MGMT gene promoter methylation (O^6-methylguanine DNA methyltransferase)

For in vitro diagnostic use

CE IVD

ORDERING INFORMATIONS

REF: ONC-001-25
CND Code: W01060299
RDM Code: 2256631/R
Tests: 25 Reactions: 31 x 2
Manufacturer: BioMol Laboratories s.r.l.

CONTENTS OF THE KIT

*The kit consists of reagents for modification with sodium bisulfite and for amplification in MSP-PCR *reagents for the extraction of genomic DNA are not supplied in the kit.*

PRODUCT CHARACTERISTICS

Device belonging to the family of in vitro medical devices **REAL-TIME QUALITATIVE PCR-SOMATIC MUTATIONS.** Determination of the methylation status of the MGMT (O6-methylguanine DNA methyltransferase) gene promoter by MSP (methylation-specific PCR) technique and subsequent detection by Real-Time PCR technique. Kit optimized for Real-Time PCR instrumentation Biorad CFX96 Dx, Biorad Opus Dx and Agilent AriaDx.

SCIENTIFIC BACKGROUND

DNA O^6-methylguanine methyltransferase (MGMT) is a DNA repair enzyme encoded by the MGMT gene present at the 10q26 locus. The MGMT enzyme removes the alkyl groups from the O^6 position of guanine acting itself as an acceptor and this reaction leads to an irreversible inactivation of the enzyme. MGMT transcription is regulated by epigenetic mechanisms. Indeed, methylation of CpG dinucleotides in the promoter region of MGMT causes gene silencing, loss of MGMT expression and inability to remove alkyl groups from methylated guanine with consequent alteration of the normal DNA structure.

§ Cancer Sci. 2024 Oct;115(10):3394-3402. doi: 10.1111/cas.16297. Epub 2024 Jul 30. MGMT protein expression is a reliable predictive biomarker for temozolomide-containing chemotherapy in osteosarcoma

§ Cochrane Database Syst Rev. 2021 Mar 12;3(3):CD013316. doi: 10.1002/14651858.CD013316.pub2.

§ Prognostic value of test(s) for O6-methylguanine-DNA methyltransferase (MGMT) promoter methylation for predicting overall survival in people with glioblastoma treated with temozolomide

§ Genome-wide methylation profiling of glioblastoma cell-derived extracellular vesicle DNA allows tumor classification. Neuro Oncol 2021 Jul 1; 23 (7):1087-1099. doi: 10.1093/neuonc/noab012

§ MGMT methylation may benefit overall survival in patients with moderately vascularized glioblastomas. Eur Radiol 2021 Mar; 31(3):1738-1747. doi: 10.1007/s00330-020-07297-4. Epub 2020 Oct 1.

§ The significance of MGMT methylation in Glioblastoma Multiforme prognosis. J Pak Med Assoc 2018 Jul; 68(7):1137-1139.

§ Role of MGMT as biomarker in colorectal cancer. World J Clin Cases 2014 Dec 16; 2(12): 835–839.

§ Characterizing DNA methylation alterations from The Cancer Genome Atlas. J Clin Invest 2014 Jan 2; 124(1): 17–23.

§ Detection of aberrant promoter hypermethylation of tumor suppressor genes in serum DNA from non-small cell lung cancer patients. Cancer Res 1999 59: 67-70.

§ Methylation-specific PCR: a novel PCR assay for methylation status of CpG islands. Proc Natl Acad Sci U S A 1996 Sep 3; 93(18): 9821–9826.

CLINICAL SIGNIFICANCE

MGMT protects normal cells from carcinogens, but the activity of MGMT also protects cancer cells from the lethal effects of chemotherapy with alkylating agents such as dacarbazine (DTIC) or temozolomide (TMZ), which are widely used for the treatment of melanoma and glioblastoma. In fact, MGMT removes the methyl groups from the O^6 position of the guanines, thus making TMZ ineffective.

In glioblastomas, MGMT promoter methylation is predictive of the therapeutic benefit of the alkylating agent temozolomide, as shown in several phase III clinical trials, and MGMT gene methylation status has become the first predictive biomarker in neuro-oncology.

MGMT gene promoter methylation also plays an important role in colorectal carcinogenesis, occurring in approximately 30%-40% of metastatic colorectal cancer. Its prognostic role is not yet defined, but the loss of MGMT expression, which is secondary to gene promoter methylation, results in an interestingly high response to alkylating agents.

BIOMOL LABORATORIES S.R.L.
Via Arcora 110 (Palazzo Gecos)
80013 Casalnuovo di Napoli, NA
info@biomollaboratories.com
biomollaboratories.it

ISO 9001 :2015
ISO 13485:2021

ED.1 REV.0 of 01/10/2024

BioMol LABORATORIES

MGMT gene promoter methylation (O^6-methylguanine DNA methyltransferase)

For in vitro diagnostic use

CE IVD

ORDERING INFORMATIONS

REF: ONC-001-25
CND Code: W01060299
RDM Code: 2256631/R
Tests: 25 Reactions: 31 x 2
Manufacturer: BioMol Laboratories s.r.l.

CONTENTS OF THE KIT

*The kit consists of reagents for modification with sodium bisulfite and for amplification in MSP-PCR *reagents for the extraction of genomic DNA are not supplied in the kit.*

CONTENTS OF THE KIT

DESCRIPTION	LABEL	VOLUME	STORAGE
		ONC-001-25	
Conversion reagent	Conversion reagent	3 tubes	RT
Dilution buffer	Buffer A	900 µl	RT
Buffer	Buffer B	200 µl	RT
Binding Buffer	Buffer C	15 ml	RT
Wash buffer	Buffer D	3 ml	RT
Desulphonation Buffer	Buffer E	5 ml	RT
Elution Buffer	Buffer F	750 µl	RT
Columns	Columns	25	RT
Collection tubes	Collection tubes	25	RT
Mix oligonucleotides	Mix A methylated 10X	1 x 77,5 µl	-20°C
Mix oligonucleotides	Mix B unmethylated 10X	1 x 77,5 µl	-20°C
Mix buffer and Taq polymerase enzyme	Mix Real time PCR 2X	1 x 775 µl	-20°C
Deionized H_20	Deionized H_20	1 x 1 ml	-20°C
Genomic or recombinant DNA methylated and unmethylated	Control +	100 µl	-20°C

TECHNICAL CHARACTERISTICS

COD. ONC-001-25

STABILITY	18 months
REAGENTS STATUS	Ready to use
BIOLOGICAL MATRIX	Genomic DNA extracted from whole blood, tissue, cells
POSITIVE CONTROLS	Recombinant DNA for at least 4 analytical sessions
TECHNOLOGY	Real-time PCR; oligonucleotides; 1 SYBR-GREEN/FAM fluorescence channel
VALIDATED INSTRUMENTS	Biorad CFX96 Dx, Biorad Opus Dx and Agilent AriaDx
RUNNING TIME	150 min
THERMAL CYCLING PROFILE	1 cycle at 95 °C (15 min); 45 cycles 95 °C (40 sec) +60 °C (40 sec) + 72 °C (40 sec); 1 dissociation cycle at 70 °C with an increase of 0,2 °C
ANALYTICAL SPECIFICITY	Absence of non-specific pairings of oligonucleotides; absence of cross-reactivity
LIMIT OF DETECTION (LOD)	≥ 2.5 ng of sodium bisulfite modified DNA
LIMIT OF BLANK (LOB)	0% NCN
REPRODUCIBILITY	99,9%
DIAGNOSTIC SPECIFICITY / DIAGNOSTIC SENSITIVITY	100%/98%

BIOMOL LABORATORIES S.R.L.
Via Arcora 110 (Palazzo Gecos)
80013 Casalnuovo di Napoli, NA
info@biomollaboratories.com
biomollaboratories.it

ISO 9001 :2015
ISO 13485:2021

ED.1 REV.0 of 01/10/2024

BioMol LABORATORIES