

## **PCR Array FAQ's**

### **PCR Arrays: Placing an order**

#### **What PCR Array services does the facility provide?**

The DNA Analysis Facility provides support for gene expression studies using PCR Array technology from a number of different companies, SABiosciences, Applied Biosystems, and Lonza for example. This service includes the cDNA reaction, running the PCR Array and training for the data analysis.

It is the responsibility of the user to purchase the PCR Array plates and to supply the facility with RNA which meets certain quality specifications. In many cases we can provide a quote to purchase PCR Arrays at a discounted price. Please see the facility to learn about these discounts. You will need to purchase PCR Array plates that are compatible with our real-time instruments. Most PCR Arrays will be run on our ABI Prism 7900HT Sequence Detection System (Standard NOT Fast block). Please check with the staff before ordering your plates.

#### **How do I place an order?**

1. Login to iLab.
2. Find the 'Request for Services' tab, and then locate 'Sample Submission and Laboratory Services' heading, open the VCC DNA Analysis Facility folder.
3. Select the [Gene Expression] – PCR Arrays request and fill out the necessary information on the order form. **\*\*Note:** Please adjust the quantity field in both locations on the order form'

**1) Forms and Request Details** (see bottom of list to add items to this request) ⌵

May 20 10:18 AM [Gene Expression] - PCR Arrays VCC DNA Analysis Facility Quantity: 1.0 \$144.40 Billing Status: Not Ready To Work Status: Proposed

[View Form: PCR Array Order Form](#) Visible Not Started page guide

**ATTENTION: Please adjust the Quantity above to reflect the number of samples you are submitting for this order.**  
 Please contact the facility staff before purchasing your plates. We can provide you a quote to purchase plates from SABiosciences at a cheaper rate and it is IMPORTANT to purchase the correct type of plates for our qPCR Instrument.  
 Fill out all information, SAVE the completed form and then SUBMIT the form.

[Save Progress](#)

★ Number of Samples (Be sure to change the Quantity field above to reflect the number of samples!)

★ Species

★ Pathway

★ Catalog #

Comments or Special Conditions

★ How was RNA integrity tested?

★ Would you like the DNA Facility to test your RNA for genomic contamination?

★ Amount of RNA you would like us to use in the cDNA Reaction

**Sample Information** Click the green plus button to add one row for each of your samples.

Sample Information Sample Name RNA Concentration (ng/uL)

Add  row(s)

Please save your form! [save completed form](#) [save draft of form](#) [lock form](#)

4. When you have completed filling out the service request, please press ‘save completed form’

Please save your form! [save completed form](#) [save draft of form](#) [lock form](#)

5. Remember to press ‘submit request to core’

[submit request to core](#)

## **What information will I need to provide when I place an order?**

Please read the iLab order form thoroughly. All of the information that we need to process your order in a timely fashion is contained in the order form.

**Array Information:** You will need to indicate the amount of RNA that you would like to use in the cDNA reaction and if you would like a gDNA test of the RNA done prior to the cDNA set up. You will need to include the species, pathway and catalog number of your PCR Array plate along with the number of plates to be run.

**Sample Information:** You will need to provide the sample name, RNA concentration, and indicate if the sample is a control sample or a test sample.

## **How do I print a copy of my service request?**

After your service request has been submitted, locate the pencil icon on the right hand side of the screen. We suggest that you print to the web format.



## **Where do I put my samples?**

Your RNA samples should be brought in a box labeled with your name, the date and the Principle Investigator of the Lab. Your PCR Array plates can be brought in their original box, but should also be labeled with your name, the date, and the Principle Investigator of the Lab. These should be placed in the freezer marked “TaqMan Samples”. Please include any strip caps that come with your order. If you have opted for the facility to test your RNA for gDNA contamination, you need to provide a diluted sample of your RNA for testing. To determine the proper concentration, take the starting amount of RNA you want to use on your PCR Array plates and divide by 100. (500ng starting RNA/100= 5ng/uL). These tubes should be carefully labeled so as not to be confused with the RNA tubes for the PCR Array plates.

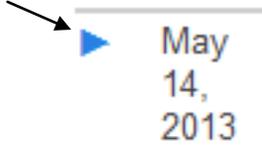
## **When will my samples be run and how will I be notified?**

Turn-around will vary depending on the number of plates being submitted.

You will receive an email from iLab when your samples are ready. We sometimes edit this message to have specific remarks about how your samples ran, so please look at this message!

## How do I download my data from iLab?

You will receive an email from iLab letting you know you have data available. Please login to iLab and navigate to your service request. To find your data, open your service request by clicking the blue arrow.



Your data will be attached to your service request under the heading “Attachments and URL’s in a zipped file.



Service requests are only stored on the iLab system for 18 months. We strongly urge you to download your data to a safe place.

## PCR Arrays: Sample Information

### RNA Quality:

-All RNA **must** be tested for intactness through a Bioanalyzer analysis or a gel analysis. We recommend that all samples have a RIN over 6.

-A DNase 1 treatment **must** be performed on all RNA submitted since not all primer sets on the plate cross exon/exon boundaries and WILL amplify gDNA. It is **HIGHLY** recommended that you test your RNA for gDNA contamination before submitting RNA for PCR Array analysis. This can be done by the facility in our standard real-time gene expression service. The facility has primer sets available for use, or you can supply your own.

## **RNA Quantity:**

-We recommend using 100ng-1ug RNA per plate for the cDNA reaction. The reaction conditions are set up to deliver this total amount of RNA in  $\leq 8\mu\text{L}$ . Just as with other qPCR gene expression studies, it is important to set up all samples with the same starting amount of RNA. It is possible to go below 100ng starting RNA, but remember that you will begin to lose expression of low amplified targets.

## **PCR Arrays: Data Analysis**

It is important to look at the raw data from your plates before moving on to the data analysis. We have copies of the SDS software in our workstation, or we can provide you with the software for your own computer. This will allow you to look at the amplification plots, set baseline and thresholds, and to view the dissociation curves. The data can then be exported for data analysis. The facility will provide the training for this.

-SABiosciences has developed a web-based data analysis for easy selection of house-keeping genes, calculation of differential gene expression as compared to a control group, level of gDNA contamination, PCR inhibition, and RT efficiencies. You can find the website at:

<http://www.sabiosciences.com/pcr/arrayanalysis.php>

Lonza StellarArray™ qPCR Arrays can be analyzed with their web based data analysis known as Global Pattern Recognition™ Data Analysis Tool. You can find the website at:

<http://array.lonza.com/gpr/>

Applied Biosystems TaqMan® Array Gene Signature Plates can be analyzed with their freeware known as Data Assist.

<http://www.invitrogen.com/site/us/en/home/Products-and-Services/Applications/PCR/real-time-pcr/real-time-pcr-assays/taqman-gene-expression/taqman-expression-arrays.html>

These companies all provide support for their data analysis, but the facility is happy to provide a tutorial for you.

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