Supplemental Information (Supporting Online Material).

Supplemental Figure Legends

Figure S1. Juxtanuclear inclusions generated by various disease-associated proteins are positive for ubiquitin and vimentin (A) Representative confocal images of SH-SY5Y cells ectopically expressing GFP-tagged huntingtin mutant (Q150Htt), α-synuclein and myc-tagged synphilin-1, myc-tagged tau P301L mutant, AIMP2 (p38) or desmin mutant immunostained with relevant primary protein-directed antibodies or anti-vimentin. Arrows in merge pictures show the co-localization of inclusions generated by the various aggregation-prone proteins with or vimentin (yellow), as indicated. (Scale Bar, 10 μm). (B) Bar graph showing the percentage of transfected cells containing aggresome-like inclusions. Each of these experiments was repeated at least three times.

Figure S2. Clearance of inclusions under normal or serum-starved culture conditions (A) Representative confocal images of SH-SY5Y cells cultured under normal or serum starved conditions (as indicated) and visualized with lysotracker (*top panels*) or anti-LAMP1 immunostaining (*bottom panels*). Untreated control cells are shown in leftmost panels. (Scale Bar, 10 μm). (B) Graph showing the number of inclusions over a 24 h recovery period under conditions of normal serum (normal) or serum starvation (starved) (*P <0.05, **P < 0.01, ***P < 0.001). Each of these experiments was repeated at least three times.

Figure S3. Intact microtubule network in aggresome-positive cells. Representative confocal images of SH-SY5Y cells ectopically expressing various aggregation-prone

proteins (as indicated) immunostained with relevant primary protein-directed antibodies (red) or anti-α-tubulin (green). Arrows in merge pictures show the absence of co-localization of inclusions generated by the various aggregation-prone proteins with α-tubulin (yellow). Note that the microtubular network is not disrupted in these aggresome-positive cells. Each of these experiments was repeated at least three times. (Scale Bar, 10 μm).

Figure S4. Presence of α-synuclein/synphilin-1 in AIMP2 (p38)-positive inclusion promotes its recruitment of LAMP1 and mTOR. (A & B) Representative confocal images of SH-SY5Y cells containing AIMP2 (p38)-positive inclusions in the absence (AIMP2 (p38) only) or presence of α-synuclein/synphilin-1 co-localization (AIMP2 (p38) + Syn/Synph) immunostained with anti-α-synuclein or (A) anti-LAMP1 or (B) anti-mTOR, as indicated. Arrows in merge pictures show the co-localization of (AIMP2 (p38) + Syn/Synph)-positive, but not AIMP2 (p38) only, inclusions with (A) LAMP1 and (B) mTOR. α-Synuclein and AIMP2 (p38) were stained blue and green with AlexaFluor647 and 488 respectively. LAMP1 and mTOR were visualized in red by staining with AlexaFluor555. Each of these experiments was carried out in cells cultured under normal serum conditions but in the presence of proteasome inhibition and was repeated at least three times. (Scale Bar, 10 μm).

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Figure S5. Clearance of AIMP2 (p38)- and α-synuclein-positive inclusions. (A) Bar graph showing the relative fold difference in the number of AIMP2 (p38)-positive inclusions in the absence (AIMP2 (p38) only) or presence of α-synuclein (AIMP2 (p38) + α-synuclein) (*left*) or synphilin-1 co-localization (AIMP2 (p38) + synphilin) (*right*) under normal or serum-starved conditions, as indicated (B) Anti-α-synuclein immunoblot showing the relative expression level of endogenous and exogenously-introduced α-synuclein in SH-SY5Y cells. (C) Representative confocal images of SH-SY5Y cells containing AIMP2 (p38)-positive inclusions in the absence (AIMP2 (p38) only) or presence of endogenous α-synuclein co-localization (AIMP2 (p38) + endogenous α-synuclein) immunostained with anti-α-synuclein or AIMP2 (p38) (Scale Bar, 10 μm). (D) Bar graph showing the relative percentage (*left*) or fold difference (*right*) in the number of AIMP2 (p38)-positive inclusions in the absence (AIMP2 (p38) only) or presence of endogenous α-synuclein co-localization (AIMP2 (p38) + endogenous α-synuclein) under normal or serum-starved conditions, as indicated. Each of these experiments was repeated three times. (**P* <0.05, ***P* < 0.01, ****P* < 0.001).

Figure S6. Co-localization of AIMP2 (p38)- or desmin-positive inclusion with various autophagy-susceptible inclusions (A) Representative confocal images of SH-SY5Y cells containing AIMP2 (p38)-positive inclusions in the absence (AIMP2 (p38) only) or presence of tau-positive inclusions (AIMP2 (p38) + tau P301L) immunostained with anti-α-synuclein or anti-myc (tau P301L). No co-localization between tau-positive and AIMP2 (p38)-positive inclusions is apparent in the image shown (B) (*Left*) Representative confocal images of SH-SY5Y cells containing desmin-positive inclusions

in the absence (Desmin only) or presence of tau P301L co-localization (Desmin + Syn/Synph) immunostained with anti-desmin or anti-α-synuclein, as indicated. Arrows in merge pictures show the co-localization between desmin- and α-synuclein/synphilin-1-positive inclusions (*Right*) Bar graph showing the relative fold difference in the number of desmin-positive inclusions in the absence (Desmin only) or presence of α-synuclein/synphilin-1 co-localization (Desmin + Syn/Synph) under normal or serum-starved conditions. (C) Same as (B) except that mutant tau replaced α-synuclein/synphilin-1. Each of these experiments was repeated at least three times. (Scale Bar, 10 μm)

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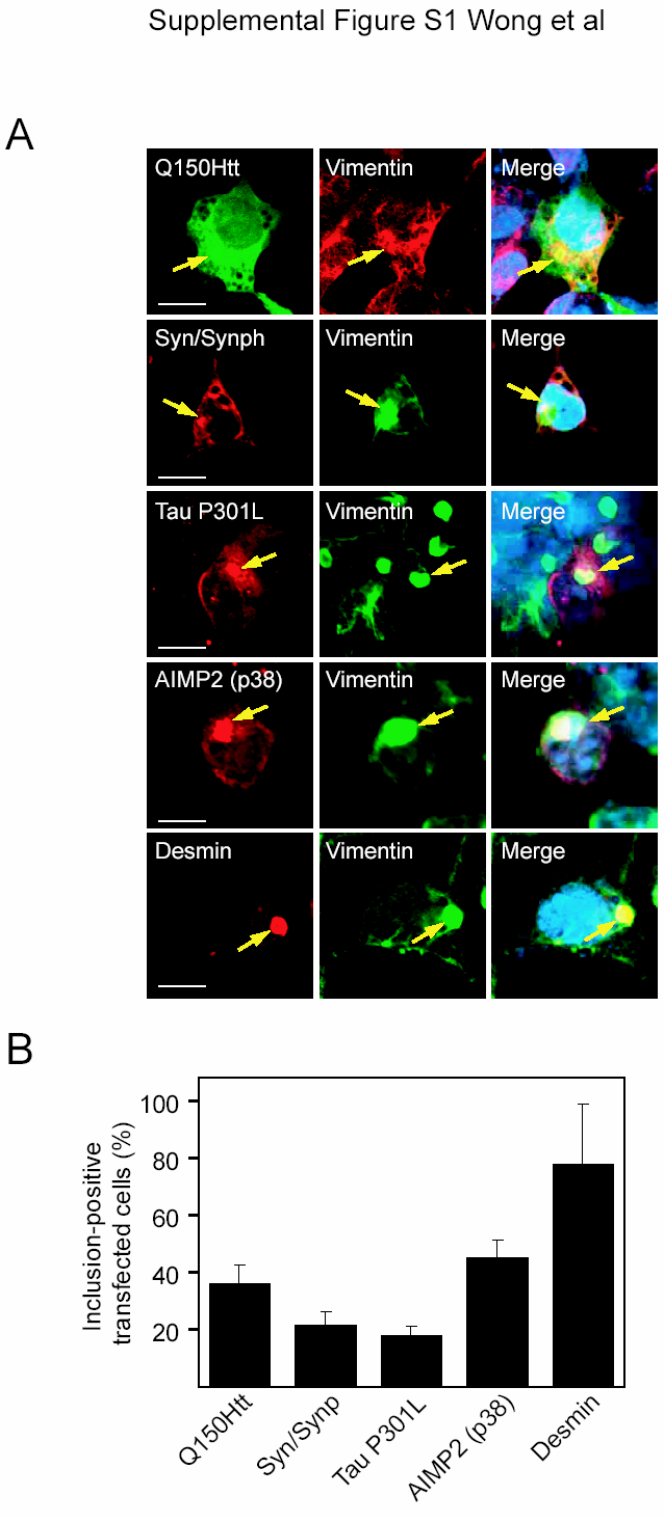

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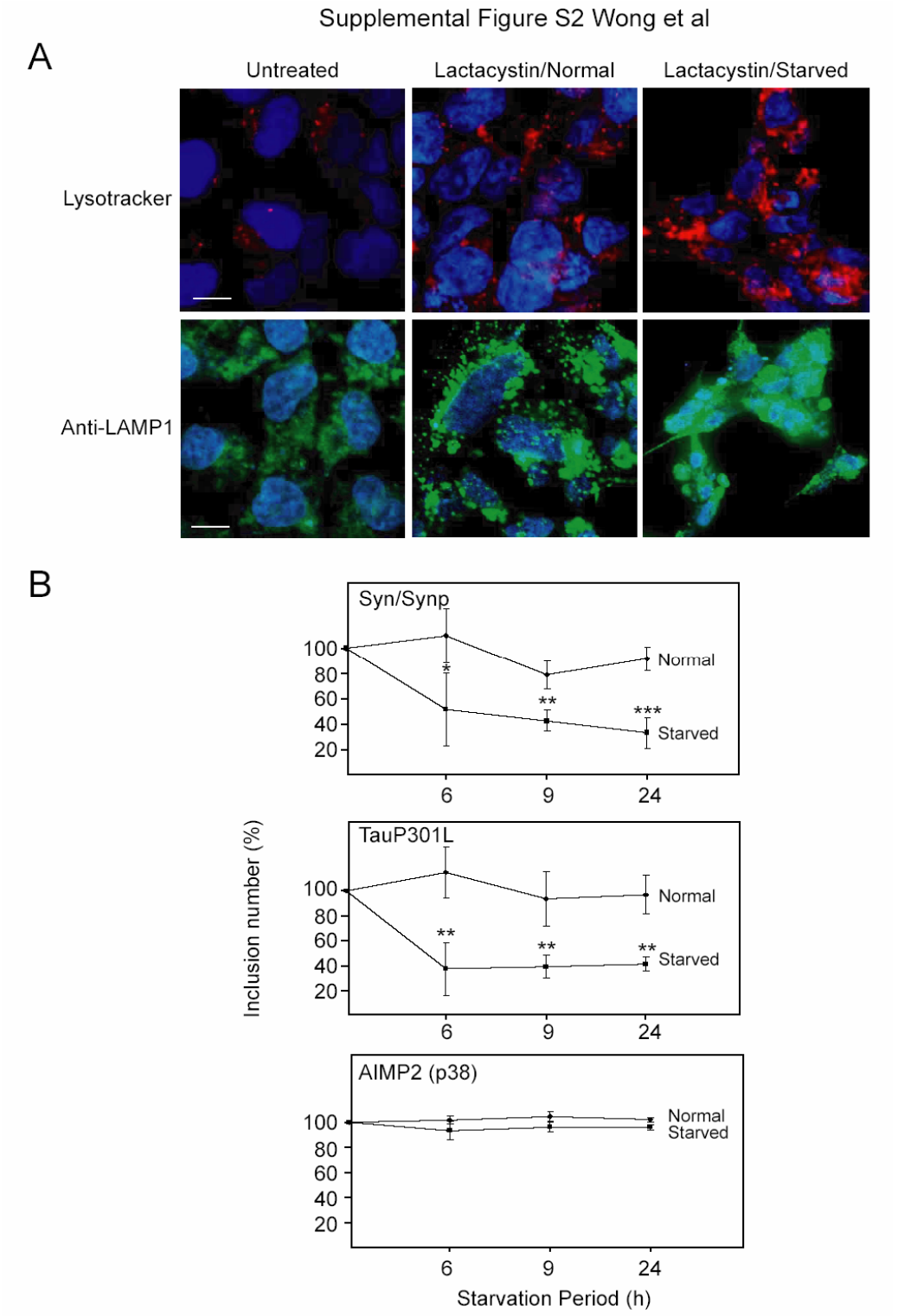

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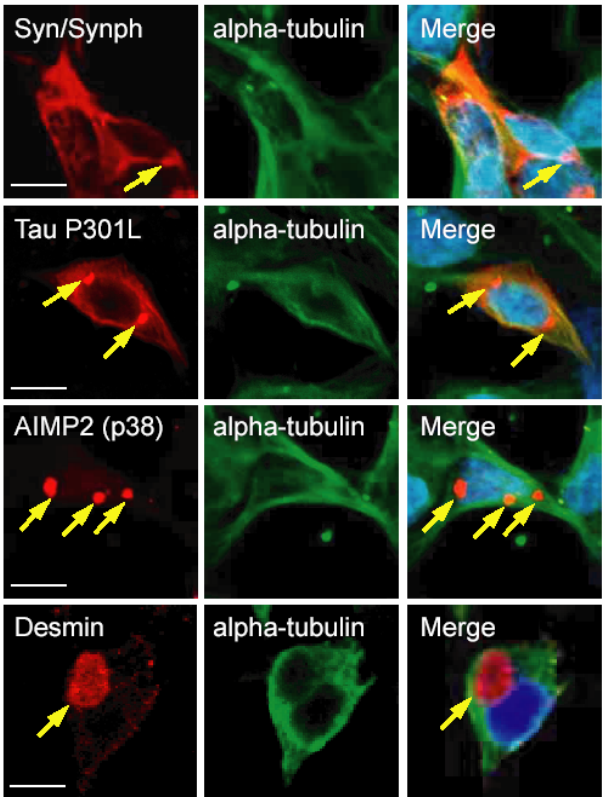

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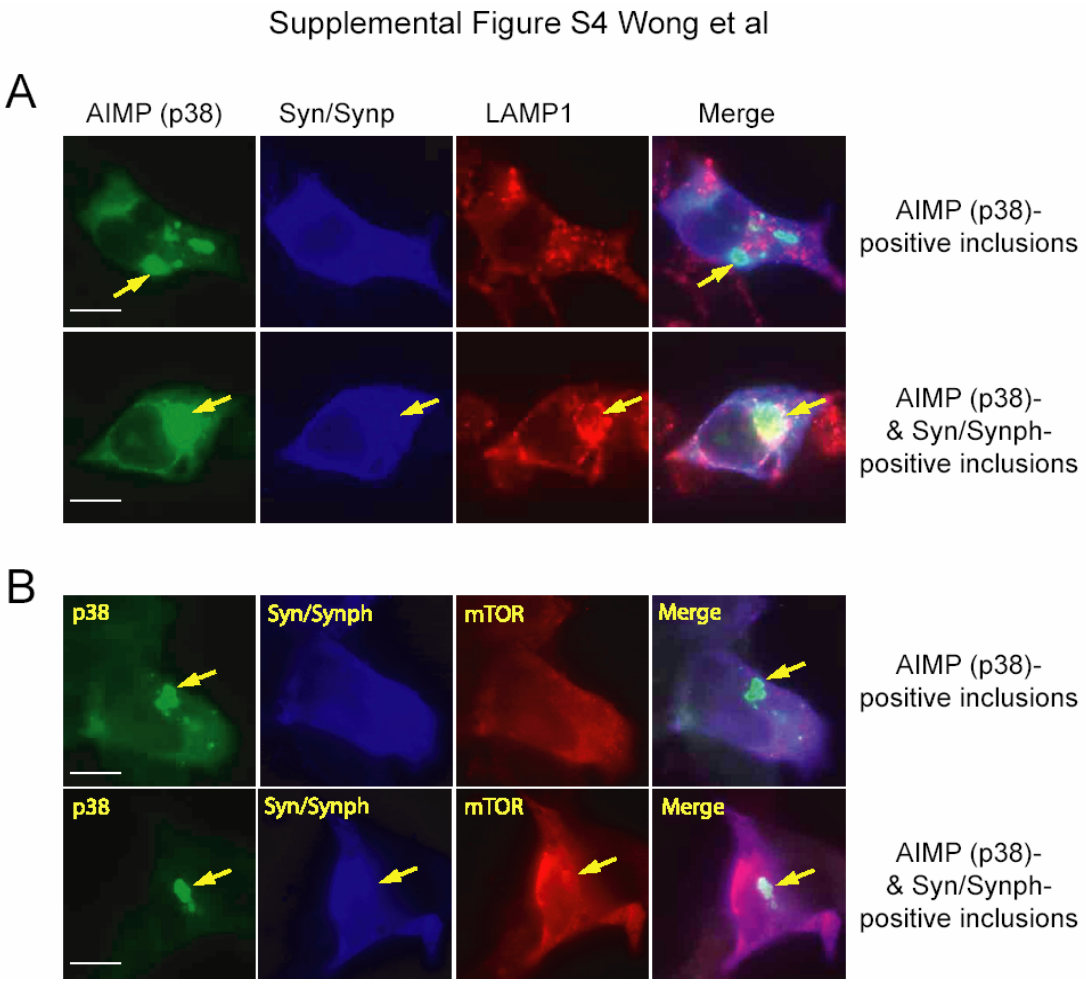

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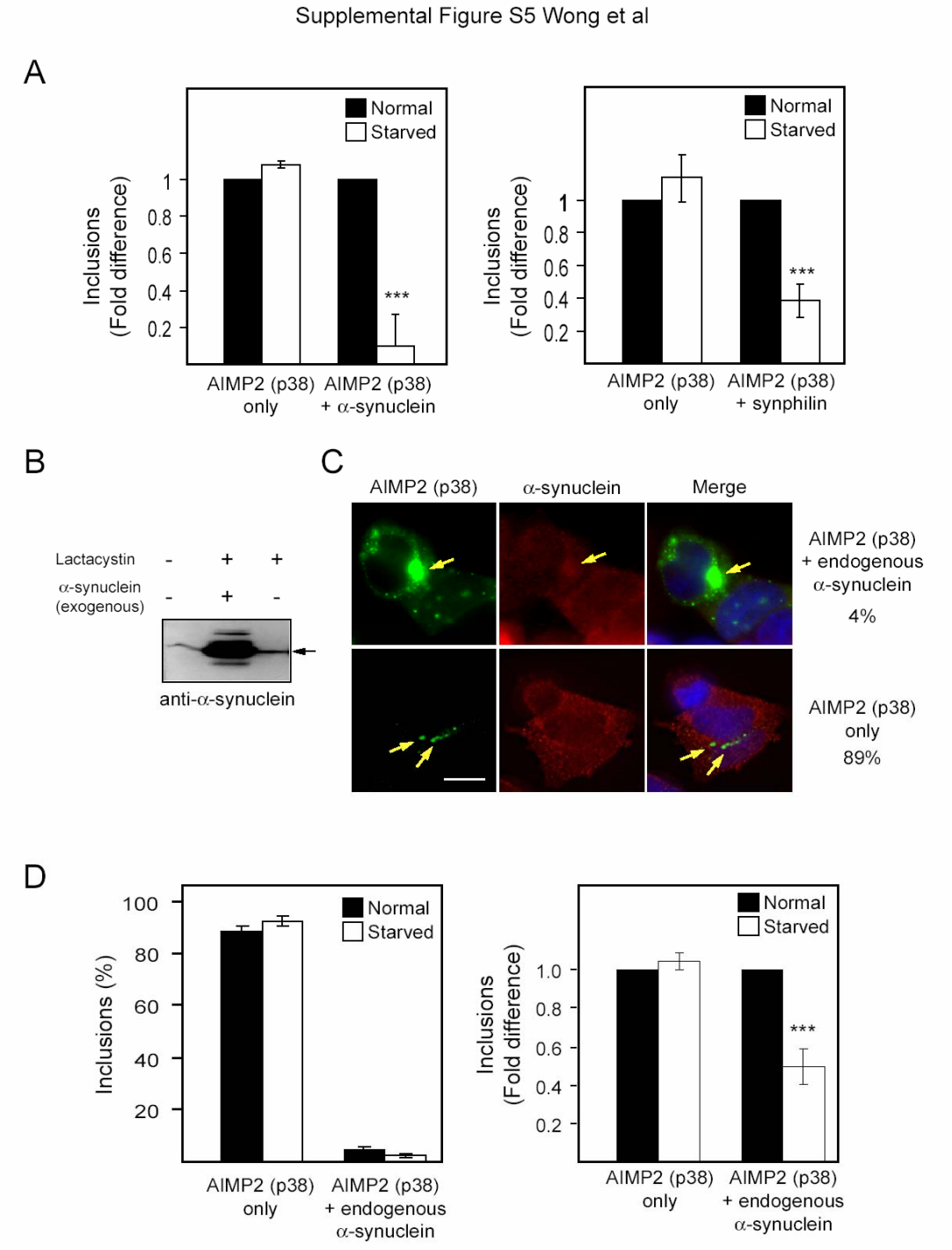

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