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online data

SUPPLEMENTARY ONLINE DATA

Docking interactions of the JNK scaffold protein WDR62

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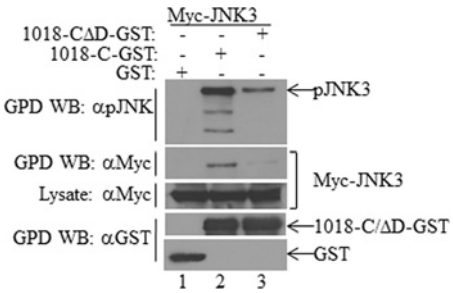

Figure S1 A putative D domain within WDR62 is responsible for WDR62–JNK3 association

HEK-293T cells were transfected with plasmids encoding GST, 1018-C–GST or 1018-CΔD–GST together with HA–JNK3 as indicated. GST-containing complexes were isolated from cell lysates using glutathione–agarose beads, washed extensively and eluted with reduced glutathione. The protein complexes were subjected to Western blotting (WB) with anti-pJNK (top panel), anti-Myc (second and third panels from top) or anti-GST (bottom two panels) antibodies. The expression level of Myc–JNK3 was determined by blotting the total cell lysate with anti-Myc antibody (third panel from top). The migration of the relevant proteins is indicated by arrows.

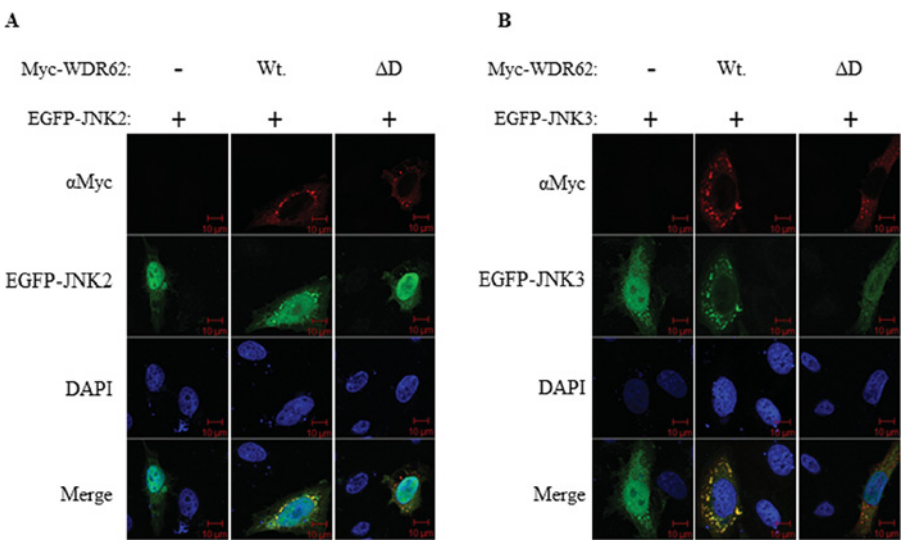

Figure S2 WDR62 recruits JNK2 and JNK3 to cellular granules through its putative consensus D domain

HeLa cells were co-transfected with EGFP–JNK2 (**A**) or EGFP–JNK3 (**B**) expression plasmids together with plasmids encoding either wild-type WDR62 (Wt.) or the D domain deletion mutant (ΔD). Cells were fixed and stained with anti-Myc antibody (red). EGFP–JNK2/3 is shown in green. Co-localization appears in yellow in the merged image. Nuclei were stained with DAPI (blue). Representative confocal microscopy images are shown. Scale bar, 10 μm.

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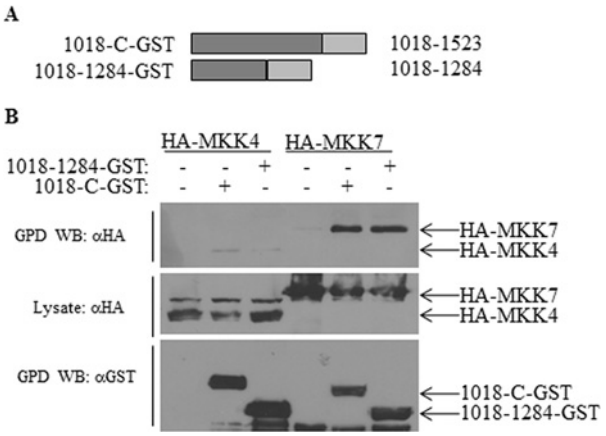

Figure S3 Both MKK7 and MKK4 associate with WDR62

HEK-293T cells were transfected with a plasmid encoding 1018-C–GST or 1018-1284-C–GST together with HA–MKK7 or HA–MKK4 as indicated. An empty GST plasmid was used as a negative control. GST-containing complexes were isolated from cell lysates using glutathione–agarose beads, washed extensively and eluted with reduced glutathione. The protein complexes were subjected to Western blotting (WB) with either anti-HA or anti-GST antibody (top and bottom panels respectively). The expression level of HA–MKK was determined by blotting the total cell lysate with anti-HA antibody (middle panel). The migration of the relevant proteins is indicated by arrows.

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