

HLA-G 5' UTR-725 C/G POLYMORPHISM

ORDERING INFORMATIONS

REF: HLA-002-25
Tests: 25 Reactions: 31
Manufacturer: BioMol Laboratories s.r.l.

CONTENTS OF THE KIT

The kit consists of reagents for Real-Time PCR amplification
*the 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-GENETIC VARIANTS-RESEARCH USE ONLY**. Detection of 5' UTR-725 C/G polymorphism of the HLA-G gene (rs1233334) by Real-Time PCR technique. Kit optimized for Real-Time PCR instrumentation Biorad CFX96 Dx, Biorad Opus Dx and Agilent AriaDx. Thermofisher QuantStudio™ 5 Real-Time PCR System

SCIENTIFIC BACKGROUND

The HLA complex is a 3,6 Mb high-density gene region located at the 6p21.3 chromosome region, encompassing more than 200 genes. The HLA-G gene is a nonclassical class I HLA locus, which, like its classical counterparts, is composed of eight exons and seven introns. In contrast to classical class I loci, HLA-G has a stop codon at exon 6, leading to a short cytoplasmic tail, exhibits a 5' upstream regulatory (or promoter) region (5' URR) extending at least 1.4 kb and presents an extended 3' untranslated region (3' UTR). HLA-G is predominantly expressed at the maternal-fetal interface, particularly in the fetal extravillous cytotrophoblast cells, placental macrophages, and mesenchymal chronic villi and has primarily been associated with maternal-fetal tolerance.

§ HLA-G: Too Much or Too Little? Role in Cancer and Autoimmune Disease. *Front Immunol.* 2022 Jan 27;13:796054.

§ The genetic diversity within the 1.4 kb HLA-G 5' upstream regulatory region moderately impacts on cellular microenvironment responses. *SCIENTIFIC RePoRTS* (2018) 8:5652.

§ The impact of HLA-G, LILRB1 and LILRB2 gene polymorphisms on susceptibility to and severity of endometriosis. *Molecular Genetics and Genomics* (2018) 293:601–613 <https://doi.org/10.1007/s00438-017-1404-3>.

§ A Comprehensive Study of Polymorphic Sites along the HLA-G Gene: Implication for Gene Regulation and Evolution. *Mol. Biol. Evol.* 28(11):3069–3086. 2011 doi:10.1093/molbev/msr138

§ Variation in the HLA-G Promoter Region Influences Miscarriage Rates. *Am. J. Hum. Genet.* 72:1425–1435, 2003

§ Comprehensive analysis of HLA-G: implications for recurrent spontaneous abortion (2010) *Reprod Sci.* 17:331–338.

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CLINICAL SIGNIFICANCE

HLA-G is believed to protect the fetus against trophoblast damage caused by maternal natural killer (NK) and T-cytotoxic cells during pregnancy. Apart from pregnancy, HLA-G expression in non-pathological conditions is restricted, detected in the thymus, cornea, proximal nail matrix, pancreas, and hematopoietic stem cells. In pathological situations, HLA-G expression is observed in numerous tumors, viral infections, inflammatory and autoimmune diseases. HLA-G polymorphisms are associated with abnormal HLA-G levels and linked to reproductive disorders such as implantation failure, recurrent miscarriage, preeclampsia, and placental abruption. HLA-G -725 C>G polymorphism located at the 5' upstream regulator region (5'URR) or promoter region is reported to change the methylation profile of CpG dinucleotide, resulting in a modification of HLA-G expression and also linked to miscarriage. Both a deficiency expression and the overexpression of HLA-G can be harmful during pregnancy. If the polymorphism HLA-G -725 C>G is present in both parents, it may represent a risk factor for recurrent miscarriages, although it is not the main cause of the loss of the pregnancy itself.

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DESCRIPTION	LABEL	VOLUME	STORAGE
		HLA-002-25	
Mix oligonucleotides and probes	Mix HLA-G -725 C>G 10 X	1 x 77,5 µl	-20°C
Mix buffer and Taq polymerase enzyme	Mix Real-Time PCR 5X	1 x 155 µl	-20°C
Deionized H ₂ O	Deionized H ₂ O	1 x 1 ml	-20°C
Genomic DNA or recombinant DNA	Control 1	1 x 22 µl	-20°C
Genomic DNA or recombinant DNA	Control 2	1 x 22 µl	-20°C
Genomic DNA or recombinant DNA	Control 3	1 x 22 µl	-20°C

TECHNICAL CHARACTERISTICS

COD. HLA-002-25

STABILITY	18 months
REAGENTS STATUS	Ready to use
BIOLOGICAL MATRIX	Genomic DNA extracted from whole blood, tissues, cells
POSITIVE CONTROLS	Recombinant DNA for at least 3 analytical sessions
TECHNOLOGY	Real-time PCR; oligonucleotides and specific probes; 2 fluorescence channels HEX/JOE and FAM
VALIDATED INSTRUMENTS	Biorad CFX96 Dx, Biorad Opus Dx and Agilent AriaDx, Thermofisher QuantStudio™ 5 Real-Time PCR System
RUNNING TIME	85 min
THERMAL CYCLING PROFILE	1 cycle at 95 °C (10 min); 45 cycles at 95 °C (15 sec) + 60 °C (60 sec)
ANALYTICAL SPECIFICITY	Absence of non-specific pairings of oligonucleotides and probes; absence of cross-reactivity
LIMIT OF DETECTION (LOD)	≥ 5 ng of genomic DNA
LIMIT OF BLANK (LOB)	>40 Cq
REPRODUCIBILITY	99,9%
SPECIFICITY / SENSITIVITY	99,9%/98%