

DNA Methylation control package

Cat. No. C02040012

Size: 40 rxns

Expected PCR product size:

Unmethylated control: 92 bp (A. thaliana chr3:20074482 - 20074573)

Methylated control: 81 bp (A. thaliana chr1:30084240 - 30084160)

Format: In solution in TE. The DNA controls are at a concentration of 0.2 ng/μl. The primer sets are at a concentration of 10 μM [5 μM of each primer].

Storage: Store at -20°C; for long storage, store at -80°C. Avoid multiple freeze-thaw cycles.

Precautions: This product is for research use only. Not for use in diagnostic or therapeutic procedures.

Product description

The DNA Methylation control package includes unmethylated and in vitro methylated DNA together with specific primer sets for assessing the efficiency of your Methylated DNA IP (MeDIP) performed with Diagenode’s MagMeDIP and AutoMeDIP kits.

The control DNA has been optimized to be used in combination with Diagenode’s (MeDIP) kits. The MeDIP kit allows you to perform DNA methylation analysis of your sample together with the internal controls in one tube, which minimizes error and permits quality control of each sample (Figure 1). Although the controls have been optimized for MeDIP, they can also be used for other applications studying DNA methylation.

The methylated DNA is obtained from BAC F19K16 which contains chromosome 1 of A. thaliana. The BAC is sheared and the resulting DNA is methylated in vitro with Sss I methylase. The unmethylated DNA is obtained by shearing BAC F24B22 which contains A. thaliana chromosome 3.

The two primer pairs specifically amplify a CpG region of the methylated and unmethylated DNA, respectively. Both primer pairs have been optimized for qPCR.

	Format	Comments	Storage
Methylated DNA	60 μl	0.2 ng/μl	-20°C
Unmethylated DNA	60 μl	0.2 ng/μl	-20°C
Primer pair for methylated DNA	100 μl	10 μM	-20°C
Primer pair for unmethylated DNA	100 μl	10 μM	-20°C

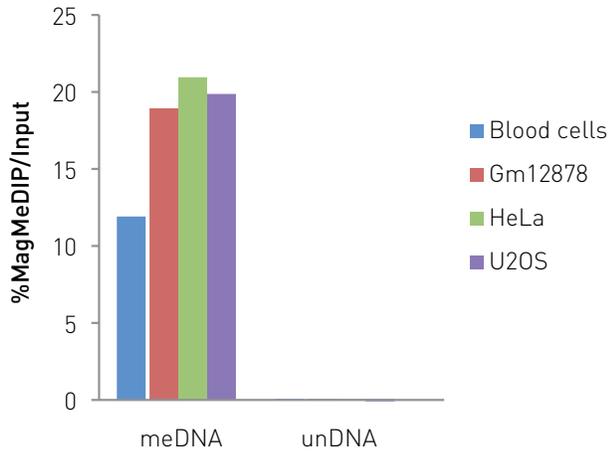


Figure 1.

MeDIP was performed with the MagMeDIP kit using DNA from blood, Gm12878, HeLa and U20S cells. The methylated DNA (meDNA) and unmethylated DNA (unDNA) from the DNA Methylation control package were added as internal positive and negative controls prior to the IP. After the IP, the DNA is isolated and analysed by qPCR with the primer pairs from the package and the recovery (% of input) is calculated. Figure 1 shows that the methylated DNA is recovered, whereas no signal is obtained for the unmethylated DNA.

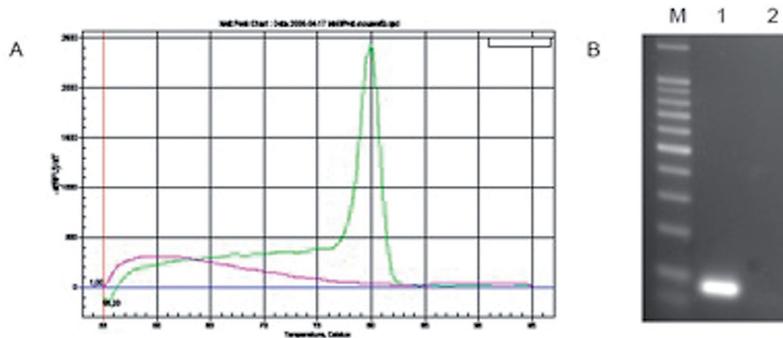


Figure 2

A: Melting curve of the PCR product obtained with the primer pair specific for methylated DNA. Real-time PCR was performed in 25 µl of final volume with 1 µl of provided primers. PCR conditions were as follows: 95°C for 3 min, followed by 40 cycles of [95°C for 15 seconds, 60°C for 45 seconds] and 1 cycle at 72°C for 2 min. The melt curve was obtained with 0,5°C temperature increments from 55°C to 95°C. The result obtained with the methylated DNA is shown in green; the unmethylated DNA shows no amplification (red). **B:** The PCR products were analysed by electrophoresis on a 2% agarose gel. The left lane shows a 100 bp molecular weight ladder. Lane 1 shows an 81 bp amplified fragment. No amplification is observed in lane 2 (PCR with the unmethylated control).

Diagenode sa. BELGIUM | EUROPE

LIEGE SCIENCE PARK
 Rue Bois Saint-Jean, 3
 4102 Seraing (Ougrée) - Belgium
 Tel: +32 4 364 20 50
 Fax: +32 4 364 20 51
 orders@diagenode.com
 info@diagenode.com

Diagenode Inc. USA | NORTH AMERICA

400 Morris Avenue, Suite 101
 Denville, NJ 07834 - USA
 Tel: +1 862 209-4680
 Fax: +1 862 209-4681
 orders.na@diagenode.com
 info.na@diagenode.com