Sample Labeling Procedure
Step 2: Clean-up Labeled samples

Note: Proceed with the remaining procedures in the dark.

1. Remove the samples from the PCR thermocycler and centrifuge the samples in a picocentrifuge.
2. Place the columns provided upright into the tubes that are provided with the kit.
3. To purify the labelled test samples, to each of the purification columns, add 430 ul of 1X TE (pH 8.0) buffer. To purify the labelled reference, use 1 column to purify 8 reference labelling reactions. So, to the column for purifying 8 labelled reference add 230 ul of TE buffer.
4. To each of the columns containing 430 ul of 1X TE buffer (pH 8.0), add 25 ul of the test sample. To the column used for the reference, containing 230 ul of 1X TE buffer (pH 8.0), pool all of the reference samples into that one column (25X8 ul).
5. Close the column and invert the tube to mix. Spin the samples at 14,000 x g for 10 minutes. Discard the supernatant.
6. Add 480 ul of 1X TE buffer (pH 8.0) to each column. Close cap and spin for 10 minutes at 14,000 x g. Discard the flow through.
7. Repeat step 15 for a total of 3 times, until the flow through is clear.
8. Invert the column into a fresh 2 ml tube (provided in the kit) and spin the sample at 1000 x g for 1 minute.
9. Take OD and concentration at 550 nm for Cyanine-3 dUTP labeled test samples and at 650 nm for Cyanine-5 dUTP labeled reference sample, as well as at 260 nm. Be sure to use 1X TE (pH 8.0) as your blank. Store your samples in the dark at -20 degrees Celsius or proceed to the next step.