

## *Division of Signal Transduction Therapy*

### **Standard Operation Procedure**

#### **Preparation of TRIAD1 [H158A]**

|                                                         |                             |
|---------------------------------------------------------|-----------------------------|
| <b><u>Enzyme description:-</u></b>                      | TRIAD1 [H158A]              |
| <b><u>Clone number:-</u></b>                            | DU20493                     |
| <b><u>Source:-</u></b>                                  | Recombinant                 |
| <b><u>Tag:-</u></b>                                     | cleaved from N-terminal GST |
| <b><u>Purification method:-</u></b>                     | GSH-Sepharose               |
| <b><u>Expression level:-</u></b>                        | < 0.1mg/L                   |
| <b><u>Calculated molecular mass:-</u></b>               |                             |
| Monoisotopic                                            | 58126 Da                    |
| Average Mass                                            | 58162 Da                    |
| [cysteines reduced, methionines have not been oxidised] |                             |
| <b><u>Theoretical pI:-</u></b>                          | 5.59                        |
| <b><u>Purity:-</u></b>                                  | 90%                         |
| <b><u>Enzyme storage buffer:-</u></b>                   |                             |
| 50 mM HEPES pH 7.5, 10% glycerol, 150mM NaCl, 1mM DTT   |                             |
| <b><u>Storage temperature:-</u></b>                     | -80°C                       |
| <b><u>Assay:-</u></b>                                   |                             |

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**Clone Data Sheet**

**Protein name TRIAD1 [H158A]**

|                                             |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                 |
|---------------------------------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b><u>Protein</u></b>                       | TRIAD1 [H158A]                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                  |
| <b><u>Synonyms</u></b>                      | ARIH2, Triad domain-containing protein 1 (Drosophila Ariadne homolog 2)                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                         |
| <b><u>Clone Number</u></b>                  | DU20493                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                         |
| <b><u>Species</u></b>                       | Human                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                           |
| <b><u>Accession Number</u></b>              | Protein: O59376                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                 |
| <b><u>Tags</u></b>                          | cleaved from N-terminal GST                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                     |
| Aminoacid sequence of the expressed protein | <b>GPLGMSVDMNSQGSDSNEEDYDPNCEEEEEEEEDDPGDIEDYYVG<br/>VASDVEQQGADAFDPEEYQFTCLTYKESEGALNEHMTSLASVLKVS<br/>HSVAKLILVNFHWQVSEILDYKSNQAQLLVEARVQPNPSKHVPTS<br/>HPPHCAVCMQFVRKENLLSLACQAQFCRSCWEQHCSVLVKDGVGV<br/>GVSCMAQDCPLRTPEDFVFP LLPNEELREKYRRYLFRDYVESHYQL<br/>QLCPGADCPMVI RVQEP RARRVQCNRCNEVFCFKCRQMYHAPTDC<br/>TIRKWLTKCADDSETANYISAHTKDCPKCNICIEKNGGCNHMQCSK<br/>CKHDFCWMCLGDWKTHGSEYYECSRYKENPDI V NQSQQAQAREALK<br/>KYL F YFERWENHNKSLQLEAQTYQRIHEKIQERVMNNLGTWIDWQY<br/>LQNAAKLLAKCRYTLQYTYPYAYMESGPRKKLF EYQQAQLEAEIE<br/>NLSWKVERADSYDRGDLENQMHI AEQRRRTLLKDFHDT</b> |
| Native sequence                             | in bold                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                         |
| Protease cleavage                           | Prescission protease (RV-3C)                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                    |
| Cloning sites                               | BamH1 Not1                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                      |

**DNA sequence of  
the expression  
cassette**

ATGTCCCCTATACTAGGTTATTGGAAAATTAAGGGCCTTGTGCAACCCACT  
CGACTTCTTTTGGAAATATCTTGAAGAAAAATATGAAGAGCATTGTATGAG  
CGCGATGAAGGTGATAAATGGCGAAACAAAAAGTTTGAATTGGGTTTGGAG  
TTTCCCAATCTTCTTATTATATTTGATGGTGATGTTAAATTAACACAGTCT  
ATGGCCATCATACTGTTATATAGCTGACAAGCACAACATGTTGGGTGGTTGT  
CCAAAAGAGCGTGCAGAGATTTCAATGCTTGAAGGAGCGGTTTTGGATATT  
AGATACGGTGTTCGAGAATTGCATATAGTAAAGACTTTGAACTCTCAAA  
GTTGATTTTCTTAGCAAGCTACCTGAAATGCTGAAAATGTTCGAAGATCGT  
TTATGTCATAAAACATATTTAAATGGTGATCATGTAACCCATCCTGACTTC  
ATGTTGTATGACGCTCTTGATGTTGTTTTTATACATGGACCCAATGTGCCTG  
GATGCGTTCCAAAATTAGTTTGTTTTTAAAAACGTATTGAAGCTATCCCA  
CAAATTGATAAGTACTTGAATCCAGCAAGTATATAGCATGGCCTTTGCAG  
GGCTGGCAAGCCACGTTTGGTGGTGGCGACCATCCTCCAAAATCGGATCTG  
GAAGTTCGTCCAGGGGCCCTGGGATCCATGTCAGTGGACATGAATAGC  
CAGGGTCTGACAGCAATGAAGAGGACTATGACCCAAATTGTGAGGAAGAG  
GAAGAAGAAGAAGAAGACGACCCGGGGACATAGAGGACTATTACGTGGGA  
GTAGCCAGCGATGTGGAGCAGCAGGGGGCTGATGCCTTTGATCCCGAGGAG  
TACCAGTTCACTTGCTTGACCTACAAGGAATCTGAGGGTGCCTCAATGAG  
CACATGACCAGCTTAGCTTCTGTCCTAAAGGTATCTCATTGCTGCTAAA  
CTTATATTAGTTAATTTCCACTGGCAAGTTTCCAGAGATATTGGACAGATAC  
AAGTCCAATTCTGCTCAACTGCTTGTGAGGCTCGAGTTCAGCCTAATCCA  
TCAAAACATGTTCCACATCCCATCCCCCTCACCCTGTGCAGTGTGTATG  
CAGTTTGTGCGAAAGGAAAACCTACTCTCTCTGGCCTGTCAGgCCAGTTT  
TGCCGCAGCTGCTGGGAGCAGCACTGCTCAGTTCTCGTCAAGGACGGCGTG  
GGCGTGGGAGTCTCTTGCATGGCTCAGGACTGTCCACTCCGTACACCAGAG  
GACTTTGTGTTTCCATTGCTTCCCAATGAAGAATTGAGAGAGAAATACAGG  
CGCTACCTCTTCCAGGACTATGTGGAGAGTCATTACCAGCTCCAGCTGTGC  
CCTGGTGCAGACTGCCCCATGGTTATTTCGGGTACAGGAGCCTAGAGCTCGC  
CGAGTACAGTGAATCGGTGCAACGAGGTCTTCTGTTTCAAGTGTGTCAG  
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TGTGCAGACGACTCTGAAACAGCCAACACTACATTAGTGCTCACACTAAAGAC  
TGTCCCAAGTGAACATCTGCATTGAGAAGAATGGAGGCTGCAATCACATG  
CAATGCTCCAAATGTAAACACGACTTCTGCTGGATGTGTCTAGGAGATTGG  
AAGACTCATGGCAGTGAATACTATGAGTGCAGTCGTTACAAGGAGAATCCT  
GACATCGTGAACCAGAGCCAACAAGCCCAGGCGAGGGAAGCCCTCAAGAAG  
TACTTATTCTACTTTGAGAGGTGGGAAAACCACAATAAAAAGCTTGCAGCTA  
GAGGCACAGACATACCAGCGGATTCACGAGAAGATTGAGGAGAGGGTTCATG  
AACAACTGCGGACATGGATCGACTGGCAGTACCTACAGAATGCTGCCAAG  
CTCTTGGCCAAGTGTGATAACCCCTGCAATACACCTACCCATATGCATAT  
TACATGGAGTCCGGACCCAGGAAGAAGCTGTTTGAATACCAGCAGGCTCAG  
CTGGAGGCTGAGATCGAAAACCTCTCATGGAAAGTGGAGCGTGCAGACAGC  
TATGACAGAGGGGACTTGGAGAACCAGATGCATATAGCGGAGCAGCGGAGG  
AGAACCCTGCTGAAAGATTTCCATGACACCTAAGCGGCCGC