

PRODUCT NAME 5-mC monoclonal antibody cl. b		
Cat. No. MAB-006-100 MAB-006-500	Type: Monoclonal MeDIP-grade IgG1	Size: 100 µg/ 100 µl 500 µg/ 500 µl
Lot #: DA-0019	Source: Mouse	Concentration: 1 µg/µl

Product description: Monoclonal antibody raised in mouse against 5-mC (5-methylcytosine) conjugated to ovalbumine.

Specificity: Human and mouse: positive
Other species: tested in plants

Applications	Suggested dilution	References
MeDIP*	1-5 µg per IP	Fig1
Immunofluorescence	1:100	Fig 3

*Please note that of the optimal antibody amount per IP should be determined by the end-user. We recommend testing 1-5 µg per IP.

Purity: Monoclonal antibody purified by gel filtration, in PBS containing 0.05% azide.

Storage: Store in small aliquots at -80°C. Avoid multiple freeze-taw cycles.

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

Last data sheet update: September 9, 2011

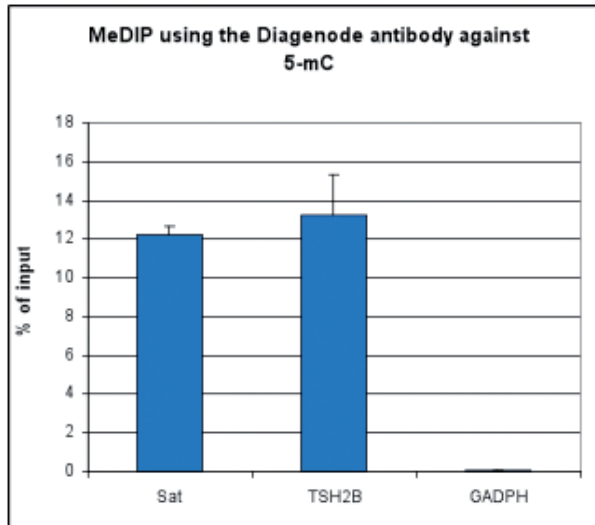


Figure 1
Methylated DNA immunoprecipitation (MeDIP) results obtained with the Diagenode monoclonal antibody directed against 5-methylcytosine

MeDIP was performed on the IP-Star automated system using fragmented genomic DNA from U2OS cells, the Diagenode monoclonal antibody against 5-mC (Cat. No. MAb-006-100/500) and optimized PCR primer sets for qPCR of the indicated regions. One μ g of antibody was used per IP experiment. Figure 1 shows the recovery (expressed as a % of the input DNA) of a satellite repeat region, of the TSH2B gene which is known to be methylated and of the promoter of the active GAPDH gene, used as a negative control.

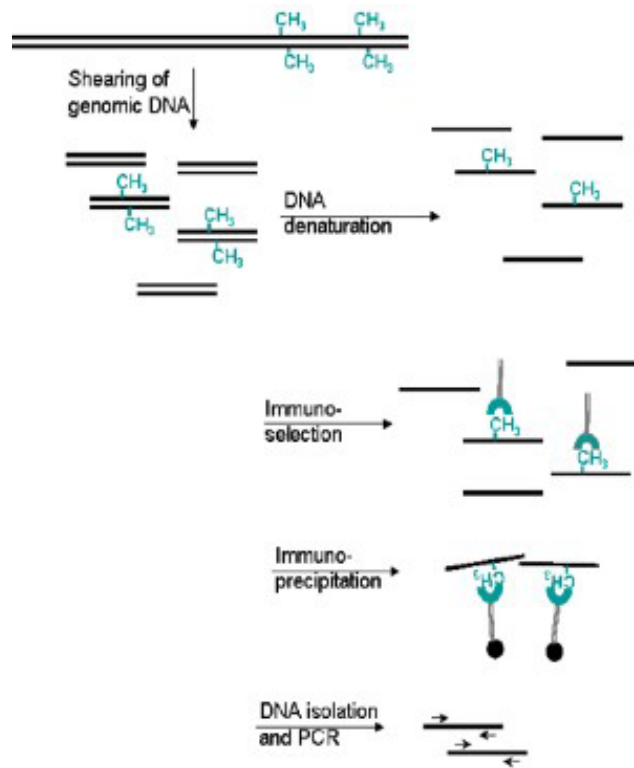


Figure 2
Methylated DNA immunoprecipitation (MeDIP) method

- Prepare genomic DNA from cultured cells
- Shear genomic DNA
- Denature the sheared genomic DNA
- Immunoprecipitate with the antibody against 5-mC
- Isolate DNA and perform PCR

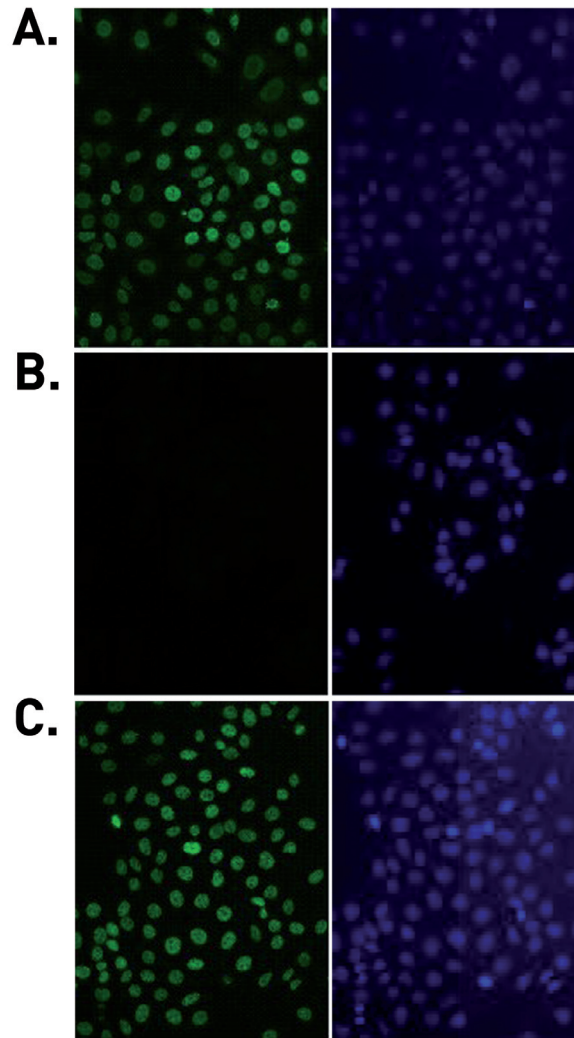


Figure 3

Immunofluorescence using the Diagenode monoclonal antibody directed against 5-mC

Human osteosarcoma (U2OS) cells were stained with the Diagenode monoclonal antibody against 5-mC (Cat. No. MAb-006-100/500) and with DAPI. Cells were fixed with 2.5% formaldehyde for 30', treated with 2N NaOH for 30' at 37°C and blocked with PBS containing 0.1% TritonX-100 and 1% BSA. Figure 3A: cells were immunofluorescently labeled with the 5-mC antibody (left) diluted 1:500 in blocking solution followed by an anti-mouse antibody conjugated to Alexa488 or with DAPI (right), which specifically labels DNA. Figure 4B and C: staining of the cells with the 5-mC antibody after incubation of the antibody with 50 μ M 5mCTP or 5hmCTP, respectively.