Supplemental Materials Molecular Biology of the Cell

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Supplemental Figure Legends

Supplemental Figure 1. Sequence alignment of Fam13a across species. Residues of the 14-3-3 binding motifs and nuclear localization signals are highlighted in blue and red boxes, respectively. The sequence of the B56-binding domain is underlined in green.

Supplemental Figure 2. Leptomycin B (LMB) blocks OA-induced cytoplasmic sequestration of Fam13a. Representative immunofluorescence images showing subcellular distribution of Fam13a in control (DMSO), LMB-, OA-, and OA+LMB-treated NIH3T3 cells. Fam13a was primarily nuclear in control (DMSO) and LMB-treated NIH3T3 cells. OA treatment induced cytoplasmic sequestration of Fam13a. LMB treatment blocked OA-induced cytoplasmic sequestration of Fam13a. The graph on the right shows the percentage of cells displaying nuclear (N), cytoplasmic (C), or even distribution between nucleus and cytoplasm (N=C).

Supplemental Figure 3. Specificity of the anti-Fam13a antibody. A. Western blot showing that overexpressed myc-Fam13a could be recognized by the anti-myc and anti-Fam13a antibodies on Western blot. **B**. Western blot showing detection of endogenous Fam13a in A549 cells by the anti-Fam13a antibody. Note that transfection of a siRNA against FAM13A (siFam13a) reduced the amount of endogenous Fam13a in A549 cells.

Supplemental Figure 4. The motif between residues 404 and 418 of Fam13a is a 14-3-3 binding motif. The left panel is a CoIP experiment, showing that mutation of the 14-3-3 binding domain located between 404-418 (Δ 315-329; T411A) abolished the weak binding between Δ 315-329 and 14-3-3. The right panel is a GST-pull down assay, showing that bacterially expressed GST-14-3-3 η could pull down the wild type Fam13a, a small amount of Δ 315-329, but not Δ 315-329; T411A.

mouse rat human chicken xenopus zebrafish	284	PMLSPRFYAYGQSRQYLDDTEVPPSPPNSHSFM-RRRSSSLGSYDDEQ-EDLTPVQLTRR PMLSPRFYAYGQSRQYLDDTEVPPSPPNSHSFM-RRRSSSLGSYDDEQ-EDLTPVQLTRR PMLSPRFYAYGQSRQYLDDTEVPPSPPNSHSFM-RRRSSSLGSYDDEQ-EDLTPAQLTRR PMLSPRFYAYGQSQQYLDDTEVPPSPPNSHSFM-RRRSSSLGSYEDDR-EDLTPAQLTRR PMLSPRFYAYGHSQQYLDDFEVPPSPPNAHSFM-RRRSSSLGSHEEDRDEDLTPAQLTRR PMLSPRFYAYGHSQQYLDDFEVPPSPPNAHSFM-RRRSSSLGSCEDER-EELTSAQLSKR	41
mouse rat human chicken xenopus zebrafish	342	IQTLKKKIRKFEDRFEEERKYRPSHSDKAANPEVLKWTNDLAKFRKQLKESKLKISEEDL IQTLKKKIRKFEDRFEEERKYRPSHSDKAANPEVLKWTNDLAKFRKQLKETKLKISEEDL IQSLKKKIRKFEDRFEEEKKYRPSHSDKAANPEVLKWTNDLAKFRRQLKESKLKISEEDL IQGLKKKIRKFEDKFEERKYRPSHSDKAANPEVLKWTNDLAKFRKQLKESKLKISEEDL IQSLKKKIRKFEDKFEEDRKYRPSHRDKATNPEVLKWMNELAKFRKQLKETKIKLSEDDL IHVLKKKIRRYEEKFEERKYRPSHGDKAGNPEVLRWMNELTRLRKDLKDHKLLKSEEDL	D1
mouse rat human chicken xenopus zebrafish	402	TPRTRQRSNTLPKSFGS2LEKEDEKKQELLDKAIRPSVEATLEGILRKLQEKRVESSRPE40TPRTRQRSNTLPKSFGS2LEKEDEKKQELVDKAIRPSVEATLEGILRKLQEKRMESSRPE40TPRMRQRSNTLPKSFGS2LEKEDEKKQELVDKAIKPSVEATLESIQRKLQEKRAESSRPE40GPVVRQRSNTLPKSFGS2LEKEDEKKQELVDKAIKPSVEATLESIQKKLQEKRTETNRPE40VPQMRQRSNTLPKSFGS2LEKEDEKKPETVEKPAKPSMEATMEAIQKKLQEKRNEVNRPETPIPRQRSNTLPKSFGS2LEKKTPETKAPKPPVESTLETVTNKLQEKRKEAGRPE	61
mouse rat human chicken xenopus zebrafish		DIKDMTKDQIANEKVALQKALLYYESIHGRPVTKTERQIMKPLYDRYRLVKQILSRASTV DIKDMTKDQIANEKVALQKALLYYESIHGRPVTKTERQIMKPLYDRYRLVKQILSRASTV DIKDMTKDQIANEKVALQKALLYYESIHGRPVTKNERQVMKPLYDRYRLVKQILSRANTI DIKDMTRDQIAAEKVALQKALLYYESIHGRPVTKNERQVMKPLYDRYRLVKQILSRANTI DIKDMTRDQIAAEKVSLQKALLYYESIHGRPVTKNERQVMKPLYDRYRLFKQILSRANTI DIKDMTRDQIAAEKVSLQKALLYYESIHGRPITKNERQVMKPLYDRYRLFKQILSRANTI DIKDMTREQIGAEKVAIQKALLYYESIHGRPITKNERQVMKPLYDRYRLVKQILCRASAI	21
mouse rat human chicken xenopus zebrafish	522	PIIGSPSSKRRSPSLQPIIEGETASFFKEIKEQEEGSEDDSSTKPDFAVTLKTDCS PIIGSPSSKRRSPSLQPIIEGETASFFKEIKEQEEGSEDDSSTKPDFTVTLKTDSS PIIGSPSSKRRSPLLQPIIEGETASFFKEIKEEEEGSEDDSNVKPDFMVTLKTDFS PIIGSPSSKRRSPLLQPIIEGETASFFKEIKEEEEGSEEDSNVKPDFTITMKTDFN PVIGSPSSKRRSPLLQPIIEGETASFFKEVKETLPEEEEGSEDDTNGNTDFTVTMKPGFS PVIGSPSSKRRSPLLQPIIEGVPALFFSDTKEEEDGSDDD-DTRTQFTVTVKPELS	77
mouse rat human chicken xenopus zebrafish	578	AHCFLDQLEDDADGFISPMDDKMPSKCSQDSGLSNLHSASIPELLEYLQEMREEKKMIRK AHCFLDQLEDDADGFISPMDDKMPSKCSQDSGLSNLHSASIPELLEYLQEIREEKKMIRK ARCFLDQFEDDADGFISPMDDKIPSKCSQDTGLSNLHAASIPELLEHLQEMREEKKRIRK VRSFLDQLEDDADGFVSPVDDKIPSRSNQDMGLSNLHEASIPELLEQLQEVREEKKRIRK VRTFLEQLDDDADGFVSPVDDNIPSKSSQDLGLGNLHSASIPELLEQLQEAREEKKRIRK MLGLLDQLDEDADGFISPVDELSPSKNTTDMRLSNLHAATMQELVEELQEAREEKKRIRK	37





