Using Mai Tai® Cassettes

Equipment Needed

- **Mai Tai® Hybridization System**, 115v/230v. — *preheated* (Includes a Model 700 or 777 Microarray Oven, a Mai Tai Rotator and 1 or more Mai Tai Cassettes, per configuration.
- **Microarrays compatible with Agilent gasket slides on 1x3 inch (25x75mm) substrates**.
- **Gasket slides matching the microarray format used:**
  - **1 x 244K microarrays** (Agilent cat. #G2534-60003 or similar)
  - **2 x 105K microarrays** (Agilent cat. #G2534-60002 or similar)
  - **4 x 44K microarrays** (Agilent cat. #G2534-60011 or similar)

Microarray Oven Set Up

Load the Mai Tai Rotator into the Microarray Oven by inserting the ball joint end of the shaft into right-side bearing of the oven chamber. Then place the other end into the left bearing. Reverse this procedure when removing the rotator. With Mai Tai Rotator installed, pre-heat the oven to the desired incubation temperature, as recommended by the microarray supplier.

Mai Tai® Cassette Components

The Mai Tai Cassette design uses just two parts, a cover and baseplate, to simplify set up and disassembly and reduce handling errors (Fig. 1).

The cassette has a standard microplate footprint so samples may be loaded manually or with robotic liquid handling equipment.

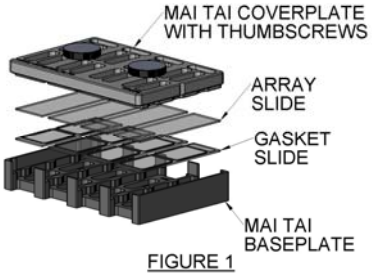

FIGURE 1

Mai Tai® Cassette Set Up

1. Place the cassette base plate on a flat surface.
2. Add **NEW** gasket slides by placing the "Agilent" label face up to align with the "barcode" (Fig. 2).

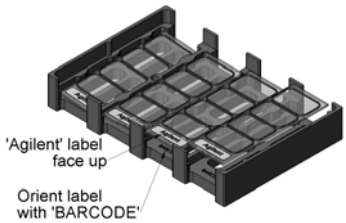

FIGURE 2

⚠ *Sample loss has been observed with **used** slides.*

3. Check to ensure each gasket slide lays down flat, so that it **does not** rest on the edge of the base plate (Fig. 3). Practice placing standard glass slides into the cassette prior to running an experiment.

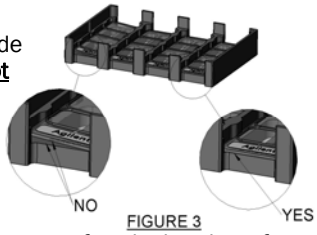

FIGURE 3

4. Pipette labeled sample into the center of each chamber of the gasket slide using the volumes shown in Table 1.

⚠ ***Load samples onto all gasket slides BEFORE placing arrays***

Table 1. Sample Volumes

Gasket Format	Probe Volume
1x microarray	490 µl
2x microarrays	245 µl
4x microarrays	100 µl

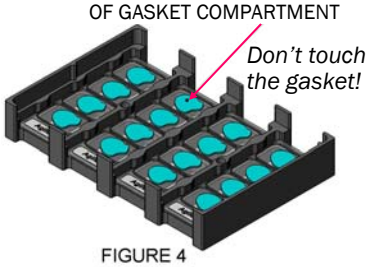

FIGURE 4

⚠ *To prevent spilling or leaking, dispense hybridization solution to the center of the gasket well without touching the gasket with the pipette tip. Some sample will "wick" over the barrier if it touches the gasket during assembly.*

Continued on next page.

Mai Tai® Cassette Set Up (continued)

5. Hold a microarray by the ends with the array side down so that the barcodes will meet. While taking care to keep it flat and level, slowly lower the array onto the gasket slide until it makes good contact over the length of the slide (Fig. 5).

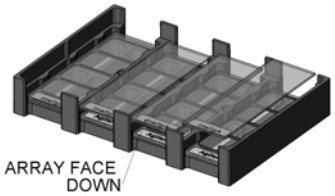

FIGURE 5

⚠ *Using microarrays compatible with Agilent gasket slides and placing them evenly onto gasket slides is crucial to prevent spilling or leakage.*

6. Repeat step 5 until all arrays have been placed, ensuring all assemblies lay flat against the base plate. See Fig. 6 for examples of proper and improper placement.
7. Place the cover plate over the arrays with the barcode window over the array barcodes (Fig. 7).

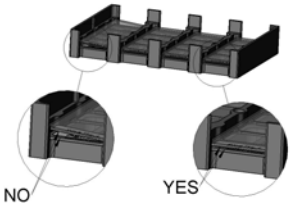

FIGURE 6

8. Tighten the thumbscrews by hand to a complete stop. ***Don't*** *use tools.*
9. Rotate the cassette by hand to ensure that all bubbles move freely inside each array chamber. If any stationary bubbles remain, gently tap each corner of the assembly on a firm surface.

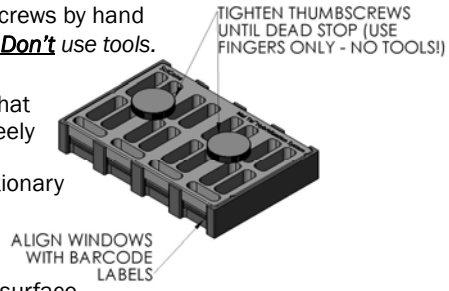

FIGURE 7

⚠ *A large single bubble which moves freely around the chamber is optimal for hybridization.*

10. Place the cassette in the pre-heated oven by raising the spring bar on the Mai Tai rotator and sliding it into an open slot (Fig. 8). When loading pairs of cassettes, place them on opposing sides of the rotator for balance.

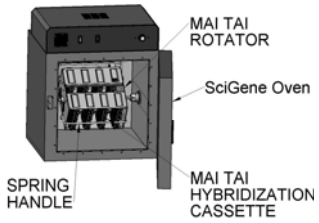

FIGURE 8

11. Adjust the oven speed control as needed and process for the time recommended by the microarray supplier.
12. At the completion of the incubation period, remove the cassette(s) from the rotator and allow to cool for a few minutes. Loosen thumbscrews and remove the top plate.
13. Remove all slide assemblies from the cassette and place them in wash buffer specified by the microarray supplier.

⚠ *Place all assemblies into wash buffer* ***before*** *separating arrays from gasket slides.*

14. With all assemblies submerged in wash buffer, separate each array from its gasket slide and place in a slide rack for processing.
15. Wash and dry arrays on the **Little Dipper® Processor** [SciGene Cat. #1080-40-X] or manually using buffer(s) specified by the supplier.

Cleaning and Storing Mai Tai Cassettes

Hybridization solution may occasionally spill, making arrays or gasket slides stick to cassette components during hybridization. Cassettes should be cleaned periodically (according to your lab schedule) to prevent this. Follow the cleaning method below:

1. Disassemble each Mai Tai cassette into its components: base plate and cover plate. Remove captive thumbscrews by unscrewing them from the cover plate.
2. Using a lint-free wipe soaked in 70% ethanol, wipe all surfaces of base plate, cover plate, thumbscrews and thumbscrew hole carefully to remove excess salts and dried hybridization solution.
3. Allow cleaned parts to dry completely, by evaporation.
4. Re-assemble the cassette, place in a clean plastic bag or in cling wrap and store in a dry place.

— End Protocol —

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