**Molecular Tech Fall 2017 Human Malate Dehydrogenase 2 Info**

**MDH2 Human [>sp|P40926|25-338].  Without Transit Sequence C term His tag plus TEV**

MAKVAVLGAS GGIGQPLSLL LKNSPLVSRL TLYDIAHTPG VAADLSHIET

KAAVKGYLGP EQLPDCLKGC DVVVIPAGVP RKPGMTRDDL FNTNATIVAT

LTAACAQHCP EAMICVIANP VNSTIPITAE VFKKHGVYNP NKIFGVTTLD

IVRANTFVAE LKGLDPARVN VPVIGGHAGK TIIPLISQCT PKVDFPQDQL

TALTGRIQEA GTEVVKAKAG AGSATLSMAY AGARFVFSLV DAMNGKEGVV

ECSFVKSQET ECTYFSTPLL LGKKGIEKNL GIGKVSSFEE KMISDAIPEL

KASIKKGEDF VKTLKENLYF QGHHHHHH

TEV cleavage site is shown in red

6xHis-tag is shown in blue

cDNA Sequence of MDH2 (Codon-optimized for expression in BL21 (DE3).

CCATGGCTAAAGTAGCTGTTCTGGGTGCATCTGGTGGTATTGGTCAACCGCTGTCCCTGCTGCTGAAGAACTCTCCACTGGTTAGCCGTCTGACCCTGTACGATATCGCACATACTCCAGGCGTTGCTGCTGACCTGTCCCACATTGAGACTAAAGCTGCGGTGAAAGGCTACCTGGGTCCAGAACAGCTGCCAGATTGTCTGAAAGGTTGTGACGTGGTAGTTATCCCAGCAGGTGTTCCACGTAAACCAGGTATGACCCGTGATGACCTGTTCAATACTAATGCGACTATCGTTGCGACCCTGACCGCTGCTTGCGCTCAGCACTGTCCGGAAGCTATGATCTGTGTGATTGCTAATCCGGTTAACAGCACCATCCCGATTACTGCCGAGGTATTCAAGAAACACGGTGTTTACAACCCGAACAAGATCTTCGGTGTTACCACCCTGGATATCGTGCGTGCAAACACCTTTGTGGCTGAACTGAAAGGTCTGGATCCAGCTCGCGTAAACGTTCCGGTAATCGGTGGTCACGCTGGCAAGACCATCATTCCGCTGATTTCCCAGTGCACTCCGAAAGTAGACTTCCCACAGGACCAGCTGACCGCTCTGACTGGTCGTATCCAGGAGGCAGGCACTGAAGTGGTGAAAGCGAAAGCAGGTGCTGGCTCTGCGACTCTGAGCATGGCCTACGCTGGTGCTCGTTTCGTATTCTCTCTGGTTGACGCAATGAACGGCAAGGAAGGTGTAGTGGAATGTTCCTTCGTGAAGTCTCAAGAAACCGAATGCACCTACTTCAGCACTCCACTGCTGCTGGGCAAGAAAGGCATCGAGAAGAACCTGGGCATCGGTAAAGTTTCTTCTTTCGAAGAGAAGATGATCTCCGACGCAATTCCGGAACTGAAAGCGAGCATCAAGAAGGGTGAAGATTTCGTGAAGACCCTGAAAGAGAACCTGTACTTCCAGGGTCATCATCACCACCATCACTAACTCGAG

Map of hMDH2/AA25-338



Synthetic gene was cloned into NcoI/XhoIdigested **pET28a**.

Open reading frame orientation as illustrated. ***Not all unique restriction sites are shown in the map. Extra nucleotides or unique restriction sites may be found on both ends of your gene for subcloning purpose.***

Seq:

LOCUS GS61493-1 pET28a-hMDH2\_AA25-338 6219 bp ds-DNA circular SYN 25-Jun-2017

DEFINITION .

ACCESSION .

VERSION .

KEYWORDS GS61493-1 pET28a-hMDH2\_AA25-338

SOURCE synthetic DNA construct

 ORGANISM synthetic DNA construct

REFERENCE 1 (bases 1 to 6219)

 AUTHORS .

 TITLE Direct Submission

FEATURES Location/Qualifiers

 source 1..6219

 /organism="synthetic DNA construct"

 /mol\_type="other DNA"

 terminator 26..73

 /note="T7 terminator"

 /note="transcription terminator for bacteriophage T7 RNA

 polymerase"

 CDS complement(140..157)

 /codon\_start=1

 /product="6xHis affinity tag"

 /note="6xHis"

 /translation="HHHHHH"

 gene complement(158..1152)

 /note="hMDH2\_AA25-338"

 CDS complement(167..184)

 /codon\_start=1

 /product="6xHis affinity tag"

 /note="6xHis"

 /translation="HHHHHH"

 protein\_bind 1194..1218

 /bound\_moiety="lac repressor encoded by lacI"

 /note="lac operator"

 /note="The lac repressor binds to the lac operator to

 inhibit transcription in E. coli. This inhibition can be

 relieved by adding lactose or

 isopropyl-beta-D-thiogalactopyranoside (IPTG)."

 promoter complement(1219..1237)

 /note="T7 promoter"

 /note="promoter for bacteriophage T7 RNA polymerase"

 promoter 1546..1623

 /gene="lacI"

 /note="lacI promoter"

 /note="

 "

 CDS 1624..2706

 /codon\_start=1

 /gene="lacI"

 /product="lac repressor"

 /note="lacI"

 /note="The lac repressor binds to the lac operator to

 inhibit transcription in E. coli. This inhibition can be

 relieved by adding lactose or

ORIGIN

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 61 ggggttatgc tagttattgc tcagcggtgg cagcagccaa ctcagcttcc tttcgggctt

 121 tgttagcagc cggatctcag tggtggtggt ggtggtgctc gagttagtga tggtggtgat

 181 gatgaccctg gaagtacagg ttctctttca gggtcttcac gaaatcttca cccttcttga

 241 tgctcgcttt cagttccgga attgcgtcgg agatcatctt ctcttcgaaa gaagaaactt

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 421 tcattgcgtc aaccagagag aatacgaaac gagcaccagc gtaggccatg ctcagagtcg

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 601 gaatgatggt cttgccagcg tgaccaccga ttaccggaac gtttacgcga gctggatcca

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 2341 actgcgatgc tggttgccaa cgatcagatg gcgctgggcg caatgcgcgc cattaccgag

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