdispenser

Overview of Service and Maintenance Tasks	5-2
Contact Us If You Have Service or Maintenance Questions	5-3
How to Return the Microdispenser or Microdispenser Parts for Service or Repair	5-3
Checking the PTFE Tips	5-4
Replacing PTFE Tips	5-5
Replacing Syringes	5-7
Removing the syringe head from the Hydra tower.	.5-7
Syringe Head Disassembly and Syringe Removal	. 5-9
Installing a New Syringe	5-12
Attaching the syringe head to the Hydra Tower	5-13
Sending in your DT head	5-14
Checking and Replacing Fuses	5-15

Service and Maintenance of the Hydra DT Microdispenser



Overview of Service and Maintenance Tasks

To keep your Hydra DT microdispenser in its best working order, you need to provide regular service and maintenance. This chapter describes basic service and maintenance tasks for the Hydra DT microdispenser. These tasks are listed in Table 5-1 on page 5-2. You do not need to call your equipment provider for help with the problem before performing the service and maintenance tasks listed in Table 5-1.



Note: If you need to perform a service or maintenance task that does NOT appear in Table 5-1 on page 5-2, please contact your equipment provider before performing the task. See page 1-9 for contact information.

Table 5-1:	Service and	Maintenance	Tasks for	the Hydra DT
Microdispe	enser			

Task Frequency	Task	Indicators that service is needed	See
Weekly	Check PTFE tips	 Unit dispenses uneven volumes. Plungers above tips or areas between tip divisions are discolored. Liquid is visible above the tips. Liquid is dripping from the needles. 	page 5-4
	Replace PTFE Tips	 Unit dispenses uneven volumes. Plungers above tips or areas between tip divisions are discolored. Liquid is visible above the tips. Liquid is dripping from the tips. Note: Heavy use = many operations daily; Moderate = few operations daily; Light = few operations weekly 	page 5-5
Use: Heavy: 2-3 mos. Moderate: 6-12 mos. Light: 12+ mos.	Replace syringes	 Needles are bent or broken. Syringe performance is unacceptable. Syringe barrel is cracked or broken. 	page 5-7
	Check & replace fuses	 When you press the power switch to ON: System does not turn on. (Check power-module fuses.) System turns on but LED display does not come on, green light on controller board does not blink, and motors do not move. (Check logic power supply fuse.) System turns on, LED display comes on, green light on controller board blinks, but motors do not move. (Check motor power supply fuse.) 	page 5-15



Contact Us If You Have Service or Maintenance Questions

If you have any questions about the service or maintenance of your Hydra DT microdispenser, please contact your equipment provider. See page 1-9 for contact information.

How to Return the Microdispenser or Microdispenser Parts for Service or Repair

Please see page vi for a detailed description of how to return the microdispenser or microdispenser parts for service or repair.

Acetic acid	Dimethyl Formamide	Triethylamine
Acetone	Dimethyl Sulfoxide	Toluene
Acetonitrile	Ethanol	Trifluoroacetic Acid
Ammonia 27-30%	Hexane	Trimethyl Orthoformate
Chloroform	Methanol	Xylenes
Dichloroethane	Methylene Chloride	Toluene
Diisopropylamine	Methylene Chloride/TFA, 1:1	
Dimethylacetamide	Pyridine	

Table 5-2: Chemical Reagents Used Routinely With Hydra DT Systems

Guidelines for Manual Wash washes

Follow these guidelines for all Manual washes:

- Use a wash volume that is equal to or greater than the sample volume.
- Change the wash solution for fresh solution after every 3 wash cycles.
 - a. [Manual Wash Only with no pump module] Wash the syringes with the same base solvent as was used in your protocol, rather than dH₂O, as the first wash solution. Wash 3 wash cycles.
 - b. [Manual Wash Only with no pump module] Wash with methanol or ethanol, 1 wash cycle.
 Wash with dH₂O, 3 wash cycles.
 - c. If washing between uses of DMSO: Wash the syringes with DMSO, rather than dH₂O, as the first wash solution, 3 wash cycles.
 - d. Wash with methanol:acetone (1:1 v/v), 1 wash cycle.

Service and Maintenance of the Hydra DT Microdispenser



- e. Wash with methanol or ethanol, 1 wash cycle.
- f. Wash with dH₂O, 3 wash cycles.
- g. Wash with dH₂O additional times if you prefer.
- h. Wash with dH_2O , 3 wash cycles.

Checking the PTFE Tips

The integrity of the PTFE tips on the syringe plungers is essential for precision dispensing. You can prolong the life of the PTFE tips by following the proper syringe-cleaning procedures and schedules.

Examine the PTFE tips weekly. If you see the following, change the tips immediately:

- Liquid above the PTFE tip
- Liquid dripping from the needles
- Discoloration on the stainless steel plunger above the tip or between the divisions of the tip
- Unit is dispensing uneven volumes



Caution: If you see spotting of a reddish-brown precipitate inside syringe barrels, do **NOT** replace the PTFE tips on your syringe plungers. Instead, send the syringes to us for factory service.

Contact the Technical Service Department for more information regarding preventative maintenance services provided

Generally, you will want to replace all the PTFE tips at the same time.

Typically the PTFE tips last six to twelve months. However, the life of the PTFE tips varies with usage. Table 5-3 provides guidelines for tip replacement. Your experience might vary from these parameters.

Table 5-3: PTFE Tip Replacement Guidelines

Usage Level Usage Definition		Replacement Frequency
Light	A few operations weekly	12+ months
Moderate	A few operations daily	6 – 12 months
Heavy	Many operations daily	2 – 3 months

See Replacing PTFE Tips, following, for instructions.



Replacing PTFE Tips

Replace the PTFE tips as needed. See Table 5-3 for guidelines on when to replace the tips.

Replace all the tips at once if you are replacing the tips for any of the following reasons:

- Unit is dispensing uneven volumes.
- Liquid is visible above the tips.
- Liquid is dripping from the needles.



Caution: If you see spotting of a reddish-brown precipitate inside syringe barrels, do **NOT** replace the PTFE tips on your syringe plungers. Instead, send the syringes to us for factory service.

If you replace a syringe, replace the PTFE tip as well (never use an old tip with a new syringe).

Procedure 5-4: Replace PTFE tips on plungers

Before you start

Set up a Hydra DT EPROM file (one of the 16 files resident on the microdispenser) to perform the operations described in this procedure.

Have these items ready to use:

- PTFE tip replacements
- Plunger Rod Tool
- Phillips-head screwdriver (Note: You will need a short screwdriver to remove the head cover plate screws that are adjacent to the front of the microdispenser.)

Replacing the PTFE tips

- 1. Empty the tips by pressing **Empty**.
- 2. Set the Air parameter in an Aspirate command to maximum volume. Press Aspr to fill the syringes with air.
- 3. Run the Empty operation to lower the head, but press the STOP button when the head has lowered halfway to pause the operation. Having the head in this position allows access to the plunger head buttons.
- 4. Turn off the power to the microdispenser.

Service and Maintenance of the Hydra DT Microdispenser



5. Use a Phillips-head screwdriver to remove the screws from the head cover plate and then remove the head cover plate. See Figure 5-1.



Figure 5-1: PTFE Tip Replacement (Showing Tip for 100µL Syringe)

- 6. Pull the plunger out of the head plate and syringe barrel.
- 7. Using the needle-nose pliers, carefully pull the PTFE tip from the plunger. Be careful not to damage the plunger when removing the PTFE tip.
- 8. Place the new PTFE tip with the hole facing upward into the hole in the head assembly. Press the plunger straight down into the PTFE tip. Make sure the PTFE tip has been fully seated onto the plunger and that the PTFE tip has not been damaged.
- 9. Place the plunger back into the head and syringe barrel. Slide the plunger up and down the length of the syringe barrel. The plunger should travel smoothly through the glass barrel. If the travel is not smooth, the PTFE tip might have been damaged during insertion. Inspect the tip and replace it if necessary.
- 10. Replace the head cover plate and tighten the screws.



Replacing Syringes

Syringe replacement will require that the Hydra DT pipettting head be removed first. A case will be provided to send a Hydra DT syringe head to the Technical Service Department for in-house services to be performed.

Please contact Matrix Technical Service Department for information regarding preventative maintenance services.

Removing the syringe head from the Hydra tower.

1. Turn the power on and allow the syringe head to home. Install the 2 shipping blocks on each side of the syringe head. Each shipping block is bolted through the top of the syringe head using a 5/64-inch Allen wrench on the 2 button head screws. Leave these blocks in place at all times while the syringe head is not installed on the Hydra tower. See Figure 5-2 for shipping blocks and placement onto Hydra DT syringe head.



Figure 5-2: (From Left to Right): Shipping blocks are supplied with the syringe head case provided by the Technical Service department; shipping block placement, shipping block bolt positions

2. Loosen and remove the 2 Phillips head screws to allow the baffle cover to be taken off. See Figure 5-3.



Figure 5-3: Top view of the Hydra DT tower Philip head screws to be removed

Service and Maintenance of the Hydra DT Microdispenser



3. Remove the 4 socket-head screws on the lower syringe head assembly with a 9/64-inch Allen wrench. See Figure 5-4 for socket screw positions.



Figure 5-4: Left and right views of the 4-socket head screws to be removed on both sides of the Hydra DT syringe head

4. Remove the 7 socket-head screws on the syringe head top with a 5/32-inch Allen wrench. See Figure 5-5.



Figure 5-5: Top view of the 7 socket head screws to be removed from syringe head



5. Lift up on the entire syringe head assembly until it releases from the two mounting pins on the Hydra tower. **Do not lift or carry the syringe head by the clamping handle.** See Figure 5-6 for proper lifting technique.



Figure 5-6: Lifting the syringe head from the Hydra DT tower

6. If you are sending the Hydra DT head in for repair or Preventative Maintenance and calibration, see the section Sending in your DT head on page 5-14. If you are replacing a syringe or syringes, go on to the next section.

Syringe Head Disassembly and Syringe Removal

1. Lay the syringe head on its back. The lower portion of the head will need to be propped up so that the head lies flat (two normal well plates stacked are the perfect height). See Figure 5-7.



Figure 5-7: Hydra DT syringe head positioned on its back stacked on 2 standard height plates

Service and Maintenance of the Hydra DT Microdispenser



2. Remove the two lower flathead screws on the handle assembly (there is one on each side of the syringe head). The tip magazine C clamp will now slide out easily. See Figure 5-8 for flathead screws to be removed and view of C-clamp.



Figure 5-8: Side view of Hydra syringe head flat head screws and bottom view of syringe head C Clamp to be removed.

3. Remove the six bolts that attach the silicone pad bracket to the syringe head. The pad bracket can now be removed. See Figure 5-9.



Figure 5-9: Silicon pad bracket



4. Using a 3/32 inch Allen wrench, remove the twelve socket-head cover bolts. See Figure 5-10.



Note: You may need a small flathead screwdriver to remove the cover from the syringe head.



Figure 5-10: Syringe head cover and bolts to be removed

5. Push the plunger rod tool through the bottom of the glass barrel until it meets with the Teflon tip. Slightly push on the Teflon tip until you can see the corresponding plunger move above the top of the head. Grasp the plunger top with your free hand and pull it out slightly more. Push down on the plunger until you can take hold of the glass barrel. Separate the glass barrel from the plunger. Continue this until you have removed all syringes necessary. See Figure 5-11 for removing syringes.



Figure 5-11 (left to right): Inserting plunger rod tool into bottom of glass barrel; removing syringes.

Service and Maintenance of the Hydra DT Microdispenser



Installing a New Syringe

- 1. Take a new syringe and remove the plunger from the barrel.
- 2. With one hand, slide the glass barrel through the barrel guide.
- 3. With the opposite hand, place the new plunger through the top of the piston retainer head, and push it into the barrel. Support the base of the glass barrel and raise the new syringe so that its base is even with the bases of the other syringes. Push the plunger until it is flush with the piston retainer head.
- 4. Once all of the syringes are replaced, you can re-install the silicone pad holder. Be sure to place the white silicone pad on the bottom, the compression plate in the middle, and the blue silicone pad on top. The beveled edge of the pad holder should be in the front position of the syringe head. See Figure 5-12 for silicone pad installation order.



Note: Make sure that the syringe barrels line up with the holes in the silicone pad. Tighten the 6 flat-head screws. Do not over tighten the screws.



Figure 5-12 (From left to right): Blue silicone pad, compression plate, white silicone pad and silicone pad holder.

5. Reposition the head plate cover and install the 12 socket-head screws carefully. Tighten them in pairs, and refrain from creating too much pressure on one screw.



Note: Do not over tighten the screws.

6. The tip magazine holder C clamp can now be re-installed. Slide the two pistons through the piston guides. Re-attach and tighten the two flat head screws to the handle assembly. Do not over tighten the screws. Your DT head is ready to be attached to the Hydra tower.



Attaching the syringe head to the Hydra Tower

1. Align the top of the head with the two guide pins on the syringe head back plate. The syringe head should slide in place with minimal force. See Figure 5-13 for replacing the syringe head on Hydra DT tower.



Figure 5-13: Guide pins to align syringe head with for reattachment on Hydra DT tower.

- 2. Reinsert the 7 socket-head screws on the syringe head top and tighten them with a 5/32-inch Allen wrench. Do not over tighten the screws. See Figure 5-5.
- 3. Reinsert the 4 socket head screws that hold the lower portion of the syringe head and tighten them with a 9/64-inch Allen wrench. Do not over tighten the screws. See Figure 5-3.
- 4. Remove the 2 shipping blocks before running the Hydra.

Service and Maintenance of the Hydra DT Microdispenser



Sending in your DT head

Call the Technical Service Department to obtain a return number and to have a Hydra DT syringe head case sent to you. If you require a loaner DT head, one can be provided with a weekly charge. Contact the Technical Service department for more details.



Note: It is required that you use this case when sending in your head for service.

1. In order to place the syringe head properly into the case, the handle will have to be removed. Loosen and remove the socket-head screw with a 5/32-inch Allen wrench on both sides of the head. Then loosen and remove the upper flathead screws from the clamping mechanism as shown. The handle should come free easily. See Figure 5-14.



Figure 5-14: Removing the Hydra DT tip clamping handle

2. The head should be placed into the shipping case with the front glass shield facing down. See Figure 5-15 for syringe head placement in shipping case.



Figure 5-15: Shipping the Hydra DT syringe head in provided case



Checking and Replacing Fuses

The Hydra DT microdispenser has 2 fuses that you might need to replace at some time.

The power module, located inside the microdispenser behind the power switch and plug on the back of the microdispenser, contains two 3-amp fuses. If the system does not turn on when you press the power switch on, you might need to replace one power-module fuse.

The third user-replaceable fuse, a 4-amp fuse, is on the logic power supply. If the LED display does not come on when you press the microdispenser's power switch on, neither the syringe head nor tray table/stage moves when you press dispenser buttons, and the green light on the controller board does not blink, then you might need to replace this fuse.

The following pages contain procedures to check and replace these fuses.

Procedure 5-8: Check and replace the fuses in the power module

If the microdispenser does not turn on when you press the power switch on, follow this procedure to see, first, whether the power-module fuses are the source of the problem and, second, to check the fuses and replace them if necessary.

Before you start

Have these items ready to use:

- Two replacement fuses
- Flat-blade screwdriver
- A voltmeter or continuity meter

Checking and replacing the power-module fuses

- 1. Ensure that the microdispenser is plugged in. If it is, go on to Step 2.
- 2. Ensure that the Emergency Shutoff Switch is installed. The switch must be installed for the system to operate. If the switch is installed, go on to Step 3.
- 3. Ensure that the Emergency Shutoff Switch is turned on. Rotate the red button on the switch in the clockwise direction, as is shown in the arrows on the button. See Figure 5-16. If the Emergency Shutoff Switch is turned on, go on to Step 4.



Figure 5-16: Emergency Switch

Service and Maintenance of the Hydra DT Microdispenser



- 4. Check the power module fuses as follows:
 - a. Turn off power to the microdispenser and unplug the power cord from the surge suppressor.
 - b. Unplug the power cord from the microdispenser.
- 5. Insert the edge of the flatblade screwdriver in the slot at the top of the powermodule cover, and gently pry the cover open. See Figure 5-17 for a closeup view of the power module.



Figure 5-17: Power Module

6. Insert the tip of the screwdriver in the slot at the top of the red fuse box. Gently pry the box away from the microdispenser. Grasp the box with your fingers and remove it from the power module. See Figure 5-18.



Figure 5-18: Removing the Fuse Box

- 7. Use the voltmeter or continuity meter to check whether the fuses are live.
 - If the fuses are live, reinsert the fuse box in the power module. Then contact your equipment provider for assistance in determining the source of the problem. See page 1-9 for contact information.
 - If the fuses are not live, replace them as follows:
- 8. Lift out the fuses on both sides of the red fuse box.
- 9. Insert the new fuses.



- 10. Reinsert the fuse box in the power module and then close the power module cover.
- 11. Plug the power cord into the receptacle on the back of the microdispenser and then into the surge suppressor.

Service and Maintenance of the Hydra DT Microdispenser



6: Getting the Most Out of Your Hydra DT Microdispenser

Basics of Liquid Handling With the Hydra DT Microdispenser
Two Aspirate/Dispense Modes6-
Priming Syringes6-6
Setting Dispense Heights6-5
Using the Microdispenser in Cell Culture Applications
Creating Dot Blots and Membrane Arrays With the Vacuum Manifold
Using the Wash Operation as a Mix Operation
Using the Manually Operated Plate Positioners DT
Using the Hydra-96 Plate Positioner for Standard 384-Well Plates6-2
Using the Hydra-96 Plate Positioner for CyclePlate-384 and 192 Plates
Dispensing Onto the Sidewalls of Wells in CyclePlate-ET Plates With a Plate Positioner 6-2
Creating Membrane Arrays With the Vacuum Manifold and a Plate Positioner
Using the Hydra DT 96 With Wide Bore D.A.R.T.s With Resin Suspensions
General Guidelines for Using the Microdispenser With Resin Suspensions
Examples of Types of Materials Used in Resin Suspensions on the Hydra DT 96
System With Wide Bore D.A.R.T.s6-23
Using Viscous Solutions
Ensuring Compatibility of Chemicals Used With the Hydra DT Microdispenser 6-26

Basics of Liquid Handling With the Hydra DT Microdispenser

To use your Hydra DT microdispenser effectively, you need to first understand some of the basics of its operation. A discussion of (1) Aspirate/Dispense modes, (2) priming syringes, and (3) setting dispense heights is provided here to give you basic information on the fundamental operations of the Hydra DT microdispenser.

Two Aspirate/Dispense Modes

Different liquid handling applications require different Aspirate/ Dispense modes.

The Hydra DT microdispenser has two Aspirate/Dispense modes:

- Aspirate with Overstroke/Dispense Specific
- Aspirate with Pre Air Gap/Dispense All

Some of the terms used in the Hydra DT EPROM-file display differ from the ones used in the *ControlMate* software. Please see the following table.

Getting the Most Out of Your Hydra DT Microdispenser



Table 6-1: Related Terms in ControlMate Software for Hydra DT & in Hydra DT EPROM-File Display

ControlMate Term	EPROM-File Term
Pre Air Gap	Air
Dispense All	Empty
Dispense Specific	Dispense
Overstroke	Prime

The rest of this discussion about the two modes will use the *ControlMate* terms *unless* a distinction must be made between the two types of terms.

When is Aspirate with Overstroke/Dispense Specific the right mode to use?

The Aspirate with Overstroke/Dispense Specific mode is best used for procedures that require:

- Highest accuracy for submicroliter and small-volume dispenses
- Incremental dispensing
- Bulk transfers

Serial dispensing using this mode is one of the most efficient means of transferring microliter volumes into any number of 96-, 384-, or 1536-well plates. (Note that your Hydra DT system must be equipped with the X/Y Plate Stage and the accompanying *ControlMate* software to dispense into 1536-well plates.)

This mode allows the most accurate method of repetitive dispensing and also allows dispensing to multiple plates without the need to refill the syringes.

How is Overstroke used in Aspirate with Overstroke/Dispense Specific mode?

The Aspirate with Overstroke command obtains the desired, programmed volume in the syringes in this way:

- 1. It first draws in the specified Aspirate volume amount.
- 2. Then it overshoots that volume by drawing in an additional Overstroke volume.



Note: The Overstroke volume can be reset on the Set Device and Pipettors Options screen under the Add-Ins menu. The Prime volume in the EPROM files is pre-set to 5% of total syringe volume and is not a user-editable value.

3. Next it pushes the plungers back down, dispensing the overshoot volume into the source vessel, leaving the desired amount in the syringes without excess.



The actions of the Aspirate with Overstroke command force the syringes against the lower holding plate to seat them in preparation for dispensing. The syringes have been "auto-primed" (primed by default) one time. Matrix Technologies recommends that you follow this default priming with 3 to 5 more manual primes just before the Dispense operation is performed. See page 6-8 for more information about priming syringes.

After the Aspirate operation has finished, you use the Dispense command to dispense the programmed volume into the receiving plate. When you select the Specific option in the Dispense command, the command empties just the volume that you have set in your file as the Dispense Volume, rather than the entire contents of the syringe.

How do I program a file to use the Aspirate with Overstroke/ Dispense Specific mode and then run the file?

The following paragraphs provide a basic description of how to program a file in this mode and then run the file. For detailed information about creating, changing, and saving the files you will use to run protocols on the Hydra DT microdispenser, see Chapter 3 if you are using Manual Operation or Chapter 4 if you are using Automated Operation.

In Manual Operation:

To program a Hydra DT EPROM file to use the Aspirate with Overstroke/Dispense Specific* mode, use the following procedure. (*Note: In the EPROM files, the "Aspirate with Overstroke/ Dispense Specific" mode is actually "Aspirate with Prime/ Dispense.")

1. In your chosen Hydra DT file, set the specified Dispense and Aspirate parameters as follows:

Screen	Parameter	Value
Dispense	Vol	Specific incremental (partial) dispense amount required by your application.
Aspirate	Prime*	ON
	Vol	Aspirate volume sufficient to provide as many of your dispense volumes as the syringes will hold, allowing space in the syringe for the Prime volume*.
	Air	0 (zero)

*Prime volume is pre-set to 5% of the total syringe volume and is not a user-editable value.

2. Set the other file parameters (for example, Table Height and Speed that are correctly calibrated for your application and labware) in the file.

Getting the Most Out of Your Hydra DT Microdispenser



To run a Hydra DT EPROM file in the Aspirate with Overstroke/ Dispense Specific mode:

- 1. Access the Ready Mode screen for the file.
- 2. Put an empty reservoir on the tray table/stage and press **Empty** to empty the syringes.
- 3. Replace the reservoir with the source plate containing the sample.
- 4. Press **Aspr** to aspirate the Vol and the Prime volume into the syringes. (Vol should be large enough to allow you to dispense the Dispense Vol amount into as many of your receiving microplates as possible while still leaving space in the syringe for the Prime volume.)
- 5. Remove the source plate and place the destination plate on the tray table/stage.
- 6. Press **Dispense** to dispense Vol into the destination plate.
- 7. Continue to remove and replace destination plates and to press **Dispense** until the sample volume in the syringes is too low to provide more dispenses.
- 8. Repeat Step 3 through Step 7 as many times as needed for multiple source and destination plates.
- 9. When you are finished dispensing, press **Empty** to empty any remaining sample volume back into the source plate or into a waste reservoir.

In Automated Operation:

To program a sequence file to using Aspirate with Overstroke/ Dispense Specific mode, use the following procedure:

1. In your sequence file, set the specified Aspirate and Dispense parameters as follows:

Command	Parameter	Value
Aspirate	Overstroke*	Option is selected.
	Aspirate volume	Aspirate volume sufficient to provide as many of your dispense volumes as the syringes will hold, with just enough space in the syringe for the Overstroke volume.*
Dispense	Specific	Option is selected.
	Volume	Specific incremental (partial) dispense amount required by your application.
* Overstroke volume is set on the <i>Set Device and Pipettor Options</i> screen under the Add-Ins menu. If you need to fine-tune the value for your applications, you can reset it there.		

- 2. Set other command parameters in the file as needed.
- 3. Click the Validate File icon in the Toolbar. If any errors are listed, correct them and then re-validate the file.



To run a sequence file in the Aspirate with Overstroke/Dispense Specific mode:

- 1. Access a Hydra DT EPROM file, and check that the Empty (Table) height is set to a safe height.
- 2. Put an empty reservoir on the tray table or on the stage in the source nest under the dispensing head and press **Empty** to empty the syringes.
- 3. Replace the reservoir with the source plate containing the sample.
- 4. Access the sequence file in the ControlMate software.
- 5. If the system is a Hydra DT with X/Y Plate Stage system, place the first destination plate in the destination nest.
- 6. Click the **Run** icon on the Toolbar.
- 7. After the Aspirate with Overstroke command has executed: On the Hydra DT base-unit system or Hydra DT with Wash Pump Module, remove the source plate and place the destination plate on the tray table.
- 8. Continue to remove and replace destination plates as your sequence file requires.
- 9. Repeat Step 4 through Step 8 as many times as needed for multiple source and destination plates.
- 10. When dispensing is completed, press **Empty** on the Hydra DT microdispenser to empty any remaining sample volume back into the source plate or into a waste reservoir.

When is Aspirate with Pre Air Gap/Dispense All the right mode to use?

The Aspirate with Pre Air Gap/Dispense All mode is best used for procedures in which:

- Source plates have minimal sample volumes.
- Residual sample cannot be returned to the source plate after the transfer.
- Multiple samples are pooled.
- Individual samples of microliter volumes are transferred.

Use of this mode is necessary when there is insufficient sample to allow for the overshoot volume that occurs with the Aspirate with Overstroke command. Sample volumes of as little as 1.0μ L have been transferred using the Aspirate with Pre Air Gap/Dispense All mode.

Aspirate with Pre Air Gap/Dispense All mode is also preferred if the user does not want to draw excess sample into the syringes only to return the remainder of the sample to the sample plate after the transfer.

When pooling multiple samples, you use the Aspirate command to draw first an air gap (see Table 6-2 on page 6-9 for recommended air-gap volumes) and then the

Getting the Most Out of Your Hydra DT Microdispenser



samples into the syringes. Then you use the Dispense All command to deliver the pooled samples and the air gap into the final receiving plate.

Microliter volumes of individual samples can also be transferred using the Aspirate with Pre Air Gap/Dispense All mode. Here, as with pooling, you first draw an air gap (see page 6-9 for recommended air-gap volumes) into the syringes using Aspirate, and then you use Aspirate to draw the single volumes to be transferred into the syringes. Finally, you use the Dispense All command to deliver the entire contents of the syringes, including the sample volumes and the air gap, into the receiving plate.

Why are air gaps used in the Aspirate with Pre Air Gap/Dispense All mode?

Aspirating an air gap into the syringes at the beginning of a procedure in the Aspirate with Pre Air Gap/Dispense All mode increases accuracy in dispensing individual small volumes and helps to force all of the sample volume out of the syringes during the dispense. For details about using an air gap, see "Priming Syringes in Aspirate with Pre Air Gap/Dispense All Mode" on page 6-9.

How do I program a file to use the Aspirate with Pre Air Gap/Dispense All mode and then run the file?

The following paragraphs provide a basic description of how to program a file in this mode and then run the file. For detailed information about creating, changing, and saving the files you will use to run protocols on the Hydra DT microdispenser, see Chapter 3 if you are using Manual Operation or Chapter 4 if you are using Automated Operation.

In Manual Operation:

To program a Hydra DT EPROM file to use the Aspirate with Pre Air Gap/Dispense All mode, use the following procedure:

- 1. In your chosen Hydra DT file, set the specified parameters as follows:
- 2. Set the other file parameters (for example, Table Height and Speed that are correctly calibrated for your application and labware) in the file.

Screen	Parameter	Value
Aspirate	Prime	OFF
	Vol	Total sample volume
	Air	Recommended air gap for your system (See Table 6-2 on page 6-9.)

To run a Hydra DT EPROM file in the Aspirate with Pre Air Gap/Dispense All mode:

- 1. Access the Ready Mode screen for the file.
- 2. Put an empty reservoir on the tray table/stage and press **Empty** to empty the syringes.



- 3. Replace the reservoir with the source plate containing the sample.
- 4. Press Aspr to aspirate Vol and Air into the syringes.
- 5. Remove the source plate and place the destination plate on the tray table/stage in the nest under the dispensing head.
- 6. Press **Empty** to dispense the total syringe contents into the destination plate.

In Automated Operation:

To program a sequence file to use the Aspirate with Pre Air Gap/Dispense All mode, use the following procedure:

1. In your sequence file, set the specified Dispense and Aspirate parameters as follows:

Command	Parameter	Value
Dispense	All	(Option is selected.)
Aspirate	Pre air gap	(Option is selected.)
	Pre air gap volume	Recommended air-gap volume for syringe size (See Table 6-2 on page 6-9.)
	Aspirate volume	Total sample volume, or as much of sample as will fit in syringe with air gap

- 2. Set other command parameters in the file as needed. If your system is a Hydra DT with X/Y Plate Stage system, include Move commands that move the source and destination nests under the dispensing head.
- 3. Click the **Validate File** icon in the Toolbar. If any errors are listed, correct them and then re-validate the file.

To run a sequence file in the Aspirate with Pre Air Gap/Dispense All mode:

- 1. Access a Hydra DT EPROM file, and check that the Empty (Table) height is set to a safe height.
- 2. Put an empty reservoir on the tray table/stage in the nest under the dispensing head and press **Empty** to empty the syringes.
- 3. Replace the reservoir with the source plate containing the sample.
- 4. Access the sequence file in the *ControlMate* software.
- 5. On the Hydra DT with X/Y Plate Stage, place the destination plate in the destination nest.
- 6. Click the **Run** icon on the Toolbar.
- 7. On the Hydra DT base-unit system or Hydra DT with Wash Pump Module: Follow software-prompt instructions to remove the source plate and insert the destination plate.

Getting the Most Out of Your Hydra DT Microdispenser



Priming Syringes

After the Hydra syringes have been filled with the solution to be dispensed, the syringes must be "primed" to ensure accuracy and precision of the operation. To explain the reasoning behind the need to prime the syringes, a short discussion of the Hydra DT design is presented here. There is a slight amount of movement (referred to as "lash") of the glass syringes that is designed into the Hydra DT micro-dispenser such that at the beginning of a Dispense or Aspirate step, the syringe barrels move up or down together with the plungers until they contact the upper or lower holding plate. (Holding plates pro-vide vertical and horizontal positioning of the syringes within the head unit of the microdispenser.) During this short interval of "lash movement" the plunger does not move inside the barrel of the syringe, so no sample is dispensed or aspirated. After the syringe barrels have been seated against the holding plate, the plunger begins to move within the syringe and aspirating or dispensing of sample begins.

The way to account for the effects of lash is to "prime" the syringes. The following two subsections tell you how to prime the syringes, first for the Aspirate with Overstroke/Dispense Specific mode and then for the Aspirate with Pre Air Gap/Dispense All mode.

Priming Syringes in Aspirate with Overstroke/Dispense Specific Mode

Aspirate with Overstroke/Dispense Specific mode uses the Aspirate with Overstroke option to fill the syringes for a dispense operation. The Overstroke action provides initial priming. The microdispenser draws the desired amount of liquid into the syringes, overshoots this volume by the specified Overstroke volume (typically an additional 5-50µL), and then dispenses the extra volume back into the reagent reservoir, leaving the desired volume in the syringes. (The necessary amount of Overstroke volume depends on syringe size. In *ControlMate* software, it is set by the user on the Set Device and Pipettors Option screen under the Add-Ins menu. In the Hydra DT EPROM files, the (Prime) value is set to 5% of total syringe volume and is not user-editable.) This default priming forces the syringes against the lower holding plate to seat them in preparation for dispensing. Matrix Technologies recommends that immediately before the dispense is to be performed you supplement the default priming by dispensing the desired volume back into the sample reservoir 3-5 times. At this point the syringes will be seated firmly against the bottom holding plate so that any further downward movement of the plungers will result in the precise volume of fluid being dispensed from the syringes. The dispense operation should be carried out immediately after the last priming.



Priming Syringes in Aspirate with Pre Air Gap/Dispense All Mode

Aspirate with Pre Air Gap/Dispense All mode requires a method to account for lash other than the one used in Aspirate with Overstroke/ Dispense Specific mode. In an aspirate-transfer, the Aspirate command (with no Overstroke) is used to draw sample into the syringes. When an Aspirate command is executed, the plungers are immediately raised, resulting in: first, the joint upward movement of the syringes and plungers; second, seating of the syringe against the upper holding plate of the head; and finally, aspirating of liquid into the syringe needles. The result is less than the desired amount drawn into the syringes. To prevent this error, program your file so that an initial air gap (see Table 6-2, below) is drawn into the syringes before the Aspirate command is used to draw sample into the syringes. Here are recommended air gap volumes:

Syringe Size	Recommended Air Gap Volume
100μL, 290μL	10µL
580µL	50µL
1mL	100µL

Table 6-2: Recommended Air Gap Volumes

Drawing air into the syringes will seat them firmly against the top holding plate in preparation for aspirating.

Setting Dispense Heights

Dispense heights are set on the Hydra DT microdispenser by raising and lowering the tray table/stage. Different dispense heights are required for different types of dispense operations. If you are dispensing very small volumes onto dry plates, for example, you will set the height differently than you would for a larger-volume dispense into a plate that contained buffer.

The following figures illustrate various dispense scenarios, with dispenses into wet and dry plates and with differences in dispense volumes. The information presented here should help you determine the optimum dispense height for your applications.



Note: For many operations you need to set the tray table/stage height to a nearbottom-of-the-well position. For instructions on how to do this, see "Set the tray table/stage height" on page 3-7

Getting the Most Out of Your Hydra DT Microdispenser



Dispensing Into Wet Plates

Figure 7-1 through Figure 7-3 illustrate dispenses into wet plates. They show three dispenses with the needle tips at slightly different heights. (They also illustrate that dispense height can vary considerably more when dispensing into a wet plate than onto a dry plate and still result in a successful dispense.)

Table 6-3: Dispensing Into Wet Plates

Figure 6-1: Dispense into wet plate, with tip just touching solution Figure 7-1 shows the tip just touching the solution already in the well. This dispense height will allow effective delivery of sample to the well while minimizing the exposure of the needle tip to the solution in the well.
Figure 6-2: Dispense into wet plate, with tip submerged in solution Figure 7-2 shows the tip submerged in solution. The sample will be delivered successfully into the solution in the well.
Figure 6-3: Dispense into wet plate, with tip above solution Figure 7-3 shows the needle tip above the solution, with the drop on the tip not touching the solution. No sample is delivered during this dispense. The drop will remain on the tip.



Dispensing Onto Dry Plates

Figure 7-4 through Figure 7-7 illustrate dispenses onto dry plates.

Table 6-4: Dispensing Onto Dry Plates

Figure 6-4: 10µL dispense onto dry plate—drop fills gap between tip and well bottom. Touch-off occurs.
Figure 7-4 shows a 10µL dispense onto a dry plate. For this type of dispense, you set the tips so that they are close to but not touching the bottom of the well — a "near bottom" position. The drop on the tip of the tip fills the gap between the tip and the bottom of the well. When the tip is retracted, the drop will "touch off" onto the well bottom. The dispense is successful.
Figure 6-5: 1μ L dispense onto dry plate—drop unable to bridge gap between needle tip and well bottom. Drop remains attached to tip. Figure 7-5 shows a needle tip at the same dispense height as the one in Figure 7-4. For this dispense, however, the dispense amount is very small— 1μ L. As illustrated, the drop is not large enough to fill the gap between the tip and the bottom of the well, and the drop remains attached to the tip, so there is no "touch off." The dispense is unsuccessful. Solution: Reset the Dispense height parameter so the tips are touching the well bottom during the dispense.
Figure 6-6: 1µL dispense onto dry plate—tip touches plate bottom and drop dispenses around tip onto well bottom. Figure 7-6 shows the tip at a dispense height that just touches the tip to the bottom of the well. To set this height, move the tray table/stage up so that you can feel that some of the needle tips are just touching the well bottom, and then move the tray table/stage up one or two more clicks. You can check whether tips are touching the plate by trying to lift the front of the plate with your forefinger. If the plate does not lift, the tips are touching the plate. This figure shows the same 1µL dispense amount as in Figure 7-5. This time, the dispense is successful because the drop squeezes out from the tip onto the bottom of the well and touches off as the tray table/stage drops down. Because the amount of dispense is so small (<3µL), pressure does not build back up into the syringe even though the needle tip is pressing down onto the plate.
Figure 6-7: 10μ L dispense onto dry plate; tip touches plate bottom. Drop dispenses around tip onto well bottom but cannot dispense quickly enough to prevent pressure buildup. Figure 7-7 shows the tip in the same position as in Figure 7-6, with the tip touching the bottom of the well. In this instance the dispense amount is 10μ L, and the needle cannot dispense the amount quickly enough to keep the pressure from building back up into the syringe. This pressure buildup is likely to damage the syringe. Solution: Reset the Dispense height parameter so the needle tips are not touching the well bottom of the plate.

Getting the Most Out of Your Hydra DT Microdispenser





Caution: In general, do not allow the tips to contact the bottom of any plate, tube, reservoir, tray, or wash basin. The only time the tips need to touch the bottom of the plate is when they are dispensing volumes of 3μ L or less onto a dry plate (as shown in Figure 7-6). For dispense volumes greater than 3μ L, the size of the droplet formed on the tip will be large enough to bridge the gap between the tip and the bottom of the plate. In such cases, set the dispense height at two to three clicks back from touching the plate. If the tips touch the plate during a dispense with volume greater than 3μ L, backpressure will build up in the syringes (as shown in Figure 7-7), because the sample cannot exit the tips as fast as the syringe plunger is pushing the sample out. That backpressure can damage the syringes. If a syringe is damaged during operation, turn off power to the unit immediately by pressing the Emergency Shutoff Switch.

For More Detailed Information About Hydra Microdispenser Operation

For more detailed information about the microdispenser's operation, see the Hydra Technical Data page on the Matrix Technologies website at *http://www.matrixtechcorp.com/*.

Dispensing Onto the Sidewalls of Wells in CyclePlate-ET Plates

To dispense droplets of different solutions into a microplate well without touching the needle tips or the solution to be dispensed to the current contents of the wells, set up the instrument to dispense onto the sidewalls rather than into the center of the well. The following procedure describes how to dispense solutions onto the sidewalls of the 96 wells in a CyclePlate-96ET without touching any other solution currently found in the wells (you can use the same procedure with CyclePlate-192 and -384 microplates).



Note: In Manual Operation, this procedure requires use of the split-well manually operated plate positioner, the Hydra-96 Plate Positioner for CyclePlate-384 and -192ET, Matrix Technologies Catalog No.1029-41-0.


In Manual Operation:

- 1. Place a CyclePlate-96 ET PCR plate in the split-well Hydra-96 Plate Positioner for CyclePlate-384 and -192ET (manually operated plate positioner).
- 2. Transfer the assembly onto the tray table of the Hydra DT 96 micro-dispenser.
- 3. Set the Dispense (Table) height so the needle tips are just touching the sides of the conical wells above the expected level of solution when the sample is transferred.
- 4. Set all other file parameters as needed.
- 5. Remove the microplate and replace it with an empty reservoir identical to that containing your sample solution. Set the Aspirate Volume, Aspirate (Table) height, and Dispense (Table) height parameters accordingly.
- 6. Remove the empty reservoir and replace it with the source reservoir containing your sample(s).
- 7. Fill the syringes with the sample solution and then remove the reservoir.
- 8. Remove the source reservoir and position your first destination CyclePlate-96 ET microplate in the manual plate positioner. Place the positioner with the CyclePlate on the tray table.
- 9. Dispense the solution into the microplate. The droplet should just touch off onto the side of the well.
- 10. Repeat Step 8 and Step 9 for as many microplates as you want to fill with the sample solution.
- 11. Change the D.A.R.T.s as needed between multiple samples.
- 12. Repeat steps 1 11 for each new solution sample plate, advancing the split-well positioner to the next position each time that you change solutions. You can perform this procedure for four separate sample or reagent solutions so that each one is placed on a different side of the well.
- 13. Centrifuge the destination plate after the final solution addition to pool all the reagents in the bottoms of the wells.

Figure 6-8 and Figure 6-9 on page 6-14 illustrate use of the Hydra-96 Plate Positioner for CyclePlate-384 and -192 (a manually operated, split-well plate positioner, Matrix Technologies Catalog No. 1029-41-0) to dispense solutions onto the sidewalls of Cycle-Plate wells.

Getting the Most Out of Your Hydra DT Microdispenser



In Automated Operation:

On the Hydra DT with X/Y Plate Stage system, use tip/well offset settings on the *Move to Position command* screen to position the depth and X-Y placement of the tips in relation to the centers of the wells.

On the Hydra DT base-unit system, use the manual plate positioners as described previously.

Table 6-5: Dispensing Solutions Onto Sidewalls of CyclePlateMicroplate Wells

Figure 6-8: Needle Position for PCR Plate on Tray Table When a CyclePlate-96ET plate is used on the Hydra DT microdispenser tray table, the needle tip will dispense into the center of the well, as shown in Figure 7-8.
Figure 6-9: Needle Position for PCR Plate on Plate Positioner Use of the Hydra-96 Plate Positioner for CyclePlate-384 and -192 plates allows you to dispense solution onto the side of a CyclePlate well rather than into the center. This technique guards against cross-contamination of samples by allowing solution to be added to a well without touching any sample currently in the plate. Figure 7-9 shows the needle position.



Using the Microdispenser in Cell Culture Applications

Some laboratories have reported success in using the Hydra DT micro-dispenser to transfer cells and cell culture media. To create the required sterile environment, take the following steps:

1. Place the Hydra DT microdispenser in a laminar flow tissue culture hood. Best results will be achieved if the unit is left in the hood. An initial wipedown of the Hydra DT microdispenser with ethanol and paper towels is recommended. You can wipe down the instrument periodically whenever you similarly clean the inside of the culture hood.



Note: The Hydra DT microdispenser can be kept in a hood with the UV (ultraviolet) light on for extended periods. However, the plastic components on the outside of the Hydra DT case (that is, the switches, the cover over the display, and the data-port connectors) might be damaged after prolonged UV exposure. If those parts become corroded, you will need to replace them.



Caution: Do NOT pass syringe needles through the flame of a Bunsen burner.

- 2. You can sterilize the syringes initially if you remove them from the Hydra DT microdispenser and soak them in a 100% ethanol solution. If you do this, you must ensure that each syringe remains paired with its original plunger tip throughout the lifespan of the plunger tip. (See Table 5-3 on page 5-4 for PTFE tip replacement guidelines.)
- 3. A less laborious approach would be simply to fill the syringes with a 100% ethanol solution, leave the reservoir with the ethanol solution on the tray table/stage, and leave the tray table/stage in the up position so that the needle tips are submerged in the ethanol solution. To leave the syringes soaking in the ethanol solution: Place the reservoir containing the ethanol solution on the tray table/stage. In an EPROM file, set Vol to full volume. Before you press the **Aspr** button, position yourself so you can easily reach the power switch on the back of the microdispenser. Press **Aspr**, and when the syringes are full and the tray table/stage is up so that the needle tips are submerged by at least 2cm, press the power switch to **OFF**. Now both the needle tips and the interior of the syringe barrels will be bathed in the ethanol solution during the storage interval. After the syringes are soaked, the ethanol should be discarded and the syringes rinsed with 3 wash cycles of sterile dH,O.

Getting the Most Out of Your Hydra DT Microdispenser



- 4. When the Hydra DT microdispenser is used to transfer culture media or media containing suspended cells, keep in mind that ethanol may lyse cells. To minimize the chances of cell lysis inside the syringes: Wash the syringes between uses of different media or reagents with 3 wash cycles of ethanol and as many as 9 wash cycles of autoclaved dH₂O. Use fresh sterile water for every 3 wash cycles.
- 5. To further ensure that the syringes are clean, you can wash the syringes with a nonionic detergent solution such as Coulter[®] Clenz (Beckman Coulter cat.#s 8546929-31). If you use a detergent wash, ensure that all residual detergent is removed by washing the syringes with multiple water washes after the detergent wash.



Creating Dot Blots and Membrane Arrays With the Vacuum Manifold

The Hydra DT 96 base-unit system and Hydra DT 384 base-unit system microdispensers can be used to prepare dot blots and membrane arrays of various densities. To prepare a simple dot blot, put several pieces (approximately 5) of filter paper on the tray table of the microdispenser and lay a membrane (dry or pre-wetted) on top of the paper. You can also place the filter paper and membrane in a Multi-Blot® tray (Catalog Nos. 1039-50-0 [standard] and 1039-51-0 [sterile]) for this purpose if you wish. Set the dispense height so that the needles will press the membrane down onto the filter paper but will not penetrate it when sample solutions are dispensed.

For creating arrays of densities greater than 96 samples on the membrane, with the samples placed within the spacing of a standard microplate, Matrix Technologies recommends using our vacuum manifold (Catalog No. 1029-42-0) because of the following advantages:

- The membrane is anchored firmly by the vacuum, so it will not shift during multiple spottings.
- The vacuum manifold ensures a uniform vacuum across the membrane and helps limit diffusion of the sample as it is being applied to the membrane, giving a more concentrated sample and a smaller spot size that allows higher density arrays.

To use the vacuum manifold, follow these steps:

- 1. Place the manifold on the Hydra DT microdispenser tray table and connect it to a vacuum source.
- 2. Place a dry or pre-wetted membrane on the manifold. Ensure that the membrane covers the top of the manifold completely but is not larger than the top of the manifold.
- 3. Adjust the Dispense (Table) height parameter so that the needles hover above the membrane by approximately 1mm. This height setting will allow effective touch off during dispense while avoiding penetration of the membrane by the needle tips.
- 4. Run the Hydra DT microdispenser dispense protocol to spot solutions containing target molecules onto the membrane.

Getting the Most Out of Your Hydra DT Microdispenser



With the Hydra DT 96 base-unit system you can use the standard plate positioner (see "Using the Manually Operated Plate Positioners for the Hydra DT 96 Microdispenser" on page 6-19) with the vacuum manifold to create membrane arrays with a density of 384 samples. Figure 6-10 shows the two manually operated plate positioners and the vacuum manifold.



Figure 6-10: Manually Operated Plate Positioners and Vacuum Manifold



Note: With the Hydra DT with X/Y Plate Stage system, you can also use the manifold to create membrane arrays with densities of 1536 to 9600 samples.

For more information on the vacuum manifold and other accessories for the Hydra DT microdispenser, including the manual plate positioners, and on MultiBlot trays, see the Matrix Technologies website at *http://www.matrixtechcorp.com/*.



Using the Wash Operation as a Mix Operation

In Manual Operation mode, you can use the Wash operation to mix multiple solutions added sequentially to the wells of a microplate. (In Automated Operation mode, use the Mix command. See page 4-65 for information about the Mix command.)

Dispense each of the solutions you will mix into the same micro-plate, one after the other. Set the Wash (Table) height close to the bottom of the plate and the wash volume (Wash Vol) to draw up less than the total volume of the combined solutions into the syringes. Push the Wash button. All the combined solutions (less a small amount) will be aspirated and then emptied back into the microplate.

When you set up your file parameters for this operation, the Wash (Table) height parameter must be set so the needle tips are submerged below the top of the combined solutions and so that no air is drawn into the syringes when the wash volume is drawn up from the microplate.

Using the Manually Operated Plate Positioners DT

To accommodate the well spacing of different 96-well and 384-well microplates, Matrix Technologies offers two manually operated plate positioners for the Hydra DT 96 microdispenser for dispensing into 384-well plates:

- Hydra-96 Plate Positioner for Standard 384 Well Plates (Catalog #1029-41-5)
- Hydra-96 Plate Positioner for CyclePlate[®]-384 and -192 (Catalog #1029-41-0)

Figure 6-10 on page 6-18 shows the manually operated plate positioners.

The positioner for standard 384-well plates is based on the 4.5mm center-to-center spacing found in 384-well plates that adhere to the Society for Biomolecular Screening (SBS) standards for size and shape. When the positioner is used to create membrane arrays of 384 samples, the spots on the membrane will also have a spacing of 4.5mm.

Getting the Most Out of Your Hydra DT Microdispenser



The "split-well" positioner is needed when using the Hydra DT micro-dispenser to transfer liquid to and from CyclePlate[®]-192ET and 384ET microplates. You can also use it with the CyclePlate-96, -192, or -384 ET plates to dispense solutions onto the sidewalls of the wells rather than into the centers of the wells. See page 6-21 for information about dispensing onto sidewalls of wells. This positioner cannot be used with 384-well plates that adhere to the SBS standards because the positioner does not align a plate according to the 4.5mm spacing grid of the standard plates.

See Figure 6-11 for illustrations of microplate well-center-to-well center spacing.



Figure 6-11: Well-Center-to-Well-Center Spacing for SBS Microplates and Matrix Technologies' CyclePlate-192 and -384 ET Microplates



6-21

Using the Hydra-96 Plate Positioner for Standard 384-Well Plates

- 1. Place the positioner on the Hydra DT 96 tray table.
- 2. Load the plate onto the positioner. (The CyclePlate-384DW ("discrete well," thin wall) microplate requires that Matrix Technologies' removable adapter tray (Matrix Technologies' Catalog No. 1029-41-2) be placed on the positioner first to secure the plate properly.)
- 3. Turn the thumbwheel on the front of the positioner to move it sequentially to each of four positions for dispensing or aspirating liquid to or from each of the four sets of 96 wells in the 384-well plate.

Using the Hydra-96 Plate Positioner for CyclePlate-384 and 192 Plates

The split-well positioner is used in much the same way as the standard positioner. You may find it more efficient to load the CyclePlate microplates onto the positioner with the positioner removed from the Hydra tray table.

Dispensing Onto the Sidewalls of Wells in CyclePlate-ET Plates With a Plate Positioner

See page 6-21 for information on using the Hydra-96 Plate Positioner for CyclePlate-384 and -192 (Matrix Technologies' split-well, manually operated plate positioner) to dispense solutions onto the sidewalls of wells in CyclePlate-96, -192, or -384ET microplates.

Creating Membrane Arrays With the Vacuum Manifold and a Plate Positioner

You can use the Hydra-96 Plate Positioner for Standard 384 Well Plates with the vacuum manifold to create membrane arrays of up to 384 samples. See "Creating Dot Blots and Membrane Arrays With the Vacuum Manifold" on page 6-17 for more information about this use of the manifold and plate positioner for creating 384-sample membrane arrays.

Getting the Most Out of Your Hydra DT Microdispenser



Using the Hydra DT 96 With Wide Bore D.A.R.T.s With Resin Suspensions

The Hydra DT 96 system with R/B syringes was designed to allow multichannel dispensing of insoluble resins and beads into 96-well microplates or blocks. The Wide Bore D.A.R.T.s design for this Hydra DT model incorporates a larger-bore tip than those of the standard D.A.R.T.s types. These design features facilitate the passage of the suspensions, preventing the solid particles from collecting and clogging the tips. When presented as homogenous suspensions, beads of up to $300\mu m$ in diameter (dry) have been drawn into the Wide Bore D.A.R.T.s of the Hydra DT 96 system and dispensed uniformly into 96-well receiving plates and blocks.

General Guidelines for Using the Microdispenser With Resin Suspensions

Keep these two points in mind as you use the Hydra DT 96 system with Wide Bore D.A.R.T.s with resin suspensions:

- The most important factor in the accuracy of delivery is that the suspension be maintained in a homogeneous state so that each syringe in the array and each dispense will contain the same concentration of the bead or resin.
- Each resin has its own characteristic behavior in suspension. Some resins have been dispensed in concentrations of up to 5%, while others can only be transferred when in concentrations of 2% or less. The principal difficulty for using some resins at the higher concentrations is that the particles begin to clump and aggregate, which can clog the syringes. Each new resin should be thoroughly tested by gradually increasing the percent concentration to verify the highest concentration that will allow trouble-free dispensing with the Hydra DT 96 system with Wide Bore D.A.R.T.s.



Note: If you have questions about types of beads and resins that can be used with the microdispenser, contact your equipment provider. See page 1-9 for contact information.



Examples of Types of Materials Used in Resin Suspensions on the Hydra DT 96 System With Wide Bore D.A.R.T.s

Examples of the types of solid materials that have been successfully used with the Hydra DT 96 system with R/B syringes and descriptions of its use follow.

Suspension Material	Bead Diameter Before Swelling	Concentration Tested
Sephadex® G-50	=<160µm	2%
Tentagel S NH2 (in DCM/DMF)	300µm	2%
Yttrium Silicate (for SPA)	2 – 5µm	20mg/mL
PVT (for SPA)	5µm	20mg/mL
CPG	500 angstrom	0.25% (w/v)

Table 6-6: Examples of Materials Dispensed With the Hydra DT 96 System With Wide Bore D.A.R.T.s

- Solid supports for chemistry—The solid supports used in chemical syntheses can be transferred using the Hydra DT 96 system with Wide Bore D.A.R.T.s when they are prepared as an isobuoyant mixture. Typically, the amount of resin needed to give a 2% (w/v) solution is weighed out and mixed with the appropriate volume of 1:1 DCM/DMF (dichloromethane/dimethylformamide). Small volumes (1mL 5mL) of DCM or DMF are added incrementally to cause the resin to rise or settle respectively within the solvent mix. Eventually the solvent mix is adjusted so that the resin neither rises nor settles. It is isobuoyant and is said to "float." The suspension can then be transferred into a receiving plate or block. Rink resins, Wang resins, Tentagels, ArgoPore® and ArgoGel® (Argonaut Technologies), SPA (scintillation proximity assay, Amersham Pharmacia BioTech) beads, and others have been successfully transferred using this method with the Hydra DT 96 system with Wide Bore D.A.R.T.s.
- SPA beads—For this application, SPA (scintillation proximity assay, Amersham Pharmacia BioTech) beads are suspended in an aqueous solution. They have a tendency to settle. This settling could present a problem if the beads were large or tended to form aggregates. But SPA beads are generally sized on the order of 5µm and remain in a uniform suspension long enough to be drawn into the syringes of the Hydra DT 96 system with Wide Bore D.A.R.T.s and subsequently dispensed. When SPA beads of yttrium silicate were tested in Matrix Technologies' laboratories, it was found that even if the beads were allowed to settle within the syringe barrels, the Hydra DT 96 system with Wide Bore D.A.R.T.s was still able to dispense them with uniform results across the syringe array.

Getting the Most Out of Your Hydra DT Microdispenser



- Sephadex beads—Sephadex (Amersham Pharmacia Biotech) beads have been used in applications in high-throughput laboratories using 96-well filtration blocks to purify cycle-sequencing reaction products prior to loading onto the gel of an automated sequencer. (For more information about this application, see the article "Reduced Volume AmpliTaq DNA Polymerase FS Cycle Sequencing Reactions in the Robbins CyclePlate® 384" on the Hydra Technical Data page on the Matrix Technologies website at *http://www.matrixtechcorp.com/*). The Hydra DT 96 system with Wide Bore D.A.R.T.s is used to transfer the Sephadex slurry into the filtration plates. Despite the fact that the suspension settles quickly within the syringes, Sephadex G-50 beads of less than 160µm diameter can be drawn into the syringes uniformly and dispensed even after the beads have settled.
- CPG beads—CPG (controlled pore glass) beads have been used as the solid support in protocols designed for the MerMade DNA oligonucleotide synthesizer (available from BioAutomation). The diameter of these beads averages approximately 500 angstrom and does not clog the syringes despite the beads' near-immediate settling within the syringe barrel.



6-25

Using Viscous Solutions

In general, do not use the microdispenser to dispense viscous solutions. The microdispenser is designed primarily to dispense clear, aqueous solutions of low viscosity.

Hydra DT microdispensers are available with syringe sizes ranging from 0.5μ L-100 μ L and 5μ L-300 μ L. Matrix Technologies studied the dispensing limits for several Hydra microdispenser models. Test results indicated that the larger bore syringe-andneedle combinations are capable of filling and dispensing the more viscous solutions.

Data for the methylcellulose solutions showed that both the Hydra DT 96 and the Hydra DT 384 with 100μ L syringes should be used with methylcellulose solutions of less than 26 cps viscosity.

Table 6-7: Pass/Fail Test Results for Filling Syringes to Full Volume With Solutions of Varying Percent Glycerol Composition

	Percer	Percent Glycerol					
	30	50	60	75	80	90	100
Hydra DT 384 100µL	+	+	-				
Hydra DT 96 100μL	+	+	+	-			
Hydra DT 96 290µL	+	+	+	+	+	-	

Table 6-8: Pass/Fail Test Results for Filling Syringes to Full Volume With Methylcellulose Solutions of Varying Viscosity

	Viscosity (cps)						
	25	50	100	250	400	1000	1500
Hydra DT 384 100µL	+	1					
Hydra DT 96 100μL	+	-					
Hydra DT 96 290µL	+	+	+	-			

For more information about these tests, refer to the article "Dispensing Viscous Fluids with the Hydra Microdispenser" on the Hydra Technical Data page on the Matrix Technologies website at <u>http://www.matrixtechcorp.com/</u>.

If you have any questions about using the microdispenser to dispense solutions with some limited viscosity, contact your equipment provider. See page 1-9 for contact information.

Getting the Most Out of Your Hydra DT Microdispenser



Ensuring Compatibility of Chemicals Used With the Hydra DT Microdispenser

To protect your microdispenser from damage and to ensure the integrity of your results, you need to know which chemicals (acids, bases, organics, etc.) can be safely used with the microdispenser and how to use them.

The basic guideline for the use of corrosive or oxidizing agents with the microdispenser is not to allow such agents (chlorine bleach, strong acid, etc.) to be in continuous contact with the microdispenser. Such agents can be used routinely with the Hydra DT microdispenser but may damage the case and the tray table/stage if left in contact with the instrument for extended periods of time. If you need to use these reagents in a protocol, ensure that you clean the microdispenser of any residual corrosive agent as soon as you finish the procedure. If you need to use any strong oxidizing or corrosive agent other than those listed in Table 7-9, contact your equipment provider. See page 1-9 for contact information. They will give you guidance on how to use these agents in such a way as to protect the microdispenser and to ensure the integrity of your samples.

Table 7-9 lists chemical reagents that have been used routinely in the Hydra DT microdispensers.

Acetic acid	Diisopropylamine	Methanol	Trifluoroacetic Acid
Acetone	Dimethylacetamide	Methylene Chloride	Trimethyl Orthoformate
Acetonitrile	Dimethyl Formamide	Methylene Chloride/TFA, 1:1	Xylenes
Ammonia 27-30%	Dimethyl Sulfoxide	Pyridine	Toluene
Chloroform	Ethanol	Triethylamine	



7: Troubleshooting and Tips

Troubleshooting Topics

Scan the following table to see if your question is listed.

Table 7-1: Troubleshooting Topics

If the problem is See p		
1. The system doesn't turn on when I press the power switch to ON.	7-2	
 The microdispenser is behaving erratically; for example, the system shuts off suddenly or recycles to the beginning of a file. 	7-2	
3. Nothing happens when I press the Dispense button. The power to the system is on and the LED display is on.	7-3	
4. One of the microdispenser's motors isn't working properly.	7-3	
5. The tray table doesn't move when I press the microdispenser buttons. I hear a grinding noise.	7-3	
6. When I place a plate on the tray table, the needles don't line up properly in the wells.	7-3	
7. The syringe barrels are breaking frequently.	7-4	
8. When I use the microdispenser to transfer small volumes, the syringes dispense uneven amounts. I've been using the Aspirate with Overstroke/Dispense Specific mode to fill the syringes with the solution to be transferred.	7-4	
 The seal on the PTFE tips appears to be defective. I can see liquid creeping up inside the barrel above the tips. 	7-5	
10. One or more syringes are not dispensing properly.	7-5	
11. When the microdispenser is doing a dry-plate dispense, some of the wells aren't receiving any sample.	7-5	
12. How do I prevent contamination between sample preparations?	7-5	
13. I need to replace some syringes	7-5	

Troubleshooting and Tips



- 1. The system doesn't turn on when I press the power switch to ON.
 - a. Ensure that the microdispenser is plugged in.
 - b. Ensure that the Emergency Shutoff Switch is installed. The switch must be installed for the system to operate.
 - c. Ensure that the Emergency Shutoff Switch is turned on. Rotate the red button on the switch in the clockwise direction, as is shown in the arrows on the button.

See Figure 7-1.



Figure 7-1: Emergency Shutoff Switch

- 2. The microdispenser is behaving erratically. The following have occurred recently:
 - System shuts off suddenly.
 - The display on the front of the microdispenser lights up and numbers in the display change when microdispenser buttons are pressed, but the syringe head and the tray table do not move.

The problems are most likely caused by electrical interference. Take the following steps:

- a. Ensure that the microdispenser is plugged into a surge suppressor.
- b. Check to see if there are other electrical appliances on the same line with the microdispenser. If there are, they could be causing the disturbance. Test this theory by removing the appliances from the line and then testing the microdispenser operation.



If the preceding steps do not fix the problem, then problems with either the printed circuit board (PCB) or the power supply are possible. Do not attempt to fix a PCB or power supply problem; contact your equipment provider. See page 1-9 for contact information. Be sure to tell them all of the "symptoms" that your microdispenser is displaying.

3. Nothing happens when I press the Dispense button [in Manual Operation]. The power to the system is on and the display is on.

You might not be in the right place in the file you are using. To run a procedure, you must be at the *ready-mode* screen. Press **Page** until you see the *ready-mode* screen.

4. One of the microdispenser's motors isn't working properly.

If either or both of the microdispenser motors (the syringe motor is the top motor; the tray table/stage motor is the bottom motor) isn't working properly, a printedcircuit board (PCB) problem or power supply problem is possible. Do not attempt to fix a PCB or power supply problem; contact your equipment provider. See page 1-9 for contact information. Be sure to tell them all of the "symptoms" that your microdispenser has displayed.

5. The tray table doesn't move when I press the microdispenser buttons [in Manual Operation]. I hear a grinding noise.

The problem could be that the tray table has moved below the limit switch (sensor). To readjust the tray table, access one of your Hydra DT files. Press **Page** until you see one of the tray-table height parameters in the display. Press the **Up** button several times to raise the tray table. Now the table will be above the sensor and should work properly. If it doesn't, the problem might be with the tray table motor. A printed circuit board (PCB) problem or power supply problem is possible. Do not attempt to fix a PCB or power supply problem; contact your equipment provider. See page 1-9 for contact information. Be sure to tell them all of the "symptoms" that your microdispenser has displayed.

6. When I place a plate on the tray table/stage, the needles don't line up properly in the wells.

a. (On the Hydra DT base-unit system and Hydra DT with 3-Position XY Stage) Probably you need to adjust the alignment of the tray table. Raise the tray table so that the needles are close to the plate and you can see where the needles align with the wells in the plate. (To move the tray table, you need to access one of your Hydra DT files and adjust one of the tray-table height parameters using the Up and Down buttons on the front of the Hydra DT case.

Troubleshooting and Tips





See Chapter 3 for information on setting tray table heights.) Now use the Allen wrench that came with the microdispenser to loosen the two screws underneath the tray table. Loosen them one ½-counterclockwise turn so that you can move the tray table slightly side to side. Loosen and tighten the two screws to adjust the tray table position until you see that the needle tips are aligned with the well centers. Then tighten the screws firmly, but do not overtighten them.

- b. (On the Hydra DT base-unit system and Hydra DT 3-Position XY Stage) If you are using a manual plate positioner on a Hydra DT 96 model's tray table, the problem might be that you are not quite clicking the positioner into one of its four positions. As you rotate the turnwheel on the positioner to move from one position to the next, ensure that the positioner clicks into place for each position.
- c. On a Hydra DT with 3-Position X/Y Stage system, the stage or labware might be miscalibrated. Recalibrate the stage or labware and then check the needle-to-well-center alignment.

7. The syringe barrels are breaking frequently.

Probably the tips are coming in contact with the labware on the tray table/stage, and the tips are becoming blocked. When that happens, pressure can build up in the syringes and damage the syringes. If a syringe is damaged, immediately turn off power to the system by pressing the **Emergency Shutoff Switch**. To fix the problem of syringe barrels breaking frequently, try setting the tray table/stage height so that the needle tips do not press into the lab-ware (microplate, reservoir, etc.) on the tray table/stage. See page 6-9 for information about how to determine the best dispense height for your application.

8. When I use the microdispenser to transfer a small volume, the tips dispense uneven amounts. I've been using the Aspirate with Overstroke/Dispense Specific mode to fill the syringes with the solution to be transferred.

Use an aspirate-transfer protocol to transfer a small volume. You should always include an air gap in an aspirate-transfer operation. To do an aspirate-transfer operation, aspirate first an air gap and then the sample solution into the syringes. Then use the Empty command [in Manual Operation] or the Dispense All command [in Automated Operation] to empty the solution into the destination plate. This procedure will give you more precise results for an aspirate-transfer than a protocol that uses the Aspirate with Overstroke/Dispense Specific mode to transfer a small volume. For details on why you should use the Aspirate with Pre Air Gap/Dispense All mode for small-volume transfers, see page 6-5.



7-5

9. The seal on the PTFE tips appears to be damaged. I can see liquid creeping up the inside of the barrel above the tips.

The PTFE tips probably need to be replaced. See "Checking the PTFE Tips" on page 5-4. If the tips are worn out, you should replace all of them.

10. One or more syringes are not dispensing properly.

- a. If the syringes are dispensing uneven volumes, then the problem might be that the PTFE tips are wearing out and should be replaced. See "Checking the PTFE Tips" on page 5-4.
- b. If the problem is not worn-out PTFE tips, then one or more syringes might be damaged. Do a careful visual check for any broken or cracked syringes. If you find any damaged syringes, replace them. See the procedure for replacing syringes on page 5-7. If you don't see any damaged syringes, go on to Step c.
- c. Follow these Cautions:



Caution: Put on safety glasses and gloves before you check for damaged syringes.

Use one of the following tests for syringe damage: (1) dispense dye into a plate and see if some of the syringes do not dispense, or (2) dispense a microliter of distilled water into the air and see if a bead forms on the ends of the tips. If a bead doesn't form on the end of a tips, then that particular syringe is damaged and needs to be replaced. If any of the syringes are damaged, replace them. See the procedure for replacing syringes on page 5-7.

11. When the microdispenser is doing a dry-plate dispense, some of the wells aren't receiving any sample.

The dispense height might need to be reset. See page 6-9 for information about setting dispense heights to dispense onto dry plates.

12. I need to replace some syringes.

See Section 5-7 of the Hydra DT User Manual. This section details the necessary steps for replacing Hydra DT syringes as well as removing the pipetting head for return to the Matrix Technical Service Department.

13. Air is aspirating into the syringes along with the sample solution.

Either the height of the tray table/stage is too low or the microplate wells don't contain enough solution for the needle tips to be submerged when aspiration takes

Troubleshooting and Tips



place. Adjust the height of the tray table/stage or the volume in the well as needed.



Quick Tips for Using the Hydra DT Microdispenser

The following tips provide answers to some frequently asked questions about using and maintaining the microdispenser.

Table 8-2: Quick Tips for Using the Hydra DT Microdispenser

How do I	See			
Use the unit safely?	Warnings and Cautions on page ix			
Change the syringe size on the microdispenser?	Contact your equipment provider. See page 1-9 for contact information.			
Find the firmware version for the system?	Note about Firmware Version on page 3-9			
Keep needle tips from touching the tray table/ stage?	"Set the tray table/stage height" on page 3-7			
Create an air gap in the syringes?	"Priming Syringes in Aspirate with Pre Air Gap/Dispense All Mode" on page 6-9			
Keep the microdispenser in good working order?	Chapter 6, Service and Maintenance of the Hydra DT Microdispenser			
Send the unit in for repair?	Warranty on page 1			
Know when to change PTFE tips?	"Checking the PTFE Tips" on page 5-4			
Change a broken syringe?	"Replacing Syringes" on page 5-7			
Connect the microdispenser to a computer so that functions can be controlled through an RS-232 port using an ASC bi-directional communications protocol?	Appendix D, Integrating the Hydra DT Microdispenser Into an Automated Laboratory			

Troubleshooting and Tips

