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OroFlex Personal Pipettor Deluxe

Orochem Technologies Inc.



# OROFLEX PERSONAL PIPETTOR DELUXE



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## INTRODUCTION

OroFlex Personal Pipettor Deluxe is an affordable, compact and easy-to-use multichannel pipetting systems. It offers 96 channel pipetting combined with the ability to do serial dilutions on the same platform.

The 96-channel pipettors can be used with 24-well, 96-well or 384-well microplate applications by installing the appropriate head. Because of the unique interchangeable head design that separates the pipetting mechanism from the pipetting tips, the user can easily install a head that is suited to the required task at hand. The head design also allows the cost of the heads to be very reasonable so that it is feasible to maintain a library of heads that can be used for different tasks. This means that a 96 or 384 channel pipettor can easily be adapted in the future for different uses with minimal additional investment.

OroFlex Personal Pipettor Deluxe can be used for a variety of liquid handling tasks including reagent & solvent addition, plate replication, plate reformatting, and serial dilutions by rows or columns.

## FEATURES & BENEFITS

Feature	Benefit
Interchangeable pipetting head	<ul style="list-style-type: none"> <li>• Pipettes into 96 and 384 standard and deep well labwares</li> <li>• Wide Choices of disposable tip sizes               <ul style="list-style-type: none"> <li>• 20µl</li> <li>• 250µl</li> <li>• 550µl</li> </ul> </li> <li>• Pipette volume range from 1µl to 550µl</li> <li>• Works with major robotic disposable tips: PerkinElmer, Beckman and others</li> <li>• Pipetting head with fixed, Teflon coated, stainless steel tips</li> </ul>
Consumable Compatibility	<ul style="list-style-type: none"> <li>• Designed to work with industry standard micro-plates and non-proprietary disposable tips</li> <li>• Certified with leading suppliers such as CORNING, Robbins Scientific and more</li> </ul>
Robotic Integration for HTS (multi-station platform)	<ul style="list-style-type: none"> <li>• Control through simple RS232 protocol</li> <li>• Compatible with Peak Robotics KINEDx, AB Controls iX!, CRS Robots, Beckman Sagian, Zymark Twister, and others</li> </ul>
Serial Dilution	<ul style="list-style-type: none"> <li>• Plate to Plate</li> <li>• Column to Column</li> </ul>
High Precision	<ul style="list-style-type: none"> <li>• 5µl dispensing under 2% CV across entire plate (typical)</li> </ul>
Tip Washing Option	<ul style="list-style-type: none"> <li>• Computer controlled Peristaltic pump with circulating wash reservoir</li> </ul>
Field proven technology	<ul style="list-style-type: none"> <li>• Field qualified before mass production</li> <li>• Long term testing through participating user groups for durability and reliability design improvement</li> </ul>
User-friendly, software interface (multi-station platform)	<ul style="list-style-type: none"> <li>• Requires minimal operator training</li> <li>• Allows for User-definable dispense rate and calibration for various liquid types</li> <li>• Records user specific operating procedures and labware used</li> <li>• Runs on various Microsoft Windows environment</li> </ul>
Low maintenance modular design	<ul style="list-style-type: none"> <li>• Speedy identification and replacement of failed components to minimize down time</li> <li>• Low service cost</li> </ul>
Option and Accessories	<ul style="list-style-type: none"> <li>• Comprehensive List of upgrades and accessories is available for your specific application</li> </ul>
Warranty	<ul style="list-style-type: none"> <li>• 12-month extensive manufacturer warranty</li> <li>• Extended 12-month warranty and Annual Service plan are also available. Contact factory or authorized dealers for detail</li> </ul>

# SOLID PHASE EXTRACTION (SPE) APPLICATION

## Automated 96-well SPE and LC-MS/MS using ESI for simultaneous determination of Artemether and its active metabolite Dihydroartemisinin in human plasma

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### Introduction:

Malaria is a leading cause of mortality and morbidity in the developing world, and resistance to available drugs is increasing. A synthetic trioxolane antimalarial drug discovered only 30 years ago Artemether, is an active synthetic derivative of Artemisinin a traditional herb used in China for over 2000 years against fevers. To date, pharmacokinetic studies on plasma levels of Artemether and its active metabolite Dihydroartemisinin have been accomplished with liquid-liquid extraction<sup>1-4</sup> followed by APCI-SIM<sup>5</sup>. We present here a process critical to sensitive and reproducible PK studies made possible with simultaneous detection of both the parent ion and its metabolite.

### Novel Aspect:

PK studies in 96-well format with EDTA-plasma were accomplished for Artemether, and its metabolite Dihydroartemisinin via LC-MS/MS of SPE eluates.

### Abstract:

A rapid and sensitive method for the determination of Artemether and its active metabolite DHA has been developed on automated solid phase extraction (SPE) and high performance liquid chromatography with electrospray tandem mass spectrometry (LC-MS/MS). The SPE method was automated on a 96-well extraction plate (Orochem Celerity Deluxe DVB LP 30mg 1cc) with 96 channel programmable liquid-handling workstation, and coupled off-line with 96-head positive pressure processor. The chromatographic separation was developed using a simple isocratic mixture of 2mM Ammonium acetate Buffer: Acetonitrile (20:80), followed by sample introduction through an ionspray interface in the positive ion mode and tandem mass spectrometry detection with multiple reaction monitoring. Method for SPE was optimized by using different EDTA plasma from different sources. The plasma was subjected to SPE plate to remove protein and other interfering impurities in the plasma to get clean extract for LC-MS/MS analysis. Sample clean up using 96 well polymer DVB LP 30mg 1cc reduces the background noise produced by ESI, enabling the development of a single, and quicker method for the simultaneous extraction of Artemether and its active metabolite Dihydroartemisinin, DHA. Compared to the conventional solid phase extraction using vacuum manifold, the automated high throughput SPE method using positive pressure processor, afforded significant time savings in sample preparation. The assay method was validated and applied successfully to the analysis of Artemether, and DHA in pharmacokinetic and bio-equivalence studies.

### Methods:

A manual SPE method of extraction established using 20-head vacuum manifold was transferred to the Orochem Celerity Deluxe polymer SPE cartridges in 96-well format using the Oroflex Personal Pipettor, an Orochem Technologies Inc. robotic liquid handling system. Pre-conditioning and equilibration, was accomplished using the Oroflex robotic 96 channel Pipettor and off-line processing with the EZYPRESS

an Orochem positive pressure processor. The eluate was collected through positive pressure processing in a 96 -well format and directly injected into an HPLC system using 96 well autosampler. Detection of the parent ion and the metabolite was accomplished by mass spectrometry using multiple reaction monitoring mode. Results are presented for LLOQ, LQC, MQC and HQC (N=6), with 3-day inter-assay measured for precision and accuracy.

**Results:**

Both the drug and metabolite were validated successfully. The resulting assay was sensitive, selective, precise and accurate. Inter assay Precision and accuracy ranged from 4.6 to 9.7 % and 95.7 to 106.4 %

Batch No.	Nominal Concentration (ng/ml)							
	2.5	5	20	40	80	100	150	200
P&A – I	2.6	4.5	20.3	38.5	81.4	90.2	156	202
P&A – II	2.6	4.6	19	41.8	79.5	95.3	167	218
P&A – III	2.5	5.2	18.3	42.6	78.2	87.5	165	207
N	3	3	3	3	3	3	3	3
<b>Mean</b>	2.6	4.8	19.2	41.0	79.7	91.0	162.7	209.0
<b>SD (±)</b>	0.090	0.390	1.015	2.173	2.263	5.515	6.364	8.185
<b>Precision (%CV)</b>	3.5	8.2	5.3	5.3	2.8	6	4	3.9
<b>Nominal (%)</b>	102.5	95.4	96	102.4	99.8	91.4	107	104.5

Table 1: Back Calculated Calibration Standard Statistics for Artemether

Batch No.	Nominal Concentration (ng/ml)							
	2.5	5	20	40	80	100	150	200
P&A – I	2.59	4.6	20.2	39.2	80.9	95.1	153	203
P&A – II	2.62	4.5	20.5	39.5	75.6	98.8	162	205
P&A – III	2.51	4.9	19.9	38.3	82.3	95.9	155	205
N	3	3	3	3	3	3	3	3
<b>Mean</b>	2.57	4.71	20.2	39	79.6	97.4	156.7	204.3
<b>SD (±)</b>	0.057	0.251	0.3	0.624	3.534	2.051	4.726	1.155
<b>Precision (%CV)</b>	2.2	5.3	1.5	1.6	4.4	2.1	3	0.6
<b>Nominal (%)</b>	102.9	94.1	101	97.5	99.5	97.4	104.4	102.2

Table 2: Back Calculated Calibration Standard Statistics for Dihydroartemisinin (DHA)

Nominal Concentration	2.5	7.5	50	120
Average Concentration	2.48	7.6	52.0	127.7
Standard Deviation	0.239	0.557	2.376	8.830
Precision (%)	9.7	7.3	4.6	6.9
Accuracy (%)	99.1	101.2	104.0	106.4
N	18	18	18	18

Batch No.	Nominal Concentration (ng/ml)			
P&A I	2.7	7.0	51.5	131
	2.8	8.3	51.9	129
	2.3	8.6	55.6	120
	2.3	6.6	50.5	137
	2.3	7.8	57.0	130
P&A II	2.6	8.1	53.5	127
	2.3	7.0	54.3	151
	2.2	7.1	51.9	127
	2.3	8.2	50.1	120
	2.1	7.7	53.8	124
P&A III	2.6	7.7	50.5	136
	2.8	7.9	51.1	132
	2.3	7.4	52.1	124
	2.3	7.6	50.3	121
	2.8	7.9	53.4	122
2.7	7.4	52.6	134	
2.7	7.6	47.9	110	

Table 4: Inter-Assay Quality Control Sample for DHA

Nominal Concentration	2.5	7.5	50	120
Average Concentration	2.54	7.4	48.2	114.8
Standard Deviation	0.171	0.623	3.366	8.002
Precision (%)	6.7	8.5	7.0	7.0
Accuracy (%)	101.8	98.3	96.5	95.7
N	18	18	18	18










  

Batch No.	Nominal Concentration (ng/ml)			
P&A I	2.5	7.4	50.8	128
	2.3	7.5	51.9	114
	2.7	8.3	51.7	109
	2.2	6.7	54.0	129
	2.5	6.5	50.5	116
P&A II	2.9	7.2	45.2	110
	2.3	6.8	44.0	109
	2.6	6.9	43.9	112
	2.4	8.4	45.0	104
	2.8	6.6	44.5	106
P&A III	2.5	7.5	46.0	109
	2.6	6.8	47.0	112
	2.4	8.1	51.8	128
	2.6	7.9	47.7	116
	2.6	7.4	50.7	111
2.7	7.2	47.1	108	
2.7	6.9	47.5	117	
2.7	8.1	44.7	121	

Table 3: Inter-Assay Quality Control Sample

## ACCESSORIES

50-A-01N	<p>Pipetting Head 96 Channel - Disposable Tips - 550<math>\mu</math>L</p> <p>For use with 550<math>\mu</math>L PerkinElmer Precise Tips, and Orochem Technologies 550<math>\mu</math>L tips. Compatible with Tomtec Quadra Tips.</p>	
550-A-02TB	<p>Pipetting Head 96 Channel - Stainless Steel Tips w/PTFE Coating.</p> <p>Maximum volume: 550<math>\mu</math>L; Inner diameter of tip: 0.032 inch</p>	
550-A-02TB-SC-S	<p>Single Station System, Single Column Fix Tip Pipetting Head (8 Channels), 550<math>\mu</math>L Per Tip.</p> <p>With Teflon Coating, Inside/Outside. Pipetting tips for serial dilution by column</p> <p>Maximum volume: 550<math>\mu</math>L; Inner diameter of tip: 0.032 inch</p>	
550-A-02TB-SC-M	<p>Multi Station System, Single Column Fix Tip Pipetting Head (8 Channels), 550<math>\mu</math>L Per Tip.</p> <p>With Teflon Coating, Inside/Outside. Pipetting tips for serial dilution by column</p> <p>Maximum volume: 550<math>\mu</math>L; Inner diameter of tip: 0.032 inch</p>	
550-A-02TB-SR	<p>Single Row Fix Tip Pipetting Head (12 Channels), 550<math>\mu</math>L Per Tip.</p> <p>With Teflon Coating, Inside/Outside. Pipetting tips for serial dilution by row</p> <p>Maximum volume: 550<math>\mu</math>L; Inner diameter of tip: 0.032 inch</p>	
550-A-03	<p>Pipetting Head 96 Channel - Disposable Tips: 200 - 250<math>\mu</math>L Tips.</p> <p>For use with BioMek 25 and 250<math>\mu</math>L Tips Only</p>	
550-A-04-55	<p>Pipet Tip Loader (for 550-A-01)</p> <p>Used to manually load 550<math>\mu</math>L PerkinElmer or Orochem Tips onto the 96 channel disposable tip head, 550<math>\mu</math>L</p>	
550-A-04-20	<p>Pipet Tip Loader and Adaptor Set (for 550-A-03)</p> <p>Used to manually load BioMek disposable tips onto the 96 channel disposable tip head, 250<math>\mu</math>L</p>	
550-A-05-55	<p>Pipet Tip Ejector (for 550-A-01)</p> <p>For manually ejecting 550<math>\mu</math>L PerkinElmer tips from the 96 channel disposable tip head, 550<math>\mu</math>L</p>	
550-A-05-20	<p>Pipet Tip Ejector (for 550-A-03)</p> <p>For manually ejecting 25 or 250<math>\mu</math>L BioMek tips from the 96 channel disposable tip head, 250<math>\mu</math>L</p>	
550-A-09	<p>Buffer Reservoir with 3 divisions - Delrin Buffer trough holds 300<math>\mu</math>L of liquid</p>	

550-A-10	Wash Manifold for Stainless Steel Head White Delrin screw down cap that fits over the top of a 96 fixed tip head. When connected by the included tubing to a facet, the head can be cleaned off of the pipetting unit.	
550-A-11	Tip Washing System: Software controlled tip wash system for on deck tip washing of either FT or DT heads on a 4 station Apricot. Includes the following items: -96 individual channel wash station (550-A-13) -2 way peri-pump for circulating wash fluid -2 packs of Tubing and 1 adapter piece RS-232 Communication Cable NOTE: Wash and Waste containers must be supplied by customer.	
550-A-13	Circulating, 96 individual Channel Wash Bath Wash station contains 96 individual cones for washing a 96 tip head.	
550-A-16	Cooling Block - Horizontal Ports Includes reservoir with horizontal connecting ports and tubing. Made of anodized aluminum. Requires customer supplied re-circulating device!	
550-A-16P	Cooling Block - Perpendicular Ports Includes reservoir with horizontal connecting ports and tubing. Made of anodized aluminum. Requires customer supplied re-circulating device!	
550-O-MI	Manual Indexer for Single Station	
550-96R-NS	550 µl, non-sterile disposable tips	
550-96R-F	400 µl, non-sterile, filter barrier disposable tips	
550-96R-S	550 µl, sterile disposable tips	
550-96R-FS	400 µl, sterile, filter barrier disposable tips	

## SPECIFICATIONS

<b>Pipette Volume</b>	1 $\mu$ l to 550 $\mu$ l
<b>Precision</b>	< 2% CV at 250 $\mu$ l (typical, liquid-to-liquid transfer)
<b>Accuracy</b>	+/- 1% at 50 $\mu$ l
<b>Resolution</b>	1 $\mu$ l
<b>Pipette Tips</b>	regular disposable tips; fixed tips
<b>Dimensions</b>	94cm(37.0) width x 29cm(11.4") depth x 60cm(23.5") height
<b>Weight</b>	37.9kg(83.6lb)
<b>Maximum Allowable Labware Height</b>	For disposable tips 12.95cm(5.1") For non-disposable tips: 12.95cm(5.1") For Teflon body pipetting head: 14.22cm(5.6")
<b>Power Requirement</b>	50/60hz, 100v - 240v

### *OroFlex Personal Pipettor Deluxe Includes:*

- 1 ea. 550-A-01, Pipetting Head for Disposable Tips
- 1 ea. 550-A-04-55, Pipet Tip Loader
- 1 ea. 550-A-05-55, Pipet Tip Ejector
- 1 ea. 550-A-08, Pipetting Head Lubrication Box
- 1 ea. 550-A-15, Adaptor for Vacuum Box
- 1 ea. 550-A-09, Buffer Reservoir with 3 divisions - Delrin
- 1 ea. 550-EP-72, RS232C Communication Cable
- 1 ea. 550-EP-59, Software Installation CD
- 1 ea. 550-EP-79, AC Power Cord
- 1 ea. 550-G-01, Instruction Manual



Integratable with vacuum box

### PC System requirements

#### Software

- Windows 98, ME, NT 4.0, 2000, XP

#### Hardware

- Pentium II with 64MB of RAM
- 1 Serial Port (2 Serial Ports required if a pump is used)
- Minimum 40MB of Hard Disk space
- CD-ROM Drive