SUPPLEMENTARY INFORMATIONS

Sup. 1. The depolymerizing effect of wt α-synuclein is not due to severing of actin filaments. (A) Kinetics of actin depolymerization. Experiments were performed in a fluorometer at 30 ºC. Purified actin (5 μM; 5 % pyrenilated) was polymerized at high salt (85 mM NaCl, 30 mM KCl, 1 mM $MgCl_2$ and 0.1 μM $CaCl_2$) in the absence (CTRL = controls) or presence of 6 μM wt or A30P α-synuclein. At time 0, actin was diluted 50 fold in buffer without salts to a final concentration of 0.1 μM. (B) Fold changes of the actin maximum depolymerization rate induced by 6 μM wt or A30P α-synuclein. (C) Actin was polymerized in high salt for 1 hr and then incubated for 1 hr with either gelsolin (1 μM), wt or A30P synucleins (6 μM) and/or cytochalasin D (cytoD, 4 μM). The samples were then centrifuged at 355,000 x g for 30 min. Pellets and supernatants were loaded on an SDS-PAGE gel and stained with Coomassie blue. Gelsolin, used as a positive control for an actin-severing activity, reduces the amount of actin filaments recovered in the pellet, an effect which is observed with neither wt nor A30P α-synucleins, even in the presence of a capping agent such as cytochalasin D.

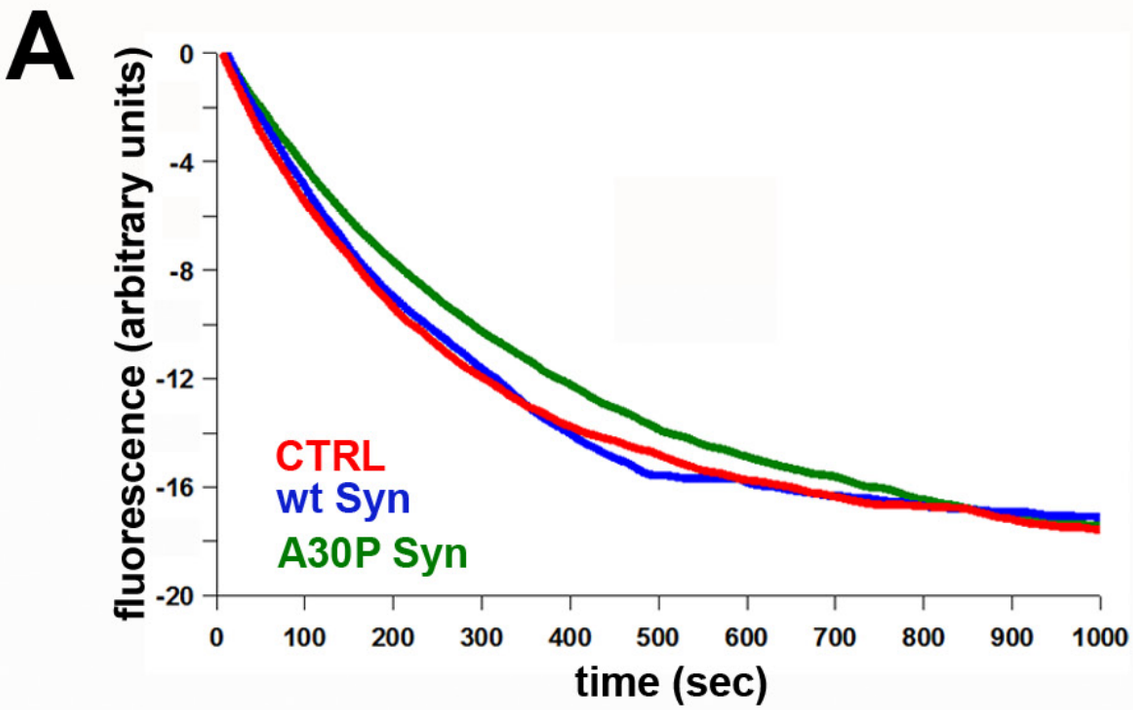
Sup. 2. α-Synuclein expression does not affect either cell proliferation or actin phosphorylation, and IPTG treatment does not affect the rate of actin cytoskeleton disassembly and reassembly.
(A) Proliferation rate of N2A cells, either non transfected (CTRL) or stably expressing wt or A30P α-synucleins. (B) non transfected MDCK cells, treated or not with IPTG and fixed either before the addition of LatA (NO LatA) or after 3 or 10 min of incubation with the drug (LatA), followed or not by 20, 40 or 60 min of recovery (LatA rec). (C) Anti-actin immunoprecipitate obtained from lysates of non transfected MDCK cells, treated or not with IPTG, and of induced clones expressing either wt or A30P α-synucleins, stained with anti-phosphoserine antibody (left panel) and with anti-actin antibody (right panel). Actin is phosphorylated at similar low levels in all four samples. As a reference for actin staining, 0.5 μg of purified actin (actin) was loaded onto the gels. Bar in B: 10 μm.

Video 1. Time-lapse movie (8 frames/sec) of a LatA experiment in a MDCK clone transiently transfected with actin-GFP. Frames, collected every 30 sec, are represented as Z-projection of 12 stacks.

Video 2. Time-lapse movie (8 frames/sec) of a LatA experiment in a MDCK clone transiently transfected with actin-GFP and induced by IPTG to express wt α-synuclein. Frames, collected every 30 sec, are represented as Z-projection of 12 stacks.

Video 3. Time-lapse movie (8 frames/sec) of a LatA experiment in a MDCK clone transiently transfected with actin-GFP and induced by IPTG to express A30P α-synuclein. Frames, collected every 30 sec, are represented as Z-projection of 12 stacks.

SUP. 1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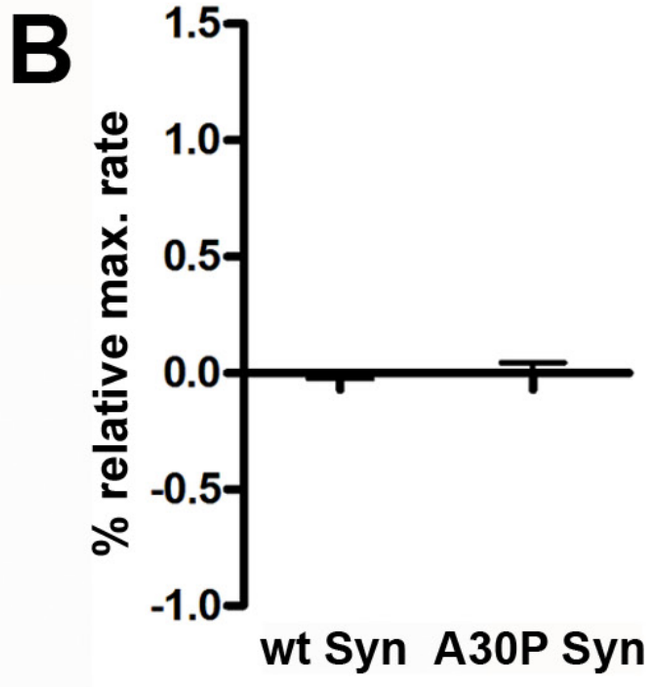
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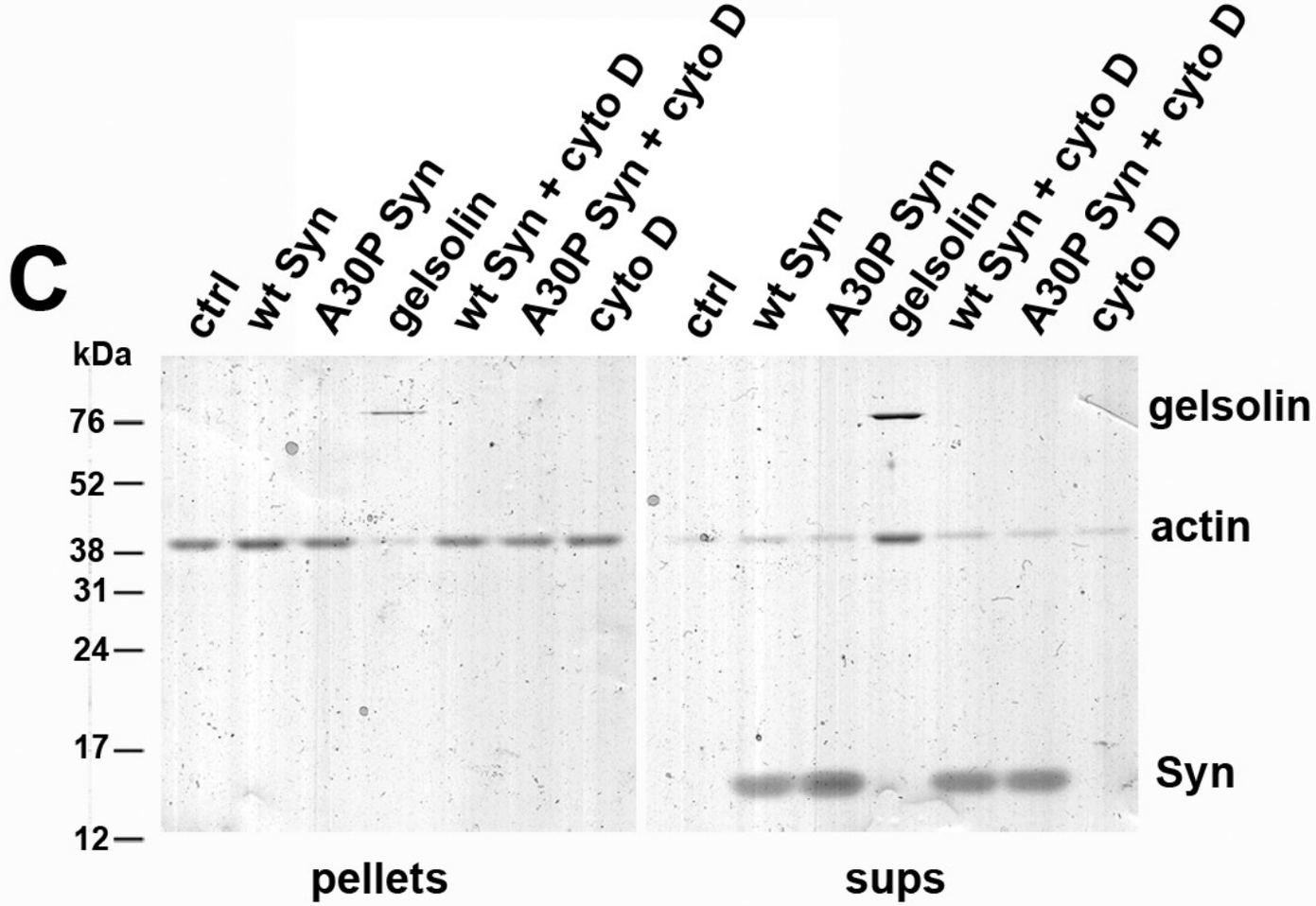
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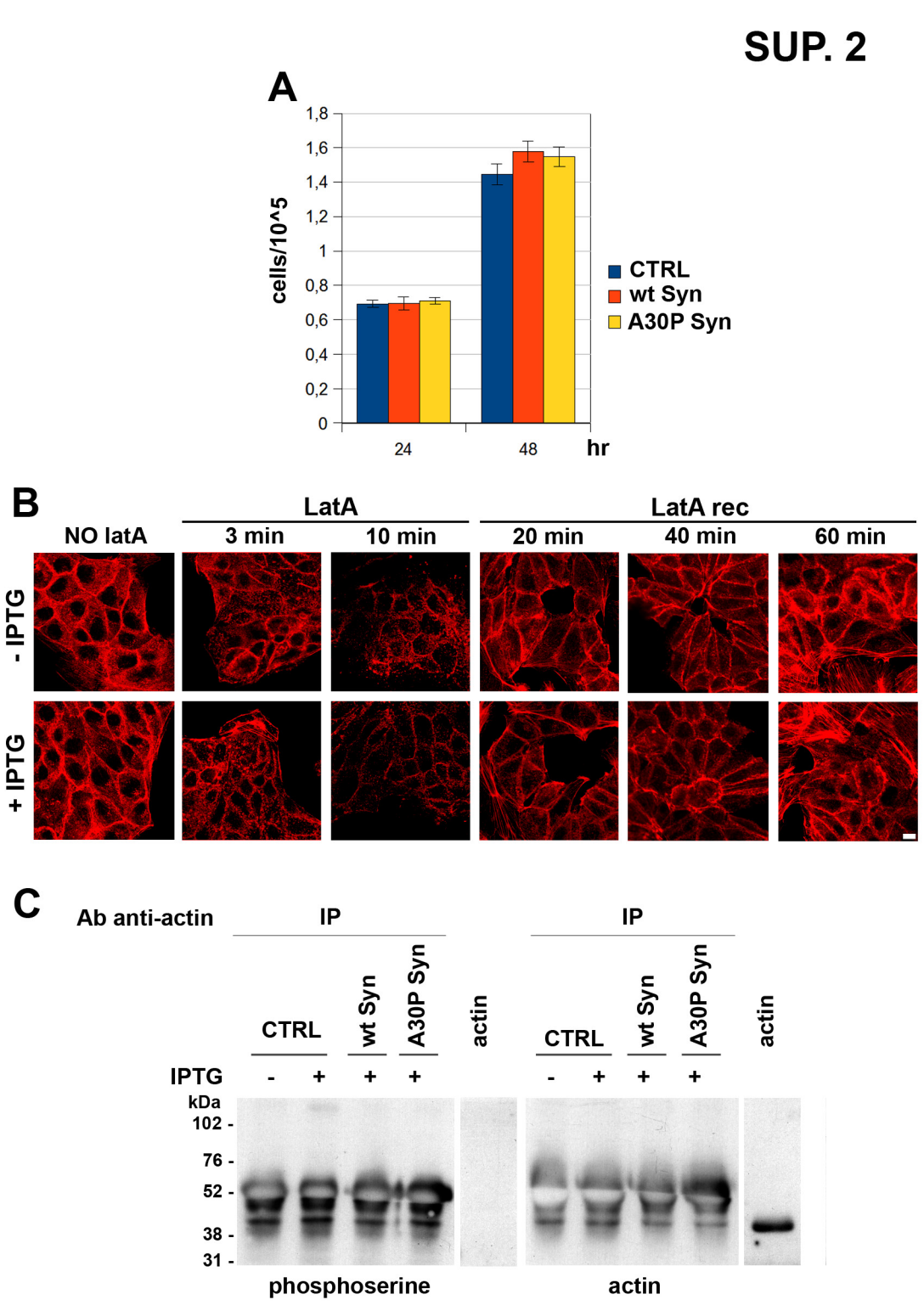
SUP. 2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