Aurora Disposable Cartridge Handling Manual

For Aurora cartridge 210-0001-CA-D

Version 1.00

106-0010-BA-D

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Introduction

This manual describes the proper method of preparing a disposable cartridge for use in the Aurora, loading the sample in the cartridge and extracting the purified DNA from the cartridge once the Aurora purification is complete. Details on preparing a variety of sample types prior to loading them into the Aurora cartridge can be found in separate Aurora protocol documents on the Boreal Genomics website.

Cartridge compatibility

The procedures described in this manual apply to Aurora cartridge 210-0001-CA-D.

Safety guidelines

Cartridges are made from non-hazardous plastics, metal, TBE buffer and agarose. Appropriate precautions should be taken if hazardous samples are used with the cartridges. Wear gloves during all stages of the protocol. Avoid skin contact with all reagents. Refer to safety guidelines in Boreal Genomics Protocol documents for further information.

Procedure overview

- **Prepare the sample**
- **Prepare the cartridge**
- **Load the sample**
- **Prepare the Aurora**
- **Begin the run**
- **Sample extraction**

Preparing the sample

Please see your Aurora protocol documentation for details on the method of preparing your sample prior to loading it into the Aurora cartridge.

Preparing the cartridge

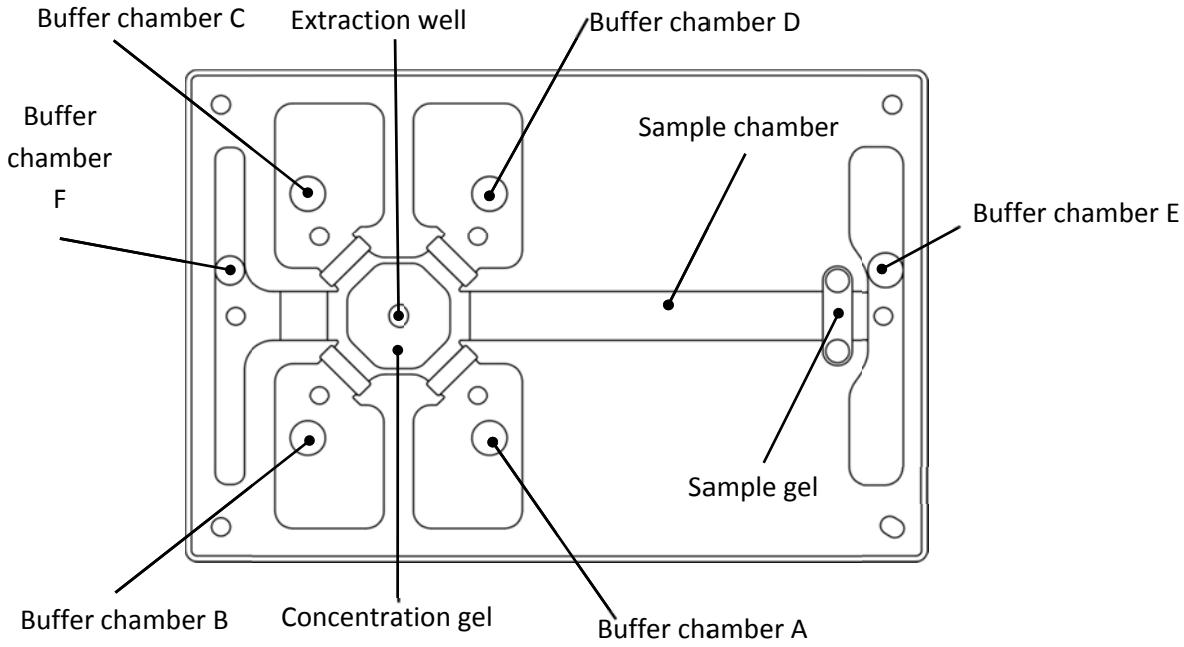

Figure 1. Top view of the cartridge.

Opening the cartridge

To remove a cartridge, open the packaging by cutting the end of the bag open using scissors.

Inspecting the cartridge

Upon removal, each cartridge should be inspected for damage or defects. Examine:

- The graphite electrodes for chips or cracks,
- The cartridge plastic for cracks,
- The concentration and sample gels for presence of bubbles,
- The cartridge for signs of leakage (wetness of the cartridge or its packaging), and
- The buffer chambers and the sample chamber for presence of buffer; each should be nearly full of liquid along with some air bubbles.

If you observe any damage or defects, please contact support@borealgenomics.com.

Preparing the cartridge: Overview

1. Clean your work surface, cut open the cartridge shipping pouch, and remove the cartridge.
2. Peel off and discard the film covering the top surface of the cartridge.
3. Remove 2 ml of buffer from each of the buffer chambers and remove the entire volume of buffer from the sample chamber.
4. Remove the tape covering the extraction well. Remove the buffer from the extraction well. Pipette 60 µl of buffer from any of the buffer wells into the extraction well. Seal the well with tape.
5. Transfer up to 5 ml of sample to the sample chamber. If the sample is smaller than 5 ml, bring volume to 5 ml with deionized water.

Removing the sealing tape

In order to gain access to the sample and buffer chambers, the clear adhesive sealing tape must be removed from the top of the cartridge. To remove the sealing tape from the cartridge:

- Place the cartridge on a sturdy, flat surface.
- Hold the cartridge down firmly.
- Peel off the sealing tape, starting from the exposed flap at one corner of the cartridge.

Figure 2. Removing the sealing film from the cartridge.

Removing packing buffer

Before using the cartridge, excess packing buffer must be removed in order to prevent buffer overflow during cartridge operation. The buffer in each chamber is 0.25x TBE. To remove excess buffer:

- Remove and discard **2 ml of buffer from each of the 6 buffer chambers** (A-F), using a 1 ml pipette or a serological pipette to access the buffer chambers through the access holes.
- Remove and discard **all of the buffer (approximately 9 ml) from the sample chamber** taking care not to disturb the sample gel or extraction gel.

Preparing the extraction well

A fresh 60 µl sample of buffer must be added to the extraction well prior to the run in order to ensure consistent run results. The extraction well must then be sealed in order to prevent output volume fluctuations. To prepare the extraction well:

- Remove the small piece of adhesive film covering the extraction well.
- Extract and discard the buffer from the extraction well (approximately 60 µl).
- Transfer 60 µl of buffer from any of the buffer chambers into the extraction well.
- Ensure the surface of the cartridge above the concentration gel is dry, so that the adhesive film will provide a good seal. Wipe off excess buffer with a Kimwipe if necessary.
- Remove the white backing from the supplied piece of adhesive film and place it on top of the extraction well as shown in Figure 3 below. One edge of the film should stick up above the cartridge slightly to allow for easier removal of the adhesive film after the run. With a gloved

finger, firmly press down on the film, ensuring a good seal on all edges and taking care to avoid wrinkles and bubbles.

- Align the extraction well sealing tool to the extraction well, as shown in Figure 4. Firmly press down on the tool until it is fully compressed, as shown in Figure 4. Release and then press down a second time to ensure a good seal.

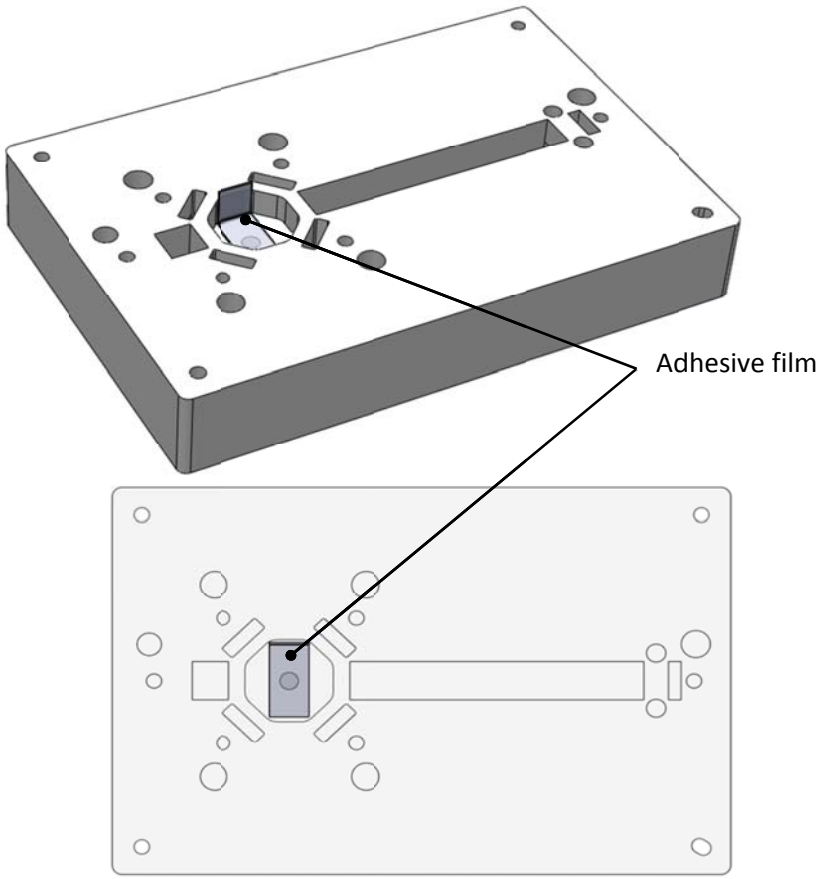

Figure 3. Cartridge with adhesive film over extraction well.

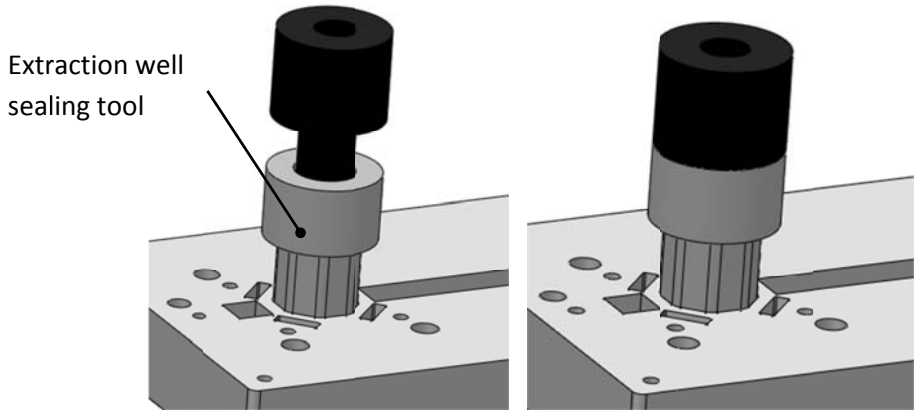

Figure 4. Extraction well sealing tool aligned and uncompressed (left) and fully compressed (right).

Placing the cartridge on the cold plate

To load the cartridge into the Aurora instrument:

- Pull gently outward at both left and right sides of the cartridge drawer until the drawer slides out and latches in the open position
- Examine the cold plate and the metal bottom of the cartridge for debris that may prevent the cartridge from sitting flat on the cold plate. Remove any debris that is found.
- Pipette 1 ml of water onto the center of the cold plate and place the cartridge onto the cold plate oriented so the concentration gel is on the left hand side, as pictured Figure 5 below.
- Ensure that the cartridge is sitting flat on the surface of the cold plate and is not resting on top of the cartridge registration features.

These steps ensure good thermal contact between the cartridge and cold plate.

Final cartridge inspection

Inspect the empty sample chamber prior to loading your sample. If any buffer has leaked into the sample from the other reservoirs, use a pipette to remove the buffer and immediately proceed to the next section, **Load the sample**. For best results, the sample should be loaded and the run started as quickly as possible once you have removed the buffer from the sample chamber.

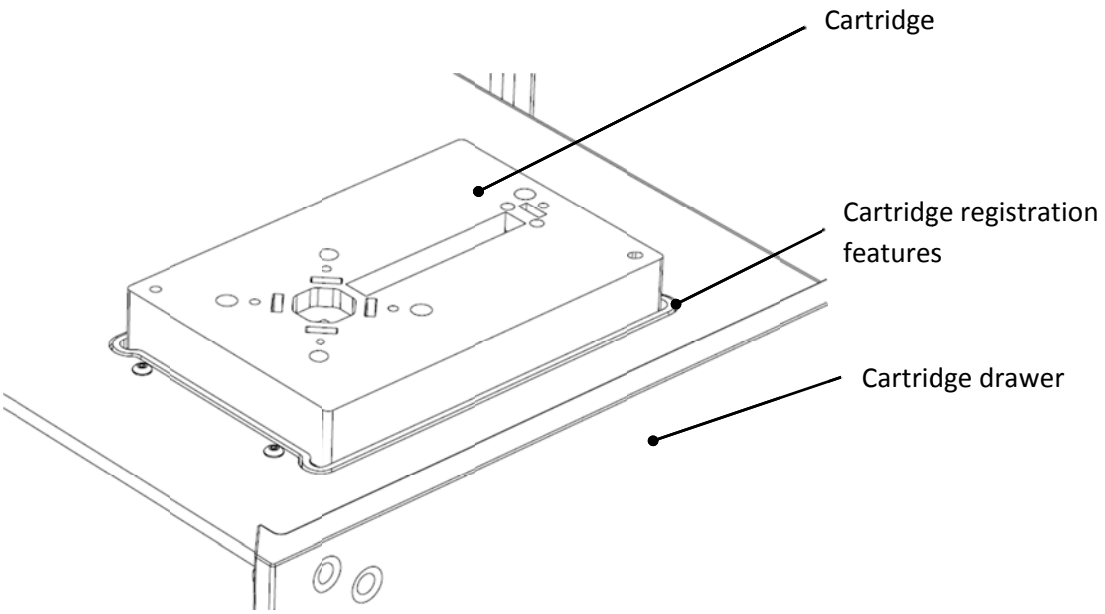

Figure 5. Cartridge oriented correctly on the cold plate. The focusing gel is on the left-hand side.

Load the sample

Bring the volume of the sample you prepared earlier up to 5 ml with deionized water. Transfer the entire 5 ml sample into the empty sample chamber (see Figure 1). Close the cartridge drawer.

Prepare the Aurora

Prepare the Aurora and chiller as described in the user manual. Briefly, turn on the chiller and the Aurora. Start the chiller and ensure that a plus or minus symbol appears on the display to indicate that thermal control is operating. If a star appears, press Start again.

Begin the run

From the home screen of the Aurora software, load and run the protocol appropriate for your application (ex. 106-0001-BA-D AURORA DNA CLEAN-UP PROTOCOL) as described in the "Operating the Aurora" section of the Aurora User Manual. Create an experiment folder for the run to save the logs.

The run will typically complete in 2 - 4 hours depending on the protocol that you are running.

Sample extraction

After a run is complete, to extract the concentrated sample:

- Pull gently outward at both left and right sides of the cartridge drawer until the drawer slides out and latches in the open position.
- Lift the cartridge from the platform, taking care not to spill any liquid, and place the cartridge on a flat work surface .
- Examine the concentration gel and ensure that there are no large bubbles present. Presence of large bubbles indicates that the gel overheated during the run, which will adversely affect the quality of the output.
- Using forceps or gloved fingers, remove the clear film covering the extraction well in the center of the gel.
- Transfer concentrated DNA from this well using a pipette, taking care not to disturb the concentration gel.
- Typically, two pipetting steps are required in order to extract most of the liquid from the extraction well. Be sure to extract any sample that is suspended on the walls of the extraction well.
- The expected output volume is 50-60 µl, but may vary from 40-70 µl depending on sample and run conditions.

Disposal

Dispose of the cartridge and contents following all applicable policies, laws, and regulations.

Aurora cartridges, buffers, and gels are non-hazardous as shipped, however cartridges that have contained hazardous samples (including many nucleic acid stains) may be considered hazardous in your jurisdiction.

Important

The user is responsible for the safe use, transport, storage, and disposal of any materials that may be considered a chemical or biological hazard. Refer to accompanying material safety data sheets (MSDS) for safety information and handling instructions, and comply with all federal, state/provincial, municipal/local, and institutional requirements and guidelines for disposal.

Troubleshooting

Please see the Aurora User Manual for more information about troubleshooting instrument faults. Troubleshooting information related to processing your sample can be found in your protocol documentation.

1. The Aurora or the camera does not connect to the computer.

Unplug the instrument's USB cable, wait five seconds, and reconnect the cable.

2. **A cartridge fails a contact test.**

   Before each run, the Aurora checks to make sure the instrument has good electrical contact with the cartridge. Poor contact can be caused by misalignment of the cartridge. Re-seat the cartridge firmly within the registration features on the cold plate. Check that the correct contact plate is installed. If your contact plate has spring pin electrodes, check to ensure that none of the spring pins are jammed in the up position. If you notice stuck pins, see the Aurora manual for cleaning instructions. If your contact plate has coiled spring electrodes, check to make sure there is nothing blocking contact with any of the springs and the graphite electrodes on the cartridge, such as PCR tape or salt.

3. **A "sample current is below minimum current limit" warning appears.**

   The Aurora may not be making electrical contact with the cartridge. Pause the run, open the cartridge drawer, check that the cartridge is firmly seated within the registration features on the cold plate, close the cartridge drawer, and press the play button to resume. If the cartridge fails the contact check, see above.

4. **A temperature controller error appears.**

   Make sure the chiller is on and operating. If you see a ∗ on the display, the chiller is pumping coolant but temperature control is not active. Press Start/Stop on the chiller to enable temperature control.

5. **Error: The Aurora control software warns that the sample is too conductive.**

   Running high conductivity samples will result in lower yields and may cause gel damage and other issues during the run. The Aurora instrument will give the warning "Injection Conductivity test failed. Sample conductivity is too high. Injection might fail" for highly conductive samples. Conductivity for a 5 ml sample should be ≤100 µS/cm. Please refer to the Troubleshooting section in your Protocol document for further instructions on reducing sample conductivity.

Ordering and support

For support for Aurora protocols or cartridges, or to order additional cartridges, please contact support@borealgenomics.com. This protocol uses cartridge part number 210-0001-CA-D.