APB830Hu01 100µg Active Transglutaminase 2 (TGM2) Organism Species: *Homo sapiens* (Human) *Instruction manual*

FOR RESEARCH USE ONLY NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Prokaryotic expression. Host: E. coli Residues: Met1~Ala687 Tags: N-terminal His-tag **Purity:** >95% **Endotoxin Level:** <1.0EU per 1µg (determined by the LAL method). Buffer Formulation: PBS, pH7.4, containing 0.01% SKL, 5% Trehalose . Original Concentration: 200µg/mL Applications: Cell culture; Activity Assays. (May be suitable for use in other assays to be determined by the end user.) Predicted isoelectric point: 4.8 Predicted Molecular Mass: 78.6kDa Accurate Molecular Mass: 100kDa as determined by SDS-PAGE reducing conditions. Phenomenon explanation: The possible reasons that the actual band size differs from the predicted are as follows: 1. Splice variants: Alternative splicing may create different sized proteins from the same gene. 2. Relative charge: The composition of amino acids may affects the charge of the protein. 3. Post-translational modification: Phosphorylation, glycosylation, methylation etc.

4. Post-translation cleavage: Many proteins are synthesized as pro-proteins, and then cleaved to give the active form.

5. Polymerization of the target protein: Dimerization, multimerization etc.

[<u>USAGE</u>]

Reconstitute in 10mM PBS (pH7.4) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

[STORAGE AND STABILITY]

Storage: Avoid repeated freeze/thaw cycles.

Store at 2-8°C for one month.

Aliquot and store at -80°C for 12 months.

Stability Test: The thermal stability is described by the loss rate. The loss rate was determined by accelerated thermal degradation test, that is, incubate the protein at 37°C for 48h, and no obvious degradation and precipitation were observed. The loss rate is less than 5% within the expiration date under appropriate storage condition.

[<u>SEQUENCE</u>]

MAEELVLERC DLELETNGRD HHTADLCREK LVVRRGQPFW LTLHFEGRNY EASVDSLTFS VVTGPAPSQE AGTKARFPLR DAVEEGDWTA TVVDQQDCTL SLQLTTPANA PIGLYRLSLE ASTGYQGSSF VLGHFILLFN AWCPADAVYL DSEEERQEYV LTQQGFIYQG SAKFIKNIPW NFGQFEDGIL DICLILLDVN PKFLKNAGRD CSRRSSPVYV GRVVSGMVNC NDDQGVLLGR WDNNYGDGVS PMSWIGSVDI LRRWKNHGCQ RVKYGQCWVF AAVACTVLRC LGIPTRVVTN YNSAHDQNSN LLIEYFRNEF GEIQGDKSEM IWNFHCWVES WMTRPDLQPG YEGWQALDPT PQEKSEGTYC CGPVPVRAIK EGDLSTKYDA PFVFAEVNAD VVDWIQQDDG SVHKSINRSL IVGLKISTKS VGRDEREDIT HTYKYPEGSS EEREAFTRAN HLNKLAEKEE TGMAMRIRVG QSMNMGSDFD VFAHITNNTA EEYVCRLLLC ARTVSYNGIL GPECGTKYLL NLNLEPFSEK SVPLCILYEK YRDCLTESNL IKVRALLVEP VINSYLLAER DLYLENPEIK IRILGEPKQK RKLVAEVSLQ NPLPVALEGC TFTVEGAGLT EEQKTVEIPD PVEAGEEVKV RMDLLPLHMG LHKLVVNFES DKLKAVKGFR NVIIGPA

[ACTIVITY]

Transglutaminase 2 (TGM2), encoded by the TGM2 gene, is belongs to the family of transglutaminases that catalyze the posttranslational modification of proteins via calcium dependent cross-linking reactions. In addition to its function in protein

cross-linking, TGM2 is also capable of hydrolyzing both GTP and ATP and has intrinsic kinase activity. TGM2 has been implicated in a variety of human diseases including celiac disease, inclusion body myositis, atherosclerosis, and neurodegenerative diseases. The activity of recombinant human TGM2 is measured by its ability to cleave a synthetic peptide Benzyloxycarbonyl-Gln-Gly and NH2OH in the assay buffer 200 mM MES, 10 mM DTT, 10 mM CaCl2, 100 mM Hydroxylamine Hydrochloride, pH 6.0. The rhTGM2 is diluted to 12.5 ug/ml in assay buffer. Loading into a clear well plate 50 μ L of 12.5 ug/mL rhTGM2 and start the reaction by adding 50 μ L of 100 mM substrate, with a substrate blank containing 50 μ L assay buffer, 50 μ L substrate, and no rhTGM2. Incubated at 37 ° C for 2 hours and stop the reaction with 400 ul stop solution of 0.37 M FeCl3, 0.67 M HCl, 0.2 M Trichloroacetic Acid. Centrifuge at 2000 rpm for 2 minutes and then load 200 ul of the supernatant into a plate and read at 525 nm (absorbance) in endpoint mode. The specific activity of recombinant human TGM2 is > 800 pmol/min/µg.



| OD (525 nm) | L-glutamic acid gamma -monohydroxamate (product) mM |
|-------------|---|
| 1.1046 | 10 |
| 0.5857 | 5 |
| 0.2946 | 2.5 |
| 0.1535 | 1.25 |
| 0.0831 | 0.625 |
| 0.0436 | 0.3125 |
| 0.0208 | 0.15625 |
| 0.0108 | 0.078125 |

Figure 1. The standard curve of L-glutamic acid gamma -monohydroxamate

Specific Activity (pmol/min/µg) =

Adjusted Vmax * (OD/min) x Conversion Factor ** (pmol/OD)

amount of enzyme (ug)

*Adjusted for Substrate Blank

**Derived using calibration standard L-glutamic acid gamma -monohydroxamate

[IDENTIFICATION]



Figure 2. Gene Sequencing (extract)



Figure 3. SDS-PAGE

Sample: Active recombinant TGM2, Human

[IMPORTANT NOTE]

The kit is designed for research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.