**Figure S1**

**C**

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TgFRM3(1705)  LFDLFYHDEKELQLKAKKSSGGTGAGEGGGPKKKSGVLDAKATQNVEICLKKYKLSTVEEFKALAAQIDDPTTLAIDLNLAENITLYWPEPSACEALKAK
NcFRM3(1691)  LFDLFYHDEKELLLKAKKSVAASPGENGGPAKKNSGVLDAKASQNVEICLKKYKLSTVDDFKALAAQIDDPTTLAIDSNLADNITQYWPEPSAAEALKAK
EtFRM3        ------------------------------------VLDPKSSQNVEICLKRYKLASQEDFEQLAEQLLQGGQLAIEGPLAESLVLYWPEAEAAAAFSSK

TgFRM3        SPENALQLPLADQLYFTLLNQVALGSEKLNFFLQRRAVDEELESKREKLDQYCGAIELLAQTLESDAFKDILGLVVRLGNCVNRGSKEGFGFKLKDFVGQ
NcFRM3        TPEEVLHMPAADQLYFTLIKHVSLGSEKLNFFLQRRAVDEELESKREKLNKYCQAISHLDNILDSDAFKGILGLVVRLGNCVNRGSKEGFGFKLKDFVGQ
EtFRM3        TAAEAAAMPAADQLYFWLLKKVPLCCDRLKFHIDRLCVEEELAAKAAALGVYFGAVAALAAVVVSSSFRALLQAVLRLGNAVNRGKKEALGFKFKDFADQ

TgFRM3        LAACTSKDRSTNLFR---------------VIVQTVLEQNPSACDEMRALQDVDAAKAIDLKDLLQFGDIEKKLGAWAKQIGDRRSAFGGAADKMEKWIE
NcFRM3        LASCTSKDRSTNLFR---------------VLVQTVLEQNPAACEEMRVLQEVDAAKGTDLKDLLQFGDIEKKLSAWTKQIGDRRSAFGGAAGKMEKWIE
EtFRM3        LAACTSRDRSTNLFRQILLVRAVATFAARRLLVETVCEQNEFAIEELRGLQLVDAAKGMEVAELANLSSIESRLSFWKKNINAKKDLLGAAAEAMLKWVE

TgFRM3        SREARLCELKKKAEDAETLQVRLRGLMGLTKADCVWPEPLRWFGQFYSHFEAIAKEYRGAKDRDRQKAEREAKKKNDVLRRATTIGAPAISSLSSEKPMQ
NcFRM3        SREALLGELKKKAEDAEDLQLRLRGLMGLTKADCVWPEPLRWFGQFYSHFETTAKEYRGAKDRDRQKAEREAKKKNDVLRRASTIAATTIS-PTQDKPTV
EtFRM3        DTEGVVAGLREEIAANEQQQEQLRLLLGLSKTDCVWPQPLKWFGKFYTLFEQTAKELKEAKEKEAAKRERELKKQQQAATSPATAAAAARR---------

TgFRM3        GVPLRPAGLDPSLARLSSATQSRFPGLLSPTGGG---GGNFFSGVGASFHSS-PPLQRFNTYANH--SGGKTGGFRSPSPLSPGAAAKSVARVRRASEAG
NcFRM3        SVPFRPSVLDPALASLPSGTQSRFP-PLSPSGSGGASGGNFFSGVGAPFQSSSLPLQRYNTYANHGNSGKTQASFHPLSPLSPGAASKSGAKGRRAGEAG
EtFRM3        ------TGTLKTLPTRAAADALAIP-----------------------------------GG-------PTQPAAAATPAAAAAAAGGSKAPPVVS

TgFRM3        K----------S---------------AE-------------------GQE-----------------APAEAAS-------------------------
NcFRM3        KLTRVLEGSAERSSVAGDDAGPETPAAGENAAGEGKENNVGSDAQAGSGVAKTAGENAAGEGKENNVGSDAQAGSGVAKTAGENAAGEGKENNVGSDAQA
EtFRM3        R-------------------------------------------------------------GSSAALS--------------------------

TgFRM3        -SGLAEKAGE------------EEKQCG-----------------------AEAGD----------DGEADEN---------------------------
NcFRM3        GSGVAKTAGENAAGEGKENNVGSDAQAGSGVAKTAGENAAGEGKENNVGSDAQAGSGVAKTAGENAAGEGKENNVGSDAQAGSGVAKTAGENAAGEGKEN
EtFRM3        -PAPAAAAGD-------------------------------------------------------EAAA---------------------------

TgFRM3        ------------------------------------------------------------ASGGQEDGAAAAPSLSPETDEGLRVEPES---
NcFRM3        NVGSDAQAGSGVAKTAGENAAGEGKENNVGSDAQAGSGVAKTAGENAAGEGKENNVGSDAQAGSGVAKTAGGNAAGEGKENNVGSDAQAGSGVAKTAGEN
EtFRM3        ------------------------------------------------------------AAAAAP----TVPAVAAAGDTPPPTP---

TgFRM3        ------------------------------------------------LLERADQREREDEGKQVDAEEDAANLMASVDEEGRGLAPRIMAAWA
NcFRM3        AAGEGKENNVGSDAQAGSGVAKTAGENAAGEGEADGTSARNSGLETGEDQAEDGPLERTDARQSEETRTQEEPEEDAANLMASVDEEGRGLAPRIMAAWA
EtFRM3        ----------------------------------------------------------------------------------------------------

TgFRM3        QARSPHLGRYQGCKRGGRGLGRFPGILRDKSKKISCDGADSSLLVPSLYQTRTAVGASGNSPLPSPRPSPTAATNASFTVRSLRDPSPLPVASGVSTLPT
NcFRM3        QARSPHLGRYQGRKRGGRGLVNFPAFLRDKTKKTAREGAHSSHSTPSPCEARAAGAAAGGTPLPSCPAAPAAGT------------APLAFHS-------
EtFRM3        -------------------SSTP--------------TST--PASTPAAAAAAAAAAASAPDLSSPSVSDTST--------------------------

TgFRM3        SSGVTTVPVPSDVSTSAEFMFPFAKA-RPKTGSRGRAR------CVEKHEQKSEDVHKVVGGSSAASFLVSAASSLGLEKTKRREDKTETVQKARAGDKA
NcFRM3        VLGVSSLPAPSGCSSPTQFMFPFSKVPRPTAPSDRRARSVPGGESAEPREHTSEDVGKTLECASAASLFVSASPSLGPEK-KGRDEKAEVVQKGRG-DKA
EtFRM3        ---MSPLPSAANLYEMQQQVCSSSEVSEDPTGPKASHMP----------QQQQHQLQQQQD----Q-----QQQQQEQQQQQQQQQQQEQQQQQQQ-QQQ

TgFRM3        TAVVPRNKAALCVSPVPTTSPHG-CSRPFTRSTRVSGGELSEPSGSVPCLSEADKEWVRCGGGYCEVS---LSSTAGPSGAR-QQRDS-RGSTAASHSFS
NcFRM3        TAAVPRSKAALPSSPAPANTPRCQGPRHPLRSVRAAAGEPS-TSGPVPFSPELDKEWVRCGGGYCRQSDSSLGAGAASSAARGQHREVGRGPAASSHSSS
EtFRM3        QQQRPPATLAWLFARTPGRRPHGLLPLQGLRPAAAPAAAPAAVAATGPAAAATTPRRVMYRALTSPGR-----AGPRAAATLAAARAAAAAAAAAADSSD

TgFRM3        --LSGAGSSRASSGRQGSVGAEAKDGSEGLTNSTRVGRRRSQGAIGAEETEKGKTGVEKGLKMLGKGLSAGKFQSTPEHQAPPPLED-----------LA
NcFRM3        ALVFGAGGSRASLGRQAPAGADEAHRPEALPDSAKLARRRSH---GAEETETGKAVLGKVSKMSGKGFSAGNFQSAVDHQASRPLREPVTVSERDFSEAG
EtFRM3        -----ESEGAAPLG---SPGVEGRAQQPLLFSSDCLSSSSSS--------------SKSCSKMN----SNSSSRSSSRNGMSKDSLG-----------RS

TgFRM3        FARDSLETPAEAGLDALVREVKAVDLARKRLEAARNRDGETDVSARDRKESHPCPFSPQAGRSGLDIGGKDFDRSMVGDSVRKVRTRVTGGRDGSIERQA
NcFRM3        HQRESNKASGEAELEGMAKEAGAVDQARKRMEATR--DGETGSSALERKECHTSRFC-DGGRGGAEGPGKDFERSAAGDSVRKVRTRVTGGREGSIERQA
EtFRM3        WPQDDPARAAAAAKTAQASAAAATDAAAAASAAAS----AAAAAAKKQPCLDPATFTVIGDAEAAAAAAAPTAAAANESPPQQHSGDCSSGSESDCSSEG

TgFRM3        TNPYIAANVPRHPRRWEPGGRMTQGRKVPADLSALGSDLNPPSAHGSGSPSEAAISDGGEVARQTKGGGESADAAV----------
NcFRM3        TNPYIAANVPRHPRRCDPGGRGTLVRNVLVDFPSSGSGISSQSANGSG-HCESEPSDAGEVAGRARSGCEPRVAAVRENLGDPERS
EtFRM3        E-PRVAAARAAAAAAAAAAAAETAARGAAATPAAAAAAADGAHRGSSS----------SRASSKAREPSYPYIAAV----------
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**Supplementary figure S1. Alignment of the FH2 domains of *Toxoplasma gondii* formins**

(A) Schematic representation of *Toxoplasma gondii* formin 3 (TgFRM3) showing the formin homology 2 domain (FH2). Expanded protein sequence above protein schematic represents the potential FH1 domain. The proline residues (P) are in bold. Numbers indicate amino acid residues bordering the different domains. Scale bar represents 500 amino acids.

(B) The FH2 domains of TgFRM1, TgFRM2 and TgFMR3 (F1, F2 and F3) have been aligned using CLUSTAL multiple alignment (http://npsa-pbil.ibcp.fr/cgi-bin/npsa_automat.pl?page=npsa_clustalw.html). The asterisks indicate the W residue, which is conserved among all formin proteins, and the two residues I and R or K that have been shown to be critical for actin binding and are conserved in all described formins. The residues W and R or K are absent in the protein sequence of the F3. Only the isoleucine residue (I) is conserved. The weakly similar residues are shown in blue, strongly similar residues are shown in green, and identical residues are in red. The amino acid numbers for the initial residues of the expressed recombinant proteins are shown in parentheses on the left.

(C) The TgFRM3 has been aligned with the available FRM3 protein sequence from *Neospora caninum* (NcFRM3) and *Eimeria tenella* (EtFRM3) using Clustal multiple alignments. The conserved residues are shown in blue, similar residues are shown in green, and identical residues are in red.