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# Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay Mini 96-Array Format Manual Workflow SITE PREPARATION GUIDE

for use with:

Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Mini 96-Array Plate
Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Mini 96-Array Plate Core
Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay Mini 96 Reagent Kit

Catalog Numbers 952380, 952424, and 952389 Publication Number MAN0019173

Revision C.0





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Affymetrix Pte Ltd | 7 Gul Circle #2M-01 | Keppel Logistics Building | Singapore 629563 Products:

Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay Mini 96 Reagent Kit

Products:

Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Mini 96-Array Plate
Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Mini 96-Array Plate Core

For descriptions of symbols on product labels or product documents, go to thermofisher.com/symbols-definition.

The information in this guide is subject to change without notice.

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Revision history: Pub. No. MAN0019173

| Revision | Date            | Description   |
|----------|-----------------|---|
| C.0      | 12 October 2020 | <ul> <li>Added Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Mini 96-Array Plate Core product information.</li> <li>Added Bio-Rad HSP9601 as an option to Bio-Rad HSP9631.</li> <li>Added ABgene<sup>™</sup> 96 Well 2.2mL Polypropylene Deepwell Storage Plate as an option to Eppendorf<sup>™</sup> DeepWell<sup>™</sup> Plate 96.</li> </ul> |
| B.0      | 18 August 2020  | Added Axiom <sup>™</sup> GeneTitan <sup>™</sup> 384HT Consumables Kit (Cat. No. 952385).  |
| A.0      | 16 June 2020    | New publication.  |

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# Assay equipment and supplies required

This chapter includes the supplier and ordering information for the equipment, software, reagents, arrays, labware, and other consumables that have been verified for use with the Applied Biosystems<sup>™</sup> Axiom PharmacoFocus Assay Mini 96-Array Format Manual Workflow.

# Applied Biosystems<sup>™</sup> equipment, software, reagents, and arrays required

| 1    | Item  | Source                 |  |
|------|---|------------------------|--|
| Equi | Equipment   |                        |  |
|      | GeneTitan <sup>™</sup> Multi-Channel Instrument <sup>[1]</sup>  | 00-0372 (110V model)   |  |
|      |   | 00-0373 (220V model)   |  |
| Soft | ware  |                        |  |
|      | GeneChip <sup>™</sup> Command Console <sup>™</sup> (GCC)  | version 4.3 or later   |  |
|      | Axiom <sup>™</sup> Analysis Suite   | version 5.1.1 or later |  |
| Rea  | gents   |                        |  |
|      | Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> Assay Mini 96 Reagent Kit, sufficient to process 1 mini 96-array format plate. Consists of: | 952389                 |  |
|      | Module A—mPCR Primers (Part No. 912896)   |                        |  |
|      | Module B—Control DNA (Part No. 912897)  |                        |  |
|      | Module 1 (Part No. 901711)  |                        |  |
|      | Module 2-1 (Part No. 901528)  |                        |  |
|      | Module 2-2 (Part No. 901529)  |                        |  |
|      | Module 3  |                        |  |
|      | <ul> <li>Axiom<sup>™</sup> Wash Buffer A (Part No. 901446)</li> </ul>   |                        |  |
|      | – Axiom™ Wash Buffer B (Part No. 901447)  |                        |  |
|      | <ul> <li>Axiom<sup>™</sup> Water (Part No. 901578)</li> </ul>   |                        |  |
|      | Module 4-1 (Part No. 901278)  |                        |  |
|      | Module 4-2 (Part No. 901276)  |                        |  |

| 1    | Item  | Source |  |
|------|---|--------|--|
| Arra | Arrays  |        |  |
|      | Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> Mini 96-Array Plate   | 952380 |  |
|      | Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> Mini 96-Array Plate Core  | 952424 |  |
| Gen  | eTitan <sup>™</sup> consumables for Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> Array Plates  |        |  |
|      | One of the following kits that contains trays that are required for processing an Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> Array Plate on the GeneTitan <sup>™</sup> Multi-Channel Instrument. |        |  |
|      | Axiom <sup>™</sup> 384HT GeneTitan <sup>™</sup> High Volume Consumables Kit, sufficient to process 5 mini 96-array format plates. Consists of:  | 902629 |  |
|      | Ten 384-Layout GeneTitan™ Stain Tray (Stain 1)     Five 384-Layout Aviers™ Stain 9 Tray   |        |  |
|      | <ul> <li>Five 384-Layout Axiom<sup>™</sup> Stain 2 Tray</li> <li>Five 384-Layout Axiom<sup>™</sup> Stabilization Tray</li> </ul>  |        |  |
|      | Five 384-Layout Axiom™ Ligation Tray  |        |  |
|      | Five 384-Layout GeneTitan <sup>™</sup> Hybridization Tray   |        |  |
|      | Five 384-Layout GeneTitan <sup>™</sup> Scan Tray  |        |  |
|      | <ul> <li>Thirty 384-Layout GeneTitan<sup>™</sup> Scan and Stain Tray Cover</li> </ul>   |        |  |
|      | Axiom <sup>™</sup> GeneTitan <sup>™</sup> 384HT Consumables Kit, sufficient to process 1 mini 96-array format plate. Consists of:   | 952385 |  |
|      | <ul> <li>Two 384-Layout GeneTitan<sup>™</sup> Stain Tray (Stain 1)</li> </ul>   |        |  |
|      | One 384-Layout Axiom <sup>™</sup> Stain 2 Tray  |        |  |
|      | One 384-Layout Axiom <sup>™</sup> Stabilization Tray  |        |  |
|      | One 384-Layout Axiom <sup>™</sup> Ligation Tray   |        |  |
|      | <ul> <li>One 384-Layout GeneTitan<sup>™</sup> Hybridization Tray</li> </ul>   |        |  |
|      | <ul> <li>One 384-Layout GeneTitan<sup>™</sup> Scan Tray</li> </ul>  |        |  |
|      | <ul> <li>Six 384-Layout GeneTitan<sup>™</sup> Scan and Stain Tray Cover</li> </ul>  |        |  |
| Othe | Other Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> kits  |        |  |
|      | Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> Assay Mini 96 Kit   | 952396 |  |
|      | Consists of:  |        |  |
|      | <ul> <li>One Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Mini 96-Array Plate</li> </ul>   |        |  |
|      | <ul> <li>One Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay Mini 96 Reagent Kit</li> </ul>   |        |  |
|      | <ul> <li>One Axiom<sup>™</sup> GeneTitan<sup>™</sup> 384HT Consumables Kit (Cat. No. 952385)</li> </ul>   |        |  |
|      |   |        |  |

| ✓ | Item  | Source |
|---|---|--------|
|   | Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> Assay Mini 96 Training Kit                                | 952398 |
|   | Consists of:  |        |
|   | <ul> <li>Two Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Mini 96-Array Plate</li> </ul>                 |        |
|   | <ul> <li>Two Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay Mini 96 Reagent Kit</li> </ul>           |        |
|   | <ul> <li>Two PharmacoScan<sup>™</sup> DNA Training Plate 96F (Cat. No. 913024)</li> </ul>               |        |
|   | • Two Axiom <sup>™</sup> GeneTitan <sup>™</sup> 384HT Consumables Kit (Cat. No. 952385)                 |        |
|   | Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> Assay Core Mini 96 Kit                                    | 952425 |
|   | Consists of:  |        |
|   | <ul> <li>One Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Mini 96-Array Plate Core</li> </ul>            |        |
|   | <ul> <li>One Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay Mini 96 Reagent Kit</li> </ul>           |        |
|   | • One Axiom <sup>™</sup> GeneTitan <sup>™</sup> 384HT Consumables Kit (Cat. No. 952385)                 |        |
|   | Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> Assay Core Mini 96 Training Kit                           | 952426 |
|   | <ul> <li>Two Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Mini 96-Array Plate Core</li> </ul>            |        |
|   | <ul> <li>Two Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay Mini 96 Reagent Kit</li> </ul>           |        |
|   | <ul> <li>Two PharmacoScan<sup>™</sup> DNA Training Plate 96F (Cat. No. 913024)</li> </ul>               |        |
|   | <ul> <li>Two Axiom<sup>™</sup> GeneTitan<sup>™</sup> 384HT Consumables Kit (Cat. No. 952385)</li> </ul> |        |

<sup>[1]</sup> For a complete list of all equipment and supplies required for GeneTitan Multi-Channel Instrument installation and operation, consult the GeneTitan Multi-Channel Instrument Site Preparation Guide (Pub. No. 08-0305)

## Special requirements

#### Preamplification/amplification staging area

Precautions are required when manipulating genomic DNA to avoid contamination with foreign DNA amplified in other reactions and procedures. It is recommended that genomic DNA manipulations are performed in a dedicated preamplification room or in an area separate from the main laboratory.

This preamplification area must have a dedicated set of pipettes and plasticware. If no dedicated area is available, use of a dedicated bench or a dedicated biosafety hood and dedicated pipettes is suggested. If no dedicated bench or biosafety hood is available, a set of dedicated pipettes is recommended.

#### **Control requirement**

A negative control is not required for this assay.

Two controls are required for proper data analysis. These controls, Control DNA 1 and Control DNA 2, are included in the Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay Mini 96 Reagent Kit.

## **Equipment required**

#### Equipment care and calibration

Lab instrumentation plays an important role in the successful completion of this assay. To aid in maintaining consistency across samples and operators, all equipment must be regularly calibrated and well maintained, including the following:

- · All pipettes, thermal cyclers, and ovens
- Plate spectrophotometer

#### Fume hood

At specific steps in the protocol, we recommend the use of adequate local or general ventilation to keep airborne concentrations low. A fume hood is suggested to achieve the desired concentration. A fume hood is recommended for several steps of this assay.

#### Oven requirements

We recommend using the BINDER<sup> $^{\text{TM}}$ </sup> ED 56 oven that is listed in the following table. If another oven is used, it must be able to maintain a constant temperature of 37°C for at least 24 hours, and have a temperature accuracy of  $\pm 1$ °C.

| ✓ | Item  | Source                        |
|---|---|-------------------------------|
|   | Oven:   |                               |
|   | Required if processing more than 3 plates per week:                           |                               |
|   | <ul> <li>BINDER<sup>™</sup> ED 56 drying and heating chamber</li> </ul>       |                               |
|   | <ul> <li>ED056UL-120V Voltage: 120 V 1~60 Hz</li> </ul>                       | BINDER <sup>™</sup> 9010-0334 |
|   | <ul> <li>ED056-230V Voltage: 230 V 1~50/60 Hz</li> </ul>                      | BINDER <sup>™</sup> 9010-0333 |
|   | Optional—for low throughput of 3 or fewer array plates per week:              |                               |
|   | <ul> <li>GeneChip<sup>™</sup> Hybridization Oven 645<sup>[1]</sup></li> </ul> | 00-0331                       |

<sup>[1]</sup> The GeneChip<sup>™</sup> Hybridization Oven 640 is currently not supported with the Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay. However, to use it in the workflow contact your field service engineer (FSE) or Thermo Fisher Scientific Technical Support regarding the compatibility of this oven with the Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay.

#### Number of ovens required

Multiple ovens are required for manual target preparation. The exact number depends on if you are running a single-sample plate and array plate through the workflow, if you are running the 3-hour precipitation workflow, or if you are running the 3-plate/week manual target preparation workflow. See Table 1 for the different temperatures required for each step.

- If you are running individual plates using standard overnight precipitation workflow, 2 ovens are required for the workflow.
- If you are running individual plates using the 3-hour precipitation workflow, a third oven is highly recommended.
- If you are running the 3-plate/week workflow using either the overnight or 3-hour precipitation workflow, a third oven is highly recommended.

Though only 2 ovens are strictly required, we recommend maintaining separate 37°C ovens for the amplification and fragmentation stages to avoid confusion of plates and to minimize excess opening and closing of oven doors during incubation periods. See Table 2 for a list of suggested settings for 3 ovens when performing the 3-plate/week workflow with overnight precipitation.

Table 1 Oven temperatures needed for each step of the workflow.

| Workflow step                | Oven temperature |
|------------------------------|------------------|
| Amplification                | 37°C             |
| Stopping amplification       | 65°C             |
| Pre-fragmentation incubation | 37°C             |
| Fragmentation incubation     | 37°C             |
| Drying                       | 37°C             |
| Hybridization <sup>[1]</sup> | 48°C             |

<sup>[1]</sup> Required for preheating of the 96-well metal chamber for hybridization transfer.

Table 2 Suggested settings for ovens when performing 3-plate/week manual target preparation workflow using overnight precipitation.

| Day of workflow | Oven 1 | Oven 2 | Oven 3 |
|-----------------|--------|--------|--------|
| Day 1           | 37°C   | N/A    | N/A    |
| Day 2           | 37°C   | 65°C   | 37°C   |
| Day 3           | 48°C   | 65°C   | 37°C   |
| Day 4           | 48°C   | 65°C   | 37°C   |
| Day 5           | N/A    | N/A    | N/A    |

The 3-plate workflow using the optional 3-hour precipitation step requires different oven settings. See the *Axiom™ PharmacoFocus™ Assay Mini 96-Array Format Manual Workflow User Guide* (Pub. No. MAN0019172).

#### Thermal cycler recommendations and protocols

We have verified the performance of this assay using the following thermal cyclers in their 96-well metal chamber configurations. The use of other thermal cyclers may result in assay failure and may violate the array and reagent replacement policy. For more information, see "PCR plate type by thermal cycler for the mPCR step" on page 15.

- Applied Biosystems<sup>™</sup> GeneAmp<sup>™</sup> PCR System 9700 (with gold-plated or silver block)
- Applied Biosystems<sup>™</sup> Veriti<sup>™</sup> Thermal Cycler
- Applied Biosystems<sup>™</sup> ProFlex<sup>™</sup> System
- Eppendorf<sup>™</sup> Mastercycler<sup>™</sup> pro S

**Note:** Two verified thermal cyclers are required if running the 3-plate/week manual target preparation workflow.

**IMPORTANT!** Always use the heated lid option when programming protocols. See the appropriate thermal cycler user guide for programming information.

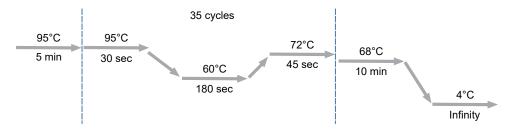


Figure 1 PharmacoFocus mPCR thermal cycler protocol (Stage 1A).

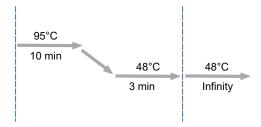


Figure 2 PharmacoFocus Denature thermal cycler protocol (Stage 4).



**WARNING!** Evaporation during denaturation can negatively affect assay performance. Use the recommended thermal cycler consumables and sealing film to eliminate condensation and evaporation.

#### Plate centrifuge

One plate centrifuge is required for the Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay Mini 96-Array Format Manual Workflow. We recommend the plate centrifuges listed in "Plate centrifuge recommendations" on page 10.

When centrifuging and drying pellets, the centrifuge must meet the following requirements:

- Temperature: 4°C.
- Rcf: 3,200 × g with an appropriate rotor-bucket combination, or 4,000 rpm for the Eppendorf<sup>™</sup>
   Centrifuge 5810 R configuration described below.

Relative centrifugal force (rcf) can be calculated as follows:

 $rcf = (1.118 \times 10-5) R S2$  where R is the radius of the rotor in centimeters and S is the speed of the centrifuge in revolutions per minute.

In addition, the bottom of the rotor buckets should be soft rubber to ensure that the deep-well plates do not crack. Do not use buckets that sit plates directly on a metal or hard plastic bottom. For the Eppendorf<sup>™</sup> Centrifuge 5810 R, do not use the A-4-62 rotor with a WO-15 hard bottom plate carrier.

#### Plate centrifuge recommendations

The following plate centrifuges are recommended for the Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay Mini 96-Array Format Manual Workflow.

| 1 | Item  | Supplier                       | Source   |
|---|---|--------------------------------|--|
|   | Sorvall <sup>™</sup> Legend <sup>™</sup> XTR Centrifuge | Thermo Fisher                  | 75004539 (bench model)                                     |
|   | (refrigerated), with:                                   | Scientific                     | 75004520 (230 V, 50 Hz, floor model)                       |
|   |   |                                | 75004521 (120 V, 60 Hz, floor model)                       |
|   |   |                                | 75004523 (230 V, 50-60 Hz,<br>USA and Canada, floor model) |
|   | ☐ TX-750 4 + 750mL Swinging Bucket Rotor                |                                | 75003180 (rotor)   |
|   | ☐ Microplate/Flask Carriers                             |                                | 75003795 (set of 2 carriers)                               |
|   |   |                                | 75003617 (set of 4 carriers)                               |
|   | Eppendorf <sup>™</sup> Centrifuge 5810 R, with:         | Fisher Scientific <sup>™</sup> | 022625551 (230 V, 50–60 Hz)                                |
|   | ☐ Rotor A-4-81, with 4 MTP/Flex buckets                 |                                | 022625501 (120 V, 50-60 Hz, 15 A)                          |
|   |   |                                | 022625101 (120 V, 50–60 Hz, 20 A)                          |
|   |   |                                | 022638807 (rotor)  |
|   | Allegra <sup>™</sup> 25R Benchtop Centrifuge,           | Beckman Coulter <sup>™</sup>   | 369434 (230 V, 50-60 Hz)                                   |
|   | Refrigerated, with S5700 Swinging-Bucket Rotor          |                                | 369435 (200 V, 50–60 Hz)                                   |
|   | . Total   |                                | 369436 (230 V, 50 Hz)                                      |
|   |   |                                | 368954 (rotor)   |

#### Plate shakers

We recommend using one of the following shakers.

| Item   | Source  |
|--|---|
| Thermo Scientific <sup>™</sup> Compact Digital Microplate Shaker | Fisher Scientific <sup>™</sup> 88880023 or 88880024             |
| Jitterbug <sup>™</sup>   | Boekel Scientific <sup>™</sup> 130000 (115V)<br>130000-2 (230V) |

### Spectrophotometer

We recommend that you use one of the following spectrophotometers, or the equivalent.

| Item  | Source   |
|---|--|
| Thermo Scientific <sup>™</sup> Multiskan <sup>™</sup> Sky Microplate<br>Spectrophotometer | 51119600   |
| SpectraMax® Plus 384 Microplate Reader  | Molecular Devices <sup>®</sup> PLUS 384                                |
| DTX 880 Multimode Detector with genomic filter slide                                      | Beckman Coulter <sup>™</sup><br>Detector 987921<br>Filter slide A30184 |

#### Other equipment and supplies required

The following additional materials are required for the Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay Mini 96-Array Format Manual Workflow.

Unless otherwise indicated, all materials are available through **thermofisher.com**. "MLS" indicates that the material is available from **fisherscientific.com** or another major laboratory supplier.

| 1   | Item   | Source   |  |  |
|-----|--|--|--|--|
| Com | Common laboratory equipment  |  |  |  |
|     | Freezer, -20°C   | MLS  |  |  |
|     | Refrigerator, 2–8°C  | MLS  |  |  |
|     | Ice bucket, 4–9 L  | MLS  |  |  |
|     | Fine-point permanent markers   | MLS  |  |  |
|     | Vortex-Genie 2   | Fisher Scientific <sup>™</sup>                         |  |  |
|     |  | 50-728-002 (120 V, 60 Hz)<br>50-728-004 (230 V, 50 Hz) |  |  |
|     | Mini Microcentrifuge for 2-mL tubes                                    | MLS  |  |  |
|     | Bel-Art <sup>™</sup> Cryo-Safe <sup>™</sup> Mini Cooler <sup>[1]</sup> | Fisher Scientific <sup>™</sup> , 03-410-497            |  |  |
|     | Pipet-Aid <sup>™</sup> Pipette Controller <sup>[1]</sup>               | VWR <sup>™</sup> , 53498-001                           |  |  |

<sup>[1]</sup> Equivalent products from other manufacturers are acceptable.

# Labware and accessories required

#### Labware and consumables required

Unless otherwise indicated, all materials are available through **thermofisher.com**. "MLS" indicates that the material is available from **fisherscientific.com** or another major laboratory supplier.

| Labware  | Source   | Image |
|--|--|-------|
| 96-deepwell plate  • ABgene <sup>™</sup> 96 Well 2.2mL Polypropylene Deepwell Storage Plate, or  • Eppendorf <sup>™</sup> DeepWell Plate 96, 2,000 μL  | <ul> <li>Fisher Scientific AB0932</li> <li>Fisher Scientific 13-864-302</li> </ul> |       |
| Greiner Bio-One <sup>™</sup> UV-Star <sup>™</sup><br>96-Well UV Spectroscopy<br>Microplate   | Fisher Scientific <sup>™</sup> 07-000-407  |       |
| Hard-Shell <sup>™</sup> 96-Well PCR Plate, high profile, semi skirted  Note: See "PCR plate type by thermal cycler for the mPCR step" on page 15 for the PCR plate type recommended for the specific thermal cycler you are using. | Bio-Rad <sup>™</sup> HSS9641   |       |
| Hard-Shell <sup>™</sup> 96-Well PCR Plate, low profile, full skirted  Note: See "PCR plate type by thermal cycler for the mPCR step" on page 15 for the PCR plate type recommended for the specific thermal cycler you are using.  | Bio-Rad <sup>™</sup> HSP9631, or<br>Bio-Rad <sup>™</sup> HSP9601                   |       |

| Labware   | Source   | Image  |
|---|--|--|
| 1.7-mL microcentrifuge tubes,<br>DNAse and RNAse-free   | MLS  |  |
| 15-mL and 50-mL conical-bottom centrifuge tubes, polypropylene  | MLS  |  |
| Matrix <sup>™</sup> Reagent Reservoir, 25 mL  | 8093 (10 bags of 10),<br>8093-11 (pack of 100 in one bag)  |  |
| Matrix <sup>™</sup> Reagent Reservoir, 100 mL   | 8085   |  |
| 96-well metal chamber Warming or cooling chamber for 0.2 mL tubes, 96 holes (4 for 1.5 mL and 6 for 0.5 mL tubes), Dimensions: 6 1/8"L x 3 1/8"W x 1" H | Diversified Biotech <sup>™</sup> CHAM-1000   | PARTIE AND LANGE OF THE PARTIES OF T |
| Adhesive film   | Use one of the following:  • MicroAmp <sup>™</sup> Clear Adhesive Film 4306311  • Microseal <sup>™</sup> 'B' PCR Plate Sealing Film Bio-Rad <sup>™</sup> , MSB1001 | MicroAmp V Constanting Constan |

#### PCR plate type by thermal cycler for the mPCR step

The following table provides details about the consumables to be used with each thermal cycler when executing the mPCR step.

| Thermal cycler model  | PCR plate type  | Seal <sup>[1]</sup>   |
|---|---|---|
| Applied Biosystems <sup>™</sup> GeneAmp <sup>™</sup> PCR<br>System 9700 | Bio-Rad <sup>™</sup> Hard-Shell <sup>™</sup> 96-Well PCR<br>Plate, high profile, semi skirted<br>(Cat. No. HSS9641)           | MicroAmp <sup>™</sup> Clear Adhesive<br>Film (Cat. No. 4306311) |
| Applied Biosystems <sup>™</sup> Veriti <sup>™</sup> Thermal<br>Cycler   | Bio-Rad <sup>™</sup> Hard-Shell <sup>™</sup> 96-Well PCR<br>Plate, high profile, semi skirted<br>(Cat. No. HSS9641)           | MicroAmp <sup>™</sup> Clear Adhesive<br>Film (Cat. No. 4306311) |
| Applied Biosystems <sup>™</sup> ProFlex <sup>™</sup> PCR<br>System      | Bio-Rad <sup>™</sup> Hard-Shell <sup>™</sup> 96-Well PCR<br>Plate, high profile, semi skirted<br>(Cat. No. HSS9641)           | MicroAmp <sup>™</sup> Clear Adhesive<br>Film (Cat. No. 4306311) |
| Eppendorf <sup>™</sup> Mastercycler <sup>™</sup> pro S                  | Bio-Rad <sup>™</sup> Hard-Shell <sup>™</sup> 96-Well PCR<br>Plate, low profile, full skirted (Cat. No.<br>HSP9631 or HSP9601) | MicroAmp <sup>™</sup> Clear Adhesive<br>Film (Cat. No. 4306311) |

<sup>[1]</sup> Microseal<sup>™</sup> 'B' PCR Plate Sealing Film from Bio-Rad<sup>™</sup> (Cat. No. MSB1001) can be used instead of MicroAmp<sup>™</sup> Clear Adhesive Film for the Applied Biosystems<sup>™</sup> thermal cyclers.

#### Plate requirements for manual target preparation

The following types of plates are required for performing manual target preparation.

- 96-deepwell plate: ABgene<sup>™</sup> 96 Well 2.2mL Polypropylene Deepwell Storage Plate or Eppendorf<sup>™</sup> DeepWell<sup>™</sup> Plate 96, 2 mL
- Bio-Rad<sup>™</sup> Hard-Shell<sup>™</sup> 96-Well PCR Plate, high profile, semi skirted (Cat. No. HSS9641) for the following thermal cyclers:
  - Applied Biosystems<sup>™</sup> GeneAmp<sup>™</sup> PCR System 9700 (with gold-plated or silver block)
  - Applied Biosystems<sup>™</sup> Veriti<sup>™</sup> Thermal Cycler
  - Applied Biosystems<sup>™</sup> ProFlex<sup>™</sup> PCR System
- Bio-Rad<sup>™</sup> Hard-Shell<sup>™</sup> 96-Well PCR Plate, low profile, full skirted (Cat. No. HSP9631 or HSP9601) for the following thermal cycler:
  - Eppendorf<sup>™</sup> Mastercycler<sup>™</sup> pro S
- Greiner Bio-One<sup>™</sup> UV-Star<sup>™</sup> 96-Well UV Spectroscopy Microplate, 370 μL/well (Cat. No. 07-000-407)

# Pipettes and tips recommendations

| 1   | Item   | Source                         |
|-----|--|--------------------------------|
|     | Pipet-Lite <sup>™</sup> Magnetic Assist Pipet, single-channel P20 <sup>[1]</sup>   | Rainin <sup>™</sup> , L-20     |
|     | Pipet-Lite <sup>™</sup> Magnetic Assist Pipet, single-channel P200 <sup>[1]</sup>  | Rainin <sup>™</sup> , L-200    |
|     | Pipet-Lite <sup>™</sup> Magnetic Assist Pipet, single-channel P1000 <sup>[1]</sup> | Rainin <sup>™</sup> , L-1000   |
|     | Pipette 12-channel P20 <sup>[1]</sup>  | Rainin <sup>™</sup> , L12-20   |
|     | Pipette 12-channel P50 (optional) <sup>[1]</sup>                                   | Rainin <sup>™</sup> , L12-50   |
|     | Pipette 12-channel P200 <sup>[1]</sup>   | Rainin <sup>™</sup> , L12-200  |
|     | Pipette 12-channel P1200 <sup>[1]</sup>  | Rainin <sup>™</sup> , L12-1200 |
|     | Pipette 8-channel P20 <sup>[1]</sup>   | Rainin <sup>™</sup> , L8-20    |
|     | Pipette 8-channel P200 <sup>[1]</sup>  | Rainin <sup>™</sup> , L8-200   |
|     | Pipette tips Green-Pak <sup>™</sup> 10-μL refill                                   | Rainin <sup>™</sup> , 30389274 |
|     | Pipette tips Green-Pak <sup>™</sup> 200-μL refill                                  | Rainin <sup>™</sup> , 30389276 |
|     | Pipette tips Green-Pak <sup>™</sup> 1,000-μL refill                                | Rainin <sup>™</sup> , 30389272 |
|     | Pipette tips RT 10 μL (racked tips)  | Rainin <sup>™</sup> , 30389225 |
|     | Pipette tips RT 200 μL (racked tips)   | Rainin <sup>™</sup> , 30389239 |
|     | Pipette tips RT 1,000 μL (racked tips)   | Rainin <sup>™</sup> , 30389212 |
| [4] |  |                                |

<sup>[1]</sup> Equivalent item is acceptable.

### Other labware and reagents required

The following additional labware and reagents are required for the Axiom<sup>™</sup> PharmacoFocus<sup>™</sup> Assay Mini 96-Array Format Manual Workflow.

Unless otherwise indicated, all materials are available through **thermofisher.com**. "MLS" indicates that the material is available from **fisherscientific.com** or another major laboratory supplier.

| 1    | Item   | Source                                      |  |
|------|--|---|--|
| Laby | Labware  |   |  |
|      | Microcentrifuge tube holder                                      | MLS   |  |
|      | 50-mL tube holder  | MLS   |  |
|      | 15-mL tube holder  | MLS   |  |
|      | VWR <sup>™</sup> Disposable 10-mL Serological Pipet              | VWR <sup>™</sup> , 89130-898                |  |
|      | VWR <sup>™</sup> Disposable 5-mL Serological Pipet               | VWR <sup>™</sup> , 89130-896                |  |
|      | Laboratory tissue  | Fisher Scientific <sup>™</sup> , 06-666A    |  |
| Reag | Reagents   |   |  |
|      | QIAGEN <sup>™</sup> Multiplex PCR <i>Plus</i> Kit (100)          | 206152                                      |  |
|      | Quant-iT <sup>™</sup> PicoGreen <sup>™</sup> dsDNA Assay Kit     | P7589                                       |  |
|      | Reduced EDTA TE Buffer (10 mM Tris-HCl pH 8.0, 0.1 mM EDTA)      | Fisher Scientific <sup>™</sup> , AAJ75793AE |  |
|      | 2-Propanol, anhydrous, 99.5% (Isopropanol)                       | Sigma-Aldrich <sup>™</sup> , 278475         |  |
| Reag | gents, gels, and instruments required to run quality-control st  | eps   |  |
|      | Mother E-Base <sup>™</sup> Device                                | EBM03                                       |  |
|      | Daughter E-Base <sup>™</sup> Device                              | EBD03                                       |  |
|      | E-Gel <sup>™</sup> 48 Agarose Gels, 4% (for Sample QC)           | G800804                                     |  |
|      | E-Gel <sup>™</sup> 48 Agarose Gels, 2% (for optional mPCR QC)    | G800802                                     |  |
|      | E-Gel <sup>™</sup> 48 Agarose Gel, 1%                            | G800801                                     |  |
|      | E-Gel <sup>™</sup> 96 High Range DNA Marker (for gDNA QC)        | 12352019                                    |  |
|      | TrackIt <sup>™</sup> Cyan/Orange Loading Buffer                  | 10482028                                    |  |
|      | Applied Biosystems <sup>™</sup> 25 bp DNA Ladder (for sample QC) | 931343                                      |  |
|      | RediLoad <sup>™</sup> Loading Buffer (for gDNA QC)               | 750026                                      |  |
|      | Water, Nuclease-free, Molecular Biology Grade, Ultrapure         | MLS   |  |
|      | 50 bp DNA Ladder (for optional mPCR QC)                          | New England BioLabs Inc., N3236S            |  |

# Documentation and support

# **Related documentation**

| Document  | Publication number | Description  |
|---|--------------------|--|
| Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> Assay Mini<br>96-Array Format Manual Workflow User<br>Guide       | MAN0019172         | This document provides instruction on running the Axiom™ PharmacoFocus™ Assay on mini 96-format plates and array processing on the GeneTitan™ Multi-Channel Instrument.  |
| Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> Assay Mini 96-<br>Array Format Manual Workflow Quick<br>Reference | MAN0019174         | An abbreviated reference for the target preparation step of the Axiom <sup>™</sup> PharmacoFocus <sup>™</sup> Assay Mini 96-Array Format Manual Workflow. This document is for experienced users.  |
| GeneTitan <sup>™</sup> Multi-Channel Instrument<br>User Guide   | 08-0308            | The GeneTitan <sup>™</sup> Multi-Channel (MC) Instrument automates array processing from target hybridization to data generation by combining a hybridization oven, fluidics processing, and state-of-the-art imaging device into a single benchtop instrument. This document detailing the use, care, and maintenance for the GeneTitan <sup>™</sup> MC.  |
| GeneTitan <sup>™</sup> Multi-Channel Instrument<br>Site Preparation Guide                                       | 08-0305            | Provides guidance on creating and maintaining the proper environment required for the GeneTitan <sup>™</sup> MC Instrument.  |
| Software and analysis   |                    |  |
| GeneChip <sup>™</sup> Command Console <sup>™</sup> User<br>Guide  | 702569             | This user guide provides instructions on using Applied Biosystems <sup>™</sup> GeneChip <sup>™</sup> Command Console <sup>™</sup> software (GCC) used to control GeneChip <sup>™</sup> instrument systems. GeneChip <sup>™</sup> Command Console <sup>™</sup> software provides an intuitive set of tools for instrument control and data management used in the processing of GeneChip <sup>™</sup> arrays. |

| Document   | Publication number | Description   |
|--|--------------------|---|
| Axiom <sup>™</sup> Analysis Suite User Guide                       | 703307             | Axiom <sup>™</sup> Analysis Suite advances genotyping data analysis with a single-source software package to enable complete genotyping analysis of all Axiom <sup>™</sup> arrays. This document provides instructions on using the software to automate the Best Practices Workflow to deliver accurate results in a single step for export in PLINK, VCF, or TXT formats.               |
| Axiom <sup>™</sup> Genotyping Solution Data<br>Analysis User Guide | MAN0018363         | This guide provides information and instructions for analyzing Axiom <sup>™</sup> genotyping array data. It includes the use of Axiom <sup>™</sup> Analysis Suite, Applied Biosystems <sup>™</sup> Array Power Tools and SNPolisher R package to perform quality control analysis (QC) for samples and plates, SNP filtering before downstream analysis, and advanced genotyping methods. |

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**Note:** For SDSs for reagents and chemicals from other manufacturers, contact the manufacturer.

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