Ncardia
Stem cell experts

This document describes handling of the Ncyte™ CardioPlate™ Maestro™ MEA 96 upon, arrival, maintainance procedures, activity measurements on the Axion Maestro MEA platform, and recommended beat detection settings for the AxIS software.

Follow the procedures described in this document to ensure optimal performance of the product. Typical compound responses using the Ncyte CardioPlate Maestro MEA 96 can be obtained through your local Ncardia representative or support@ncardia.com. It is recommended to begin compound assays after a three-day equilibration period following product delivery. An analysis template can be found at ncardia.com/ncytecp.

Technical support

email **support@ncardia.com**
phone **+31 (71) 332 2230**
web **ncardia.com**

Note for package receiver

This product contains live cells which need immediate action. Do not open the package. Please deliver the product immediately to the end user.

Equipment required	Materials required
Axion Maestro MEA™ system + AxIS software	Ncyte™ CardioPlate™ Maestro™ MEA 96
Class 2 laminar flow cabinet	50 mL conical tubes, sterile
Incubator at 37˚C, with 5% CO_2, saturated humidity	200 µL filter pipette tips, sterile
Vacuum-powered suction system (optional)	Reagent reservoirs, sterile
8 or 12-channel pipette	

Ref: QG_Nc_Ncyte_MEA_1

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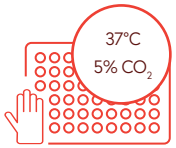
Quick Guide

Ncyte™ CardioPlate™ Maestro™ MEA 96

Information for user

1 Upon delivery, place the Ncyte™ CardioPlate™ while wearing protection in an incubator (37˚C, 5% CO_2).



2 Store the Cardiomyocyte Culture Medium at 4°C upon arrival.
Optional: Add antiibiotic of choice (not provided),

Store at 4°C upon arrival

3 On the day of arrival, remove the sealing mat and cover the plate with a new, sterile lid. Perform an activity test at 300 µV, as described below.

Measure activity

4 Exchange the medium in the Ncyte CardioPlate on the **day of arrival**. using the description provided in this Quick Guide.

Replace the medium on day of arrival

Arrival Day 0	Day 3-13
Medium refreshment	- Medium replacement every other day - Compound assay

It is recommended to use the Ncyte CardioPlate within 14 days of arrival. Contact Ncardia if the product does not arrive on the expected arrival day.

Method: MEA Activity Test

***Note:** Do NOT change the medium before finishing the heat map analysis.*

1. Start the Axion MEA AxIS software.
2. Set the temperature to 37°C (environment control -> heater control)
3. Apply the cardiac realtime spontaneous beat detector mode.
   (configuration -> cardiac -> real-time → spontaneous)

4. Set the heat map at the manual scale limit to 300 µV. (activity map -> spike amplitude)
5. Equilibrate the Ncyte™ CardioPlate™ in the device for 10 minutes before recording.
6. Start a two-minute MEA recording. Save it in a dedicated folder (experiment setup properties).
7. Place the Ncyte CardioPlate back into the incubator at 37°C, 5% CO_2.

Method: Medium Replacement

Note: *Ncyte CardioPlate, including lids and Cardiomyocyte Culture Media, should be handled under sterile conditions. To avoid contamination, wear gloves, use ethanol to disinfect all equipment and materials, and work in a laminar flow cabinet. Optional: Antibiotic of choice may be added to the medium (not provided).*

Note: *Volumes used are calculated for one Ncyte CardioPlate. When refreshing more than one Ncyte CardioPlate, multiply the volume of Cardiomyocyte Culture Medium required by the number of Ncyte CardioPlate products to be used. It is recommended to exchange one Ncyte CardioPlate at a time.*

1. Transfer 21 mL Cardiomyocyte Culture Medium from the bottle to a sterile 50 mL conical tube. Incubate the tube at 37°C in a water bath for at least 30 minutes.
2. Transfer the Ncyte™ CardioPlate™ from the incubator to the flow cabinet.
3. Remove the lid and discard it. Use the included sterile lid for the remainder of the procedure.
4. While holding the Ncyte™ CardioPlate™ firmly on a flat surface, gently remove the sealing mat from the plate.
5. Transfer the pre-warmed Cardiomyocyte Culture Medium from the 50 mL conical tube to a multi-channel reservoir in the flow cabinet.
6. Carefully aspirate the fluid from the wells using a vacuum-powered suction system. Alternatively, a multichannel pipette can be used.
   Important: Avoid touching the bottom of the CardioPlate with the pipette tips.
   Important: Prevent cells from drying out. It is recommedned to replace medium per 3 columns at a time.

7. Add 200 µL of pre-warmed Cardiomyocyte Culture Medium to each well using a multichannel pipette.
8. Place the included sterile lid on the Ncyte CardioPlate™.
9. Transfer the Ncyte™ CardioPlate™ back to the incubator (37°C, 5% CO_2).
10. Refresh medium every other day.

AxIS Beat Detection Settings

It is recommended to perform analysis of baseline and compound effetcs with the beat detection setting shown in Figure 1.

For optimal results, analyze signals in a stable region of 30 beats per segmentation during at least 3 minutes recordings to capture sufficient beats.

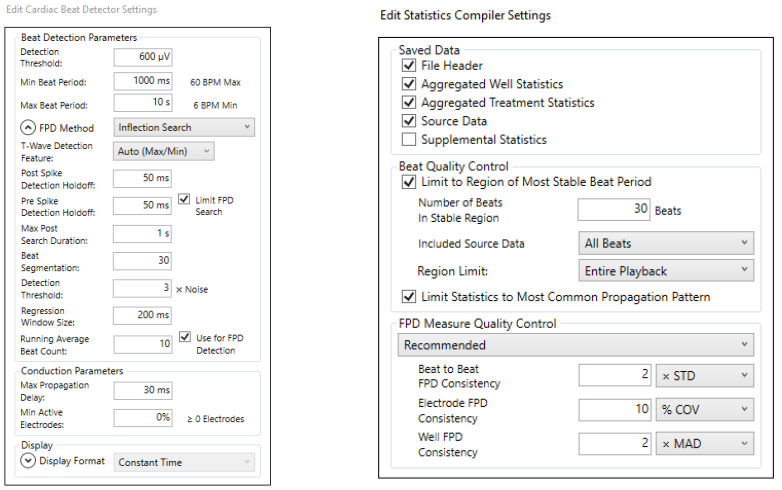


Figure 1. Recommended AxIS Cardiac Beat Detection settings (left) and Statistics Compiler settings (right) for the Ncyte CardioPlate Maestro MEA.