Frequently Asked Questions

Promoter Methylation PCR Kit [PN MP1100]

1. Can I use my own primers with the Promoter Methylation PCR Kit?

Yes, you can design your own primers for use with this kit or use primers that you already have, provided that they meet the requirements. The requirements are:

- Locate sequential Mse I restriction sites (TTAA) in your promoter region(s).
- Locate CpG islands between these Mse I sequential restriction sites.
- Forward and reverse primers should correspond to the DNA sequences before and after sequential Mse I sequences, respectively. Be sure that the region between these sequential Mse I sites contains a CpG island.
- The predicted melting temperature (T_m) of your primers should optimally be ~55°C. Primers should be at least 22 nucleotides in length.

2. Can the kit be used with mouse samples?

The Methyl Binding Protein used in the kit can bind any DNA that contains methylated CpG, regardless of the species. So, yes, the kit can be used with mouse samples, but you will need to design your own primers for PCR.

3. Does the kit contain PCR reagents?

No, the kit does not contain the reagents for PCR. You will need to purchase these separately.

4. Do I have to use Mse I to digest the DNA?

We have only tested the use of Mse I in-house. However, any appropriate restriction enzyme can be used with the kit.

5. How many reactions can be run with the kit?

The kit contains 6 columns and enough reagents for 6 isolations of methylated DNA fragments.

6. What MBP protein is used with the kit?

The MBP protein we use is MeCP2. It is the full length protein, and it belongs to the methyl binding protein family, including MeCP2, MBD2, MBD3, and MBD4. MeCP2 is a human protein that can bind DNA containing methylated CpG from any kind of species.

7. What primers are used with this kit?

You can find a list of currently available primers in the MP1100 Tech Tip located at www.media.affymetrix.com/support/technical/usb/tech_tips/MP1100_Primer_List.pdf