

Luminex Bioscience Group

FlexmiR™

FlexmiR™ MicroRNA Human Panel

Instruction Manual

for product #

BG-FMIR-H20-8.0

Using LNA™ Technology from



For Research Use Only. Not for use in diagnostic procedures.

Version A (November 2006)



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Literature citations:

Please refer to "FlexmiR™ MicroRNA Assay" whenever describing this method for publication.

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PRODUCT SUMMARY

Kit Contents

The FlexmiR™ MicroRNA Human Panel consists of 9 components as described in Table 1.

Table 1.

Kit components	Amount supplied	Cap color
Human Pool 1	20 reactions	Blue
Human Pool 2	20 reactions	Yellow
Human Pool 3	20 reactions	Green
Human Pool 4	20 reactions	White
Human Pool 5	20 reactions	Red
Reporter Molecule*	36 µL	Amber
Hybridization Solution	1.6 mL	Clear
Wash Solution	45 mL	Clear
Nuclease Free Water	3.5 mL	Clear

*SAPE: streptavidin-phycoerythrin conjugate, 1 mg/mL

Shipping and Storage

The FlexmiR MicroRNA Human Panel is shipped on blue ice. Upon receipt, the components should be stored as individually labeled. Use properly stored reagents within three months of receipt.

Additional Required Materials (user-supplied)

Luminex recommends:

FlexmiR™ MicroRNA Labeling Kit (product # BG-FMIR-L20)

For biotin labeling of microRNAs from total RNA samples

FlexmiR™ MicroRNA Control Set (product # BG-FMIR-C20)

For ensuring assay integrity

- Biotinylated total RNA test sample
- Deionized water
- Nuclease-free PCR tubes, PCR strips or 96-well plate
- Heating block or thermal cycler
- Bath sonicator
- Vortex mixer
- Adhesive aluminum foil sealer for 96-well plates, Costar cat. #6570
- Vacuum manifold (Multiscreen HTS Vacuum Manifold MSVM HTS)
- Vacuum pressure pump
- Plate shaker
- 96-well Thermowell P polycarbonate clear PCR plates, Costar cat.# 6509

- Sterile, nuclease-free filter barrier pipette tips
- 1.2 µm Millipore filter plate (Multiscreen Styrene MSBV N12XX or MSBV S1210)
- Luminex[®] 100™ or Luminex[®] 200™ analyzer with Luminex IS™ Software Version 2.3 or greater

Related Products

FlexmiR™ MicroRNA Mouse/Rat Extension Panel (product # BG-FMIR-M20-8.0)

FlexmiR™ MicroRNA Labeling Kit (product # BG-FMIR-L20)

FlexmiR™ MicroRNA Control Set (product # BG-FMIR-C20)

Product Description

The FlexmiR MicroRNA Labeling Kit labels uniformly across all RNA molecules using a simple and fast 2-part protocol (see Workflow overview, page 7). In the first part, a biotin label is attached enzymatically to the 3'-end of the RNAs molecules in the total RNA sample. This is followed by an enzyme inactivation step after which the sample is ready for hybridization.

The FlexmiR MicroRNA Human Panel sensitively measures the expression of the miRBase sequence database (version 8.0) human miRNA sequences by combining xMAP[®] technology and Locked Nucleic Acid (LNA™) technology. The integration of these technologies allows precise detection of miRNAs without prior need for sample RNA size fractionation or amplification. Discrimination between closely related miRNA family members is achieved by the use of LNA-enhanced capture probes complementary to the mature miRNA targets. The capture probes have been T_m-normalized to hybridize optimally under the conditions described in this protocol by varying the LNA content and the length of the capture probes.

Locked Nucleic Acid (LNA™) is a conformationally restricted nucleic acid analogue in which the ribose ring is "locked" with a methylene bridge connecting the 2'-O atom with the 4'-C atom. Incorporation of LNA in one strand of a nucleic acid duplex increases the melting temperature of the duplex by 2-8° C/ LNA monomer.

The general flow of the FlexmiR MicroRNA Assay allows the end-user to work on samples of total RNA to conduct the steps as follows: a biotinylation step that biotinylates the 3' end of RNA (FlexmiR MicroRNA Labeling Kit), followed by a hybridization step where the labeled miRNA hybridizes specifically to LNA-spiked capture probes on xMAP Carboxylated Microspheres. The detection of the biotinylated miRNA is achieved by the reaction with the Reporter Molecule (SAPE) and final read of the samples in a microtiter plate on a Luminex 100 or Luminex 200 analyzer with Luminex IS Software Version 2.3 or greater versions of the software.

The FlexmiR MicroRNA Human Panel includes a minimum of 97% of the annotated human sequences listed in version 8.0 of the miRBase microRNA sequence database. Please visit the Luminex microRNA resource site at www.luminexcorp.com/microRNA for information regarding the included sequences.

IMPORTANT: The assay can provide quantitative results if standard material specific to a particular miRNA is obtained separately through an oligonucleotide vendor, as standards are not provided with the panel.

Definitions

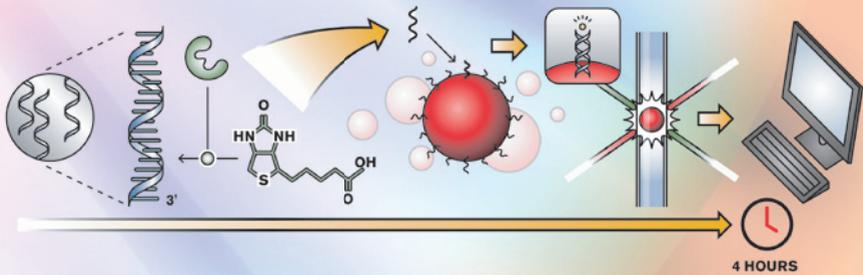
Panel

A panel refers to the entire collection of assay targets performed against one unknown sample. For example, the FlexmiR MicroRNA Human Panel collates data from multiple microsphere pools (defined below) performed against an unknown sample to provide a panel of results.

Pool

A microsphere pool is a collection of 60-100 capture probe-bound microspheres mixed together into one vial. Since the FlexmiR MicroRNA Panels involve assay targets in excess of 100, each species-specific assay includes multiple pools to achieve coverage of the miRBase sequences.

FlexmiR MicroRNA Assay - Work flow



Biotin-label total RNA

FlexmiR MicroRNA Labeling Kit

Hybridize to FlexmiR microspheres

Wash

Add SAPE

FlexmiR MicroRNA
Panel

Detect hybridized microRNAs on the Luminex analyzer

Controls

Normalization Microspheres

Four normalization microspheres, regions 72, 73, 74 and 76, are included in each human pool. These normalization microspheres each contain a unique LNA-capture probe specific for ubiquitous small nucleolar RNA (snoRNAs), which are expected to be present in any total RNA sample. The normalization microspheres are useful for normalizing the resulting Median Fluorescent Intensity (MFI) across all the pools for a given sample.

Control Microspheres and Capture Probes

Five control microspheres, regions 83, 84, 85, 86 and 87, are included in each human pool. These control microspheres each contain a unique LNA-capture probe specific to synthetic RNA oligonucleotides, which have no biological equivalents.

The FlexmiR MicroRNA Control Set includes two mixtures of five synthetic RNA oligonucleotides, one mixture is biotinylated (Biotinylated Control) and the other mixture is not (Non-Biotinylated Control). Both controls are specific to the probes attached to the microspheres in regions 83-87, so only one control, either the Biotinylated Control or the Non-Biotinylated Control, may be used per well.

The Biotinylated Control is provided as a stand-alone verification that the panel is performing as expected without the variable of the biotinylation labeling. It may also be used to spike into the biotin-labeled total RNA sample post-labeling to verify the integrity of the assay. The Biotinylated Control is also useful for becoming familiar with the assay or troubleshooting the assay without using precious RNA sample.

The Non-Biotinylated Control is provided to spike into the unlabeled total RNA sample to verify the integrity of the labeling protocol as well as the panel protocol. It also provides a reference from which to normalize results between different samples. The Non-Biotinylated Control is also useful for normalizing results within samples, similar to the normalization microspheres, when using samples that have been enriched or samples that do not contain snoRNAs.

Sample Analysis

Luminex analyzer settings

A high reporter gain setting for the Luminex analyzer is recommended to provide best results. Guidelines for modifying the gain setting and calibrating the analyzer to this setting are provided below.

IMPORTANT: If the analyzer is used for other assays that utilize the low reporter gain setting, modify and calibrate the setting back to the original setting following this assay.

After completing the analyzer startup procedures, which include warm-up, prime, alcohol flush, and two washes, the analyzer must be calibrated. Make sure the probe height is set for the plate type being used for calibration.

Tip: Steps 1-14 of the below instructions may be automated by using the calibration-control template (FMIR_Calibration_Control_Template) provided at www.luminexcorp.com/microRNA.

1. In the Maintenance tab, click on the CAL1 icon. On the Confirmation Screen that appears, click New CAL1 Target.
2. Input (or select if present in the pull-down list) the CAL1 and CAL2 lot numbers and corresponding expiration dates. For CAL1, enter (or verify) that the DD, CL1, and CL2 values on the screen for that lot match those printed on the CAL1 bottle. For CAL2, enter (or verify) that the RP1 value matches the stated RP1 Target Value on the CAL2 bottle label. When done, click OK.
3. On the Confirmation Screen use the pull-down bar at the right to select the location of CAL1 on the plate being used.
4. Click Eject/Retract to open the Luminex XYP instrument.
5. Remove CAL1 from 2° C to 8° C storage and vortex the container to ensure homogeneity. Open the bottle and invert it such that it is perpendicular to the plate and squeeze to dispense 4-5 drops from the dropper tip into the well specified on the Confirmation Screen.

IMPORTANT: The number of drops used for performing calibration and controls on the Luminex analyzer depends on the type of microtiter plate used. For example, 5 drops are recommended when using a flat bottom microtiter plate. Fewer drops may be necessary for other microtiter plate types that have lower volume capacity, but be careful to ensure that sufficient reagent volume is provided to not create air in the Luminex analyzer.

6. Click Eject/Retract and then click OK to start calibration for CAL1.
7. Once CAL1 calibration has passed successfully, on the "Maintenance" tab, click on the CAL2 icon. On the Confirmation Screen that appears repeat steps 4-6 for CAL2.
8. After both CAL1 and CAL2 have passed successfully, click on the CON1 icon in the Maintenance tab.

9. Input (or select if present in the pull-down list) the CON1 and CON2 lot numbers and corresponding expiration dates. For CON1, enter (or verify) that the Target A1, Target B1, and Target C1 values for the 5 references are correct. For CON2, enter (or verify) that the Target A1 values are correct for the 4 references. All these values can be found on the certificates of quality that accompanied the control beads or off the FAQs page at www.luminexcorp.com. When done, click OK.
10. On the Confirmation Screen use the pull-down bar at the right to select the location of CON1 on the plate being used.
11. Click Eject/Retract to open the Luminex XYP instrument.
12. Remove CON1 from 2° C to 8° C storage and vortex the container to ensure homogeneity. Open the bottle and invert it so that it is perpendicular to the plate and squeeze to dispense 4-5 drops from the dropper tip into the well specified on the Confirmation Screen.
13. Click Eject/Retract and then click OK to start confirmation of calibration using CON1.
14. Once CON1 has passed successfully, on the Maintenance tab, click on the CON2 icon. On the Confirmation Screen that appears repeat steps 11-13 for CON2.
15. After both CON1 and CON2 have passed successfully, click on the CAL2 icon in the Maintenance tab. Click on the CAL2 icon. On the Confirmation Screen that appears, click New CAL2 Target.
16. In the Update CAL Targets dialog box, set up a new CAL2 lot using the CAL2 lot number from the bottle plus a designator, such as "A1234-high". Calculate the new RP1 Target Value by multiplying the RP1 target value provided on the label of the CAL2 bottle by 4.55 (e.g. $3832 \times 4.55 = 17436$). Enter this calculated value as the new RP1 Target Value and click OK. **This increases the reporter gain setting.**

IMPORTANT: After the assay is run, the machine will require recalibration back to the original CAL2 lot number RP1 Target Value to lower the reporter gain to the original setting.

17. On the Confirmation Screen use the pull-down list at the right to select the location of CAL2 on the plate being used.
18. Click Eject/Retract to open the Luminex XYP instrument.
19. Remove CAL2 from 2° C to 8° C storage and vortex the container to ensure homogeneity. Open the bottle and invert it such that it is perpendicular to the plate and squeeze to dispense 4-5 drops from the dropper tip into the well specified on the Confirmation Screen.
20. Click Eject/Retract and then click OK to start high reporter gain setting calibration using CAL2.
21. After calibration is complete, clean the system by clicking Wash on the Maintenance Tab. Make sure the reservoir has deionized water in it before washing. Repeat for a total of 4 washes.

Sample Setup

Prior to running the sample analysis, it is important that the Luminex analyzer is loaded with the appropriate template files. Software templates are available for use with the Luminex IS Software Version 2.3 at www.luminexcorp.com/microRNA. There are five templates to download, one for each pool of microspheres. To set up the assay, create a New Multi-Batch under the File menu as described in the Luminex IS Software Manual for Version 2.3/Procedures/Batch Setup Procedures/Create a Multi-Batch.

IMPORTANT: A background control, such as nuclease-free water treated in same manner as sample, is recommended for each microsphere pool to use in determining background MFI for each pool. Also, each template begins with 2 wash cycles using deionized water from the XYP analyzer reservoir. If not using the provided templates, be sure to include 2 wash cycles between each different microsphere pool to ensure best results.

When using the Luminex IS software templates, group the samples such that each microsphere pool is run together. For example:

To perform the entire Human Panel, which consists of five microsphere pools or human pools, for five total RNA samples, where:

Key	Description
P1	IS Template for Human Pool 1
P2	IS Template for Human Pool 2
P3	IS Template for Human Pool 3
P4	IS Template for Human Pool 4
P5	IS Template for Human Pool 5
Sx	Sample number, for example: S1=Sample 1
*	Indicates the analyzer will perform 2 wash cycles from the XYP reservoir prior to testing from the indicated well
BC	Background control

Fill the XYP analyzer reservoir with deionized water and setup the plate as follows:

	1	2	3	4	5	6	7	8	9	10	11	12
A	P1-BC*	P2-S2	P3-S4	P5-BC*								
B	P1-S1	P2-S3	P3-S5	P5-S1								
C	P1-S2	P2-S4	P4-BC*	P5-S2								
D	P1-S3	P2-S5	P4-S1	P5-S3								
E	P1-S4	P3-BC*	P4-S2	P5-S4								
F	P1-S5	P3-S1	P4-S3	P5-S5								
G	P2-BC*	P3-S2	P4-S4									
H	P2-S1	P3-S3	P4-S5									

PROTOCOL

Before starting the experiment

- **Labeling of RNA:** Biotin-label 2 µg of total RNA per microsphere pool. For example, the human panel contains five microsphere pools, so use 10 µg for the entire panel. When using the human panel in combination with the mouse/rat extension panel that contains two microsphere pools, prepare 14 µg total RNA. Luminex recommends using the FlexmiR MicroRNA Labeling Kit for labeling of the RNA sample(s). Please visit www.luminexcorp.com/microRNA to learn more about this product.

IMPORTANT: Human sourced material should always be handled as potential biohazard material. Use Universal Precautions in the handling of all human sourced materials. (There is no human sourced material in the labeling kit or panel components).

- **Analyzer startup:** Ensure the Luminex analyzer has been through manufacturer-recommended start up procedures and is calibrated according to the procedures detailed under “Luminex analyzer settings” starting on page 9. Refer to the appropriate analyzer instruction manual for more information about start up procedures.

Preparations during the experiment

- **Preheating of Wash Solution:** Before reaching step 18 of the protocol, preheat an appropriate volume (min. 200 µL per well) of Wash Solution to 60° C. Keep remaining Wash Solution at room temperature.
- **Preparation of Reporter Solution:** Shortly before reaching step 23 of the protocol, prepare fresh reporter solution by diluting the Reporter Molecule (SAPE) in room temperature Wash Solution to 10 µg/mL (1:100 dilution) in a nuclease-free container. Protect this solution from light. Twenty five µL of reporter solution is required for each reaction.

Experimental Procedure

IMPORTANT: The human pools and Reporter Molecule should be protected from prolonged exposure to light throughout this procedure.

Protocol Step	Detail
1. Verify analyzer setting and setup assay	Verify that the Luminex analyzer is set to the high reporter gain as described under "Luminex analyzer settings" starting on page 9 of this manual. Setup the assay in the analyzer software as described under "Sample setup" starting on page 11.
2. Label total RNA sample(s)	Prepare biotin-labeled total RNA sample(s) using the FlexmiR MicroRNA Labeling Kit.
3. Dilute labeled total RNA sample(s)	Dilute the labeled sample in nuclease-free water to a concentration of 1 µg RNA/10µL. For example, when using 10 µg total RNA from the labeling procedure, add 60 µL of nuclease-free water to the labeled sample to achieve a volume of 100 µL. This allows for the use of 20 µL (corresponding to 2 µg labeled total RNA) per hybridization (step 9 of this protocol).
4. Select the appropriate Human Pool(s)	Please refer to www.luminexcorp.com/microRNA for information regarding specific microRNA sequences contained in each human pool.
5. Resuspend Human Pools	Resuspend microspheres in each of the selected human pools by vortexing followed by bath sonication for approximately 20 s. If preparing a large number of samples, occasionally resuspend using this technique.
6. Add 16 µL of appropriate Human Pool to each sample or background well	The assay should be prepared in nuclease-free PCR tubes, strips or 96-well plate(s). A separate well, or tube, is required for each human pool being tested. For example, if testing 5 human pools for one tissue sample, plan to use 5 wells to perform the testing of that sample.

IMPORTANT: Do not mix microspheres from different pools in the same well.

7. Add 14 μ L of Hybridization Solution to each sample or background well

8. Add 20 μ L Nuclease-Free Water to each background well

9. Add 20 μ L of diluted labeled total RNA sample to each sample well

For best results, use 2 μ g of labeled total RNA (20 μ L) per reaction or well. If the volume of the RNA is less than 20 μ L, add nuclease-free water to reach 20 μ L total volume.

The volume of the labeled RNA must not exceed 20 μ L. The sample volume can be reduced by using a Speedvac (Savant, product# 74104 or equivalent).

10. Mix reagents

Mix reagents gently by pipette mixing. Avoid foaming the reagents.

11. Incubate at 95-100° C for 3 min.

Close the tubes (or if using a 96-well plate, cover the reaction plate with adhesive aluminum foil) to prevent evaporation and light exposure. Incubate at 95-100° C for 3 min. to denature any secondary structure in the sample.

Steps 11 and 12 can be combined with the use of a thermal cycler programmed as follows:

Hold at 95° C, 3 min.

Hold at 60° C, FOREVER

12. Incubate at 60° C for 60 min.

Make sure that the samples are protected from exposure to light during the hybridization reaction.

Tip: Perform steps 13-15 during the hybridization incubation to prepare for washing the reaction and to reduce reaction temperature cooling prior to washing.

13. After the incubation, pre-wet a 96-well filter plate with 100 μ L of Wash Solution

Place the filter plate on the inverted filter plate lid and add 100 μ L of Wash Solution to the required number of wells.

Tip: Small amounts of liquid can pass through the filters and accumulate on the lower side of the filters, i.e. at the bottom of the filter plate. Placing the filter plate on the inverted plate lid between steps throughout the protocol will elevate the plate slightly and thereby reduce the risk of contamination of the filters from neighboring wells.

TIP

TIP

14. Incubate the filter plate at room temperature for 1 min.

During this incubation, cover any unused wells with adhesive aluminum foil.

15. Transfer the filter plate to the vacuum manifold and filter the Wash Solution

As soon as the liquid has passed through, place the filter plate on the inverted filter plate lid.

Tip: Remove the filter plate from the vacuum manifold as soon as the liquid has passed through as overdrying the filters can compromise their performance.

16. Transfer the hybridization reactions to the filter plate wells

Gently pipette up and down three times, and then transfer the hybridization reactions to the filter plate. Avoid foaming the reagents.

17. Transfer the filter plate to the vacuum manifold and filter the hybridization reactions

When the liquid has passed through, place the filter plate on the inverted filter plate lid.

18. Carefully add 100 μ L of preheated (60° C) Wash Solution to each well

19. Transfer the filter plate to the vacuum manifold and filter through the Wash Solution

When the liquid has passed through, place the filter plate on the inverted filter plate lid.

20. Repeat steps 18-19

Repeat steps 18 and 19 (using additional pre-warmed wash solution) for a total of 2 washes per reaction.

21. Blot the bottom of the filter plate on a clean tissue to remove surplus liquid.

Place the plate on the inverted filter plate lid and immediately proceed to the next step.

22. Add 50 μ L of Wash Solution (room temperature) to each well

TIP

23. Add 25 μ L of fresh reporter solution to each well

Prepare fresh reporter solution by diluting the Reporter Molecule (SAPE) in room temperature Wash Solution to 10 μ g/mL (1:100 dilution) in a nuclease-free container. Protect this solution from light.

24. Mix on a plate shaker at room temperature for 15 minutes at 600 rpm

Protect this reaction from light using aluminum foil.

25. Transfer each sample to a Costar 96-well ThermoWell P polycarbonate clear PCR plate

Transfer each sample to a Costar plate and fill the XYP analyzer reservoir with deionized water (as instructed in the Sample Setup starting on page 11) to allow the Luminex analyzer to perform 2 wash cycles prior to each different microsphere pool.

Tip: Verify that probe height is properly adjusted for the plate used for reading samples.

26. Analyze 50 μ L of each sample (room temperature) on the Luminex analyzer according to the appropriate system manual

27. Reset reporter gain setting

If the analyzer is used for other assays that utilize the low reporter gain setting, modify and calibrate the reporter gain setting back to the original CAL2 setting. Refer to the instructions provided under "Luminex analyzer settings" starting on page 9 of this manual, steps 1-14.

TIP

DATA ANALYSIS

Background subtraction

For each of the five human pools a background control (i.e. water “hybridized” to the microsphere pool instead of labeled RNA) should be measured. The Median Fluorescence Intensity (MFI) obtained from this background control should be subtracted from all sample MFIs in the relevant microsphere pool.

Normalization

If the quality of the RNA sample preparation and quantification is consistent between samples, the normalization methods described below will provide good quality data analysis. However, it is important to note that these methods only enable correction for variation in hybridization or labeling efficiencies. They do not normalize for variation between the biological samples (e.g. differences in RNA quality or input amount).

Normalization using Normalization Microspheres

Four normalization microspheres, regions 72, 73, 74 and 76, are included in each human pool. These normalization microspheres each contain a unique LNA capture probe specific for ubiquitously expressed small nucleolar RNA (snoRNAs). The normalization microspheres can be used to:

- Check for consistent hybridization of a labeled total RNA sample across all microsphere pools.
- Correct MFIs for variation in hybridization between microsphere pools for a given labeled total RNA sample.

IMPORTANT: The normalization microspheres do not provide reliable normalization when the RNA samples have been enriched for small RNAs prior to labeling.

After data acquisition is complete, analyze the data as follows:

1. Discard normalization microsphere signals of less than 250 MFI.
2. Calculate the coefficient of variation (CV*) of the MFI's obtained from each normalization microsphere across the five pools.
3. If the CV values are all below 10%, further normalization is not necessary.
4. If one or more of the CV values are above 10%, and if the MFIs obtained from the four normalization microspheres in one pool reflect a general tendency for higher or lower signals than in the other pools, normalization is recommended.

Example: A sample is hybridized to all five human pools. In one of the five pools, the four normalization microspheres MFIs are 13%, 20%, 15% and 16% lower than the average MFIs of the normalization microspheres in the other four human pools giving an average signal decrease of 16%. All MFIs from this pool are corrected for this factor:

$$\text{Normalized MFI} = \frac{\text{MFI}_{\text{sample}} - \text{MFI}_{\text{background}}}{1 - 0.16}$$

* The standard deviation of the replicas times 100 divided by the sample mean.

IMPORTANT: snoRNAs cannot be assumed to be equally expressed in different tissues or across different treatments of the same tissue. Therefore, the normalization microspheres cannot be assumed to be suitable for normalization between samples.

Normalization using the FlexmiR MicroRNA Control Set

Luminex recommends the use of the FlexmiR MicroRNA Control Set (product # BG-FMIR-C20) in order to enable more comprehensive normalization of FlexmiR MicroRNA Human Panel data.

The FlexmiR MicroRNA Control Set includes a mixture of five non-biotinylated synthetic control RNA oligonucleotides (Non-Biotinylated Control). Five control microspheres, regions 83, 84, 85, 86 and 87, are designed to specifically target these five control oligonucleotides that have no biological equivalents. The control microspheres are included in each of the five human pools.

When spiked into the total RNA sample prior to labeling, the Non-Biotinylated Control provides several advantages as compared to using the normalization microspheres alone:

- Consistent labeling and hybridization can be assured across microsphere pools and samples.
- Correction of MFIs for variation in labeling and hybridization between microsphere pools and samples is made possible.
- Both total RNA and enriched RNA samples can be normalized.

After data acquisition is complete, analyze the data as follows:

1. Calculate the coefficient of variation (CV) of the MFI's obtained from each control microsphere across the sample wells that are compared.
2. If the CV values are all below 10%, further normalization is not necessary.
3. If one or more of the CV values are above 10%, and if the MFIs obtained from the five control microspheres in a particular pool or sample reflect a general tendency for higher or lower signals than in the other pools or samples, normalization is recommended. This is done in a similar manner as described for the normalization microspheres above.

TECHNICAL SUPPORT

Additional information about microRNA is available on the Luminex website at www.luminexcorp.com/microRNA. Support for miRNA as well as other xMAP topics is available at www.luminexcorp.com.

Also review Frequently Asked Questions from the main Luminex website by clicking Support > Support Login to log into the Support FAQ site, then click on the Support tab. First time visitors may need to register to view all of the FAQ content.

Toll free technical support is available to users in the U.S. and Canada by calling 1-877-785-BEAD (-2323). Users outside of the U.S. and Canada can call at +1 512-381-4397. Inquiries may also be sent by E-mail to support@luminexcorp.com.

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