Additional file

Progressive lysosomal membrane permeabilization induced by iron oxide nanoparticles drives hepatic cell autophagy and apoptosis

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Additional figures

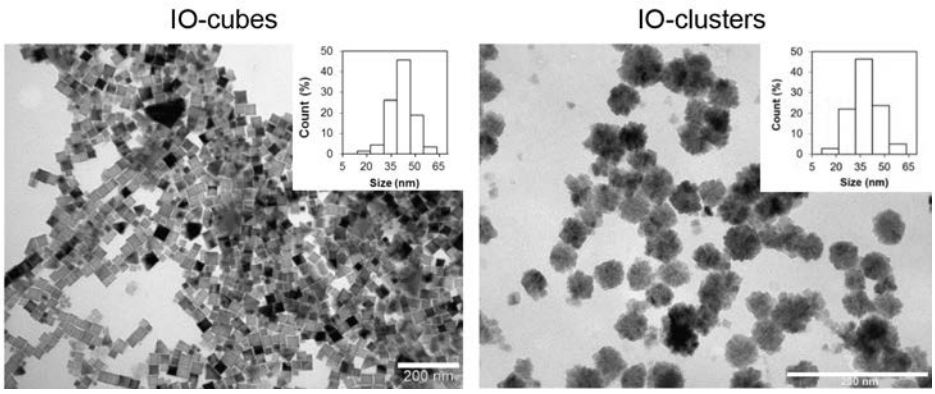

Figure S1. Transmission electron micrographs of the iron core of the nanoparticles.

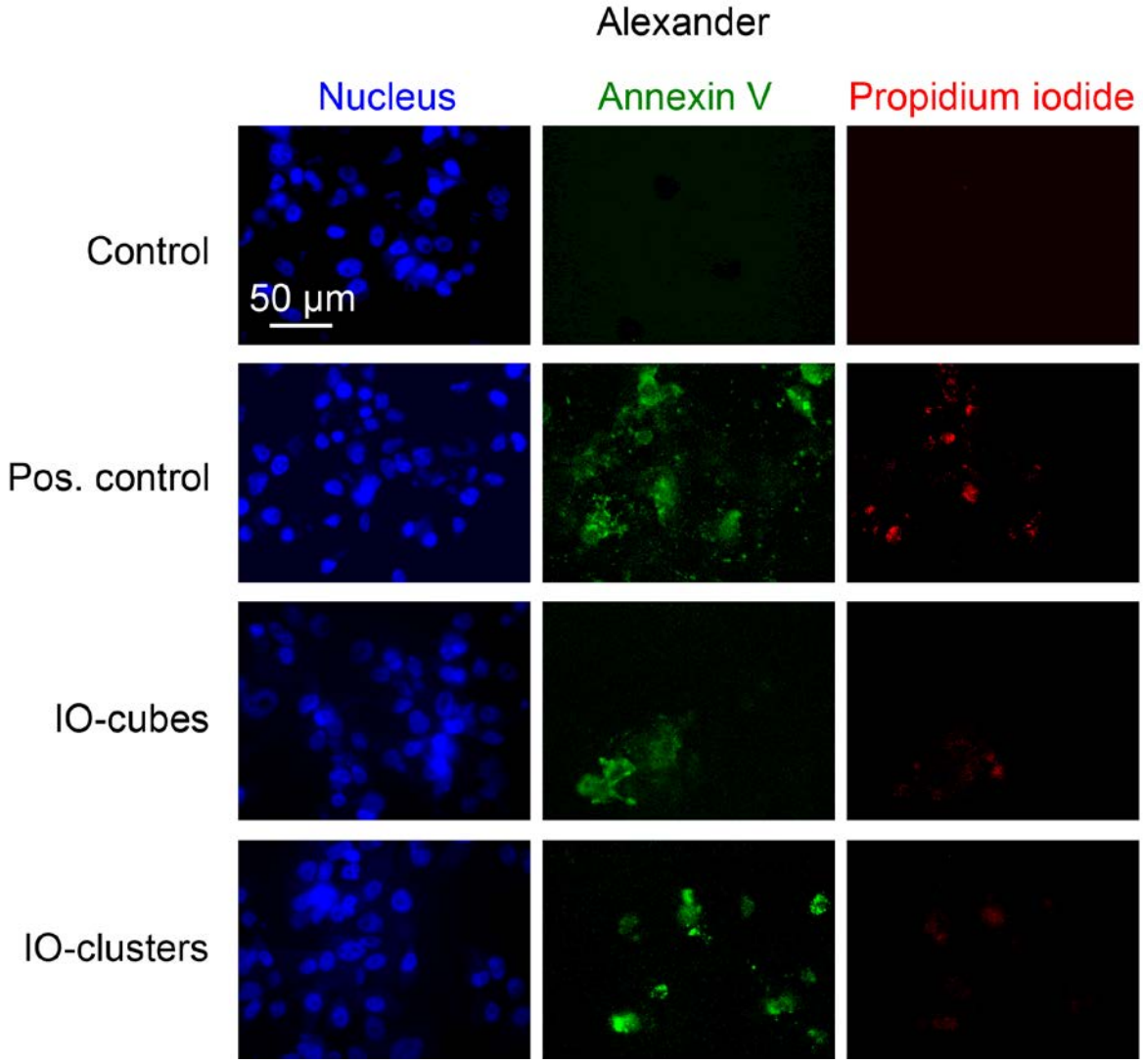

Figure S2. Alexander cells were stimulated with IO-cubes or IO-clusters (100 µg/mL) for 24 h and labeled with annexin V – green dye, propidium iodide – red dye and hoechst 33342 nuclear stain – blue. Labeled cells were imaged with epi-fluorescence microscopy.

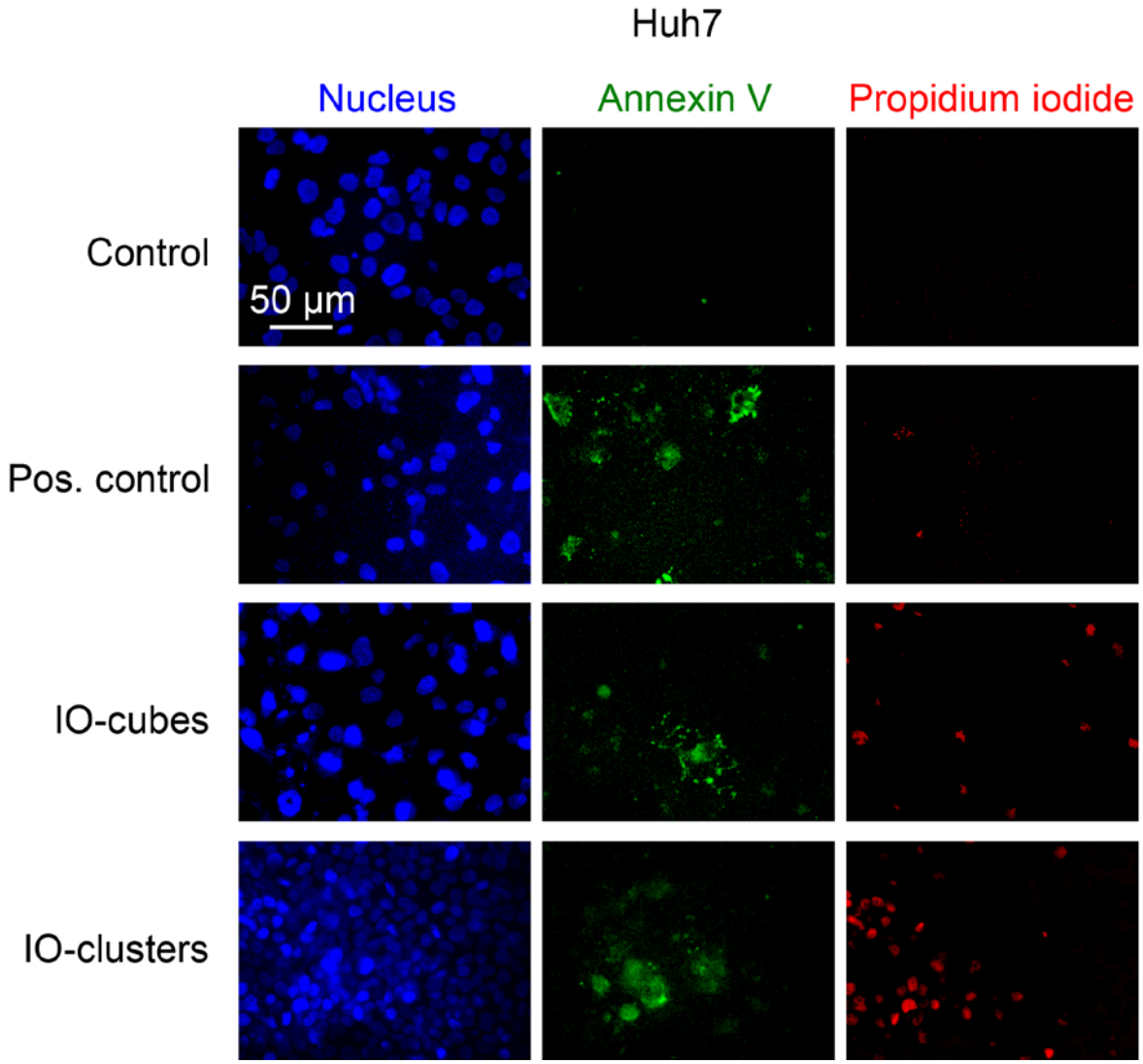

Figure S3. Huh7 cells were stimulated with IO-cubes or IO-clusters (100 µg/mL) for 24 h and labeled with annexin V – green dye, propidium iodide – red dye and hoechst 33342 nuclear stain – blue. Labeled cells were imaged with epi-fluorescence microscopy.

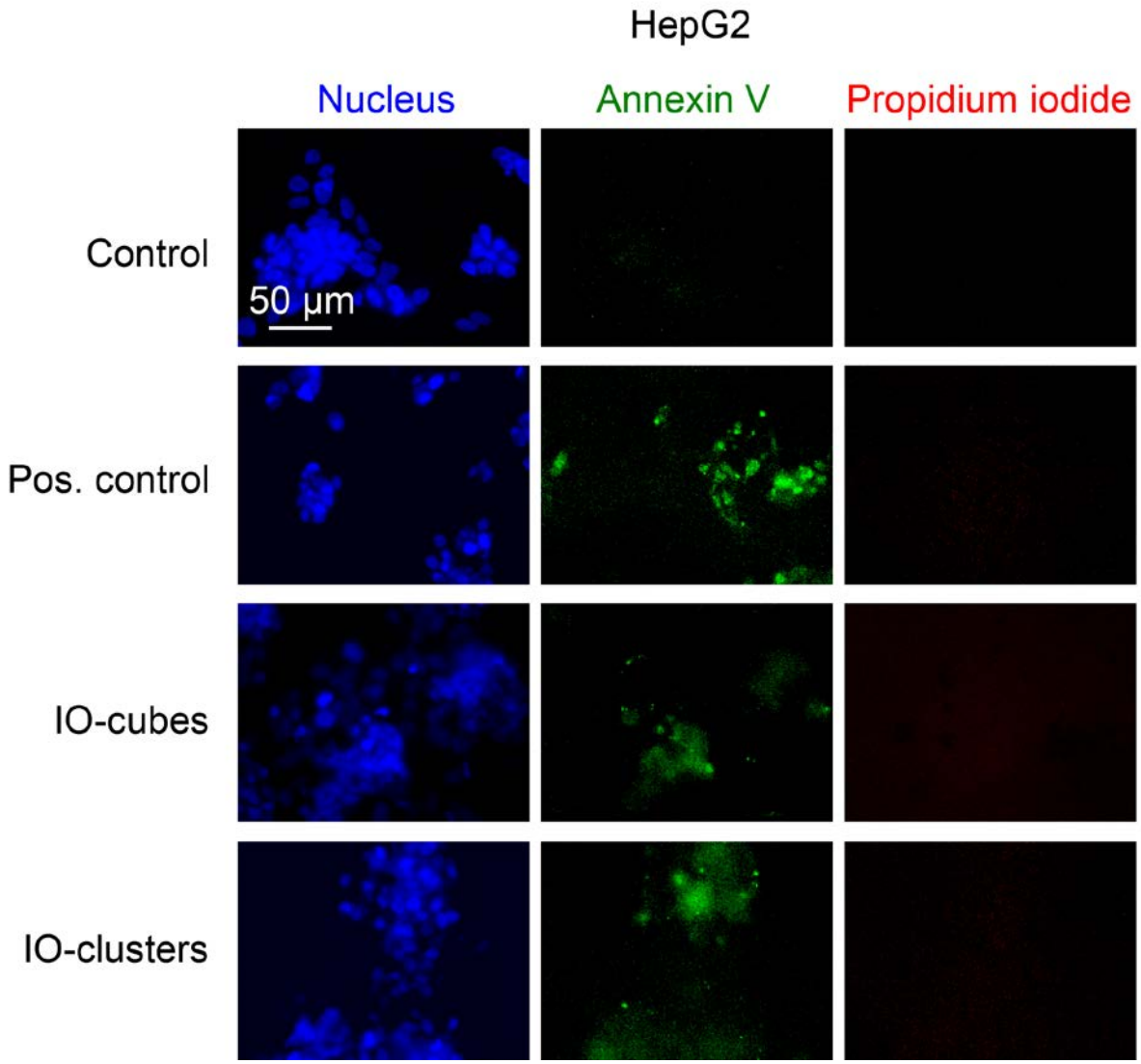

Figure S4. HepG2 cells were stimulated with IO-cubes or IO-clusters (100 µg/mL) for 24 h and labeled with annexin V – green dye, propidium iodide – red dye and hoechst 33342 nuclear stain – blue. Labeled cells were imaged with epi-fluorescence microscopy.

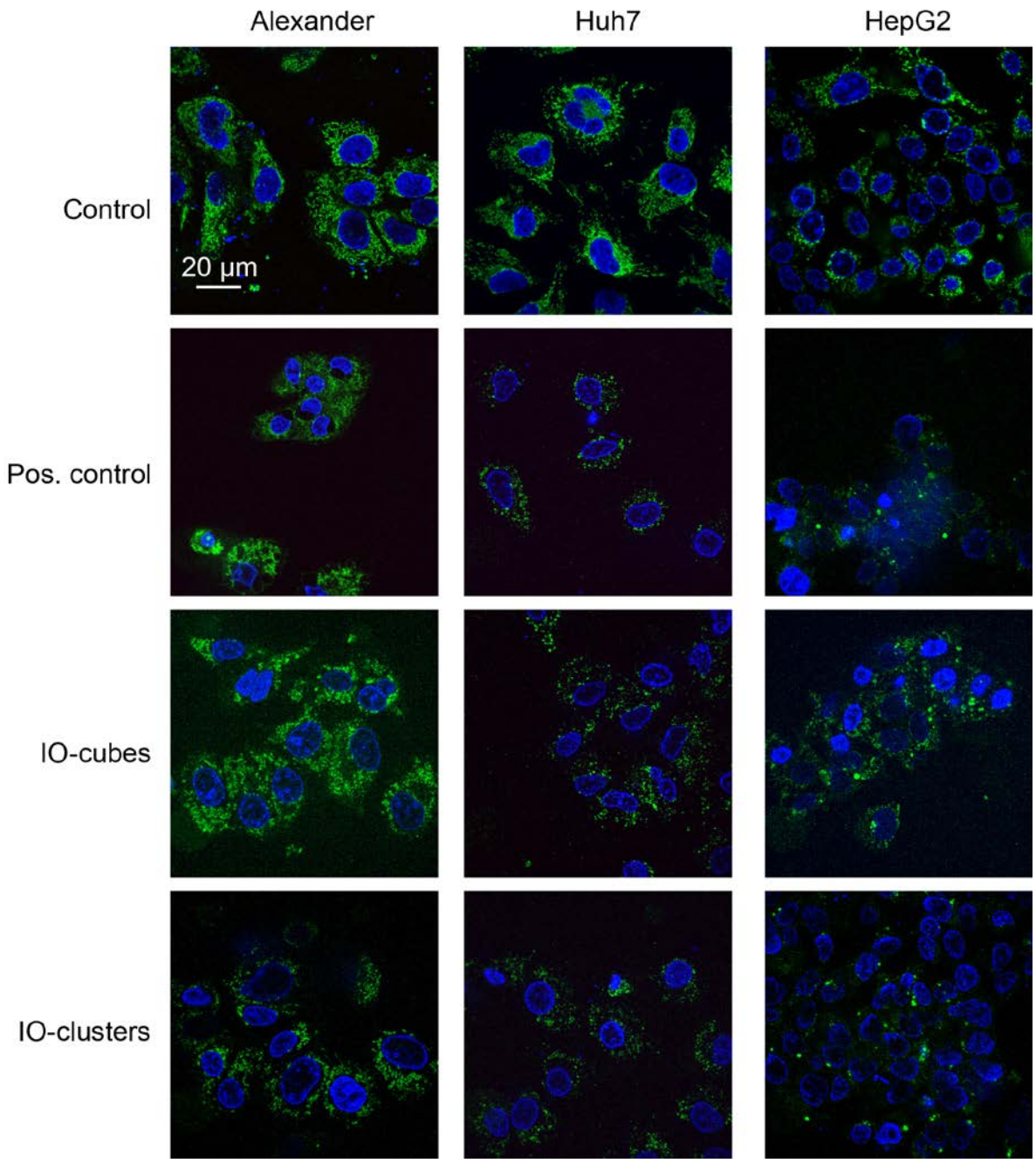

Figure S5. Alteration of mitochondrial morphology by IO-cubes and IO-clusters treatment. Alexander, HepG2 and Huh7 cells were stimulated with IO-cubes or IO-clusters (100 µg/mL) for 24 h and labeled with MitoTracker® green. Positive control – 20 % ethanol for 20 min. Nuclei were labelled with hoechst 33342 nuclear stain (blue). Labeled cells were then imaged using spinning disk confocal microscopy.

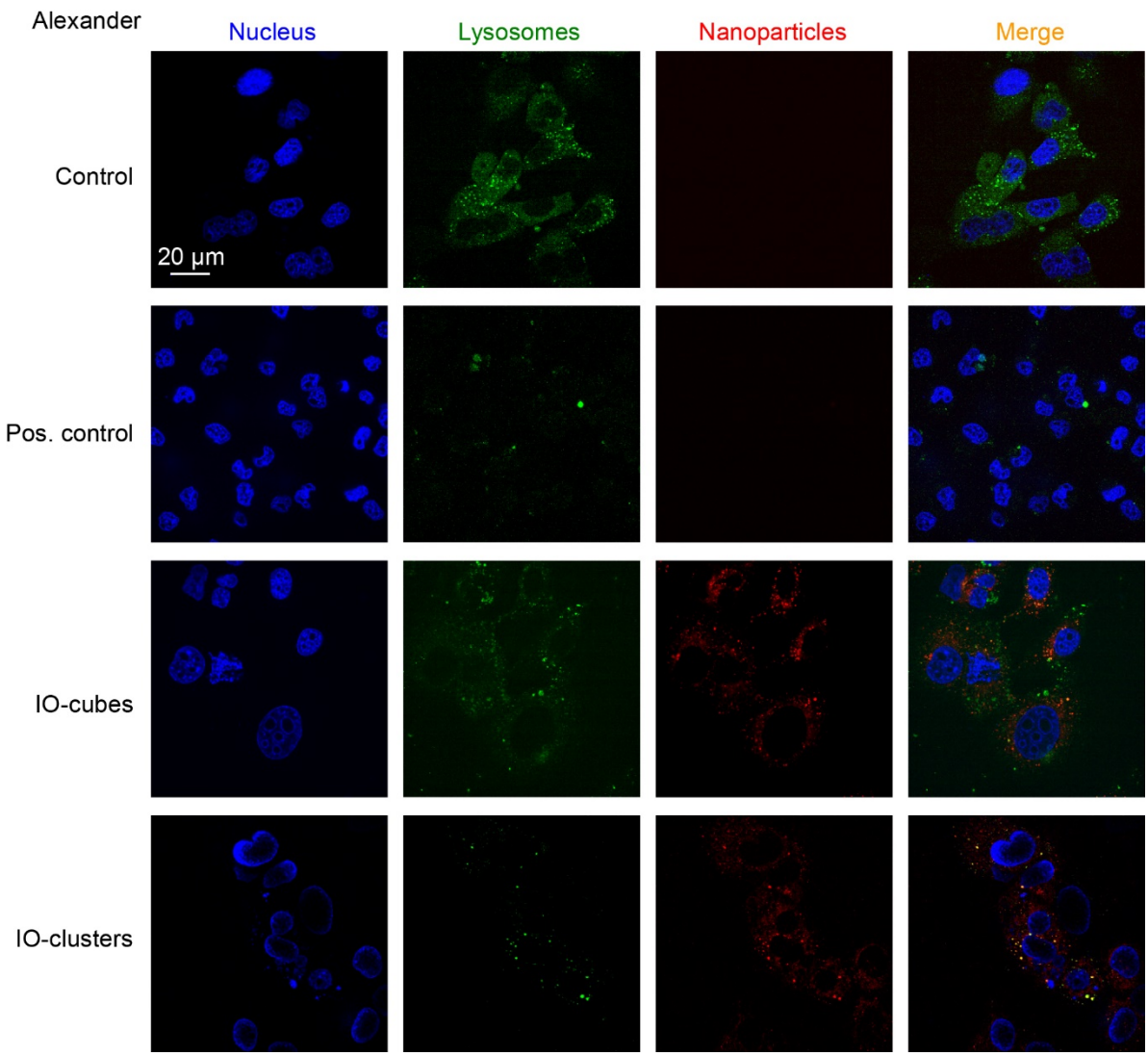

Figure S6. Alexander cells were treated with fluorescently labeled (red) IO-cubes or IO-clusters (100 µg/mL) for 24 h and stained with LysoTracker (green), colocalization of fluorescently labeled nanoparticles with lysosomes (yellow). Positive control – 20 % ethanol for 20 min. Nuclei were labelled with hoechst 33342 nuclear stain (blue). Labeled cells were then imaged using spinning disk confocal microscopy.

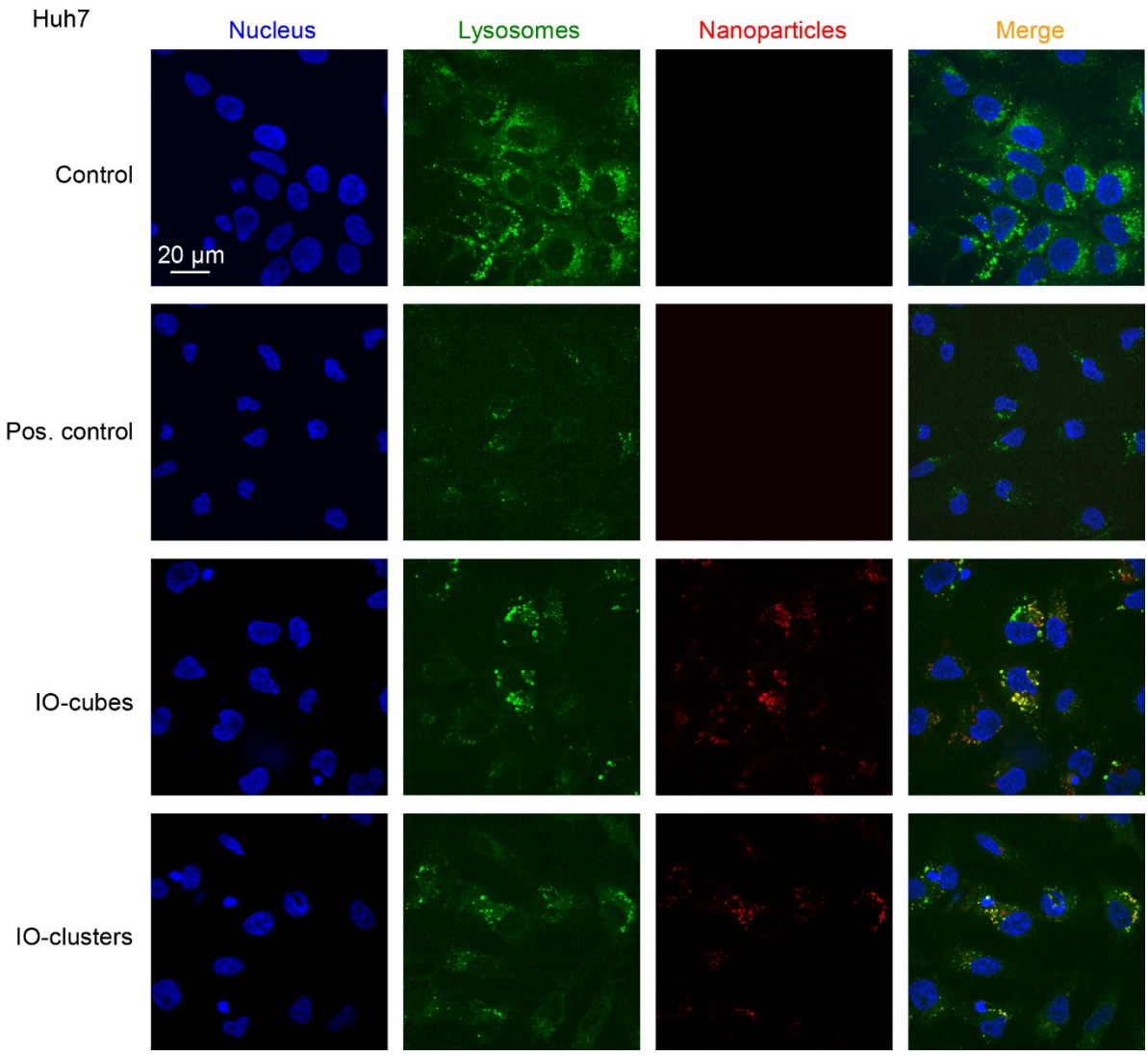

Figure S7. Huh7 cells were treated with fluorescently labeled (red) IO-cubes or IO-clusters (100 µg/mL) for 24 h and stained with LysoTracker (green), colocalization of fluorescently labeled nanoparticles with lysosomes (yellow). Positive control – 20 % ethanol for 20 min. Nuclei were labelled with hoechst 33342 nuclear stain (blue). Labeled cells were then imaged using spinning disk confocal microscopy.

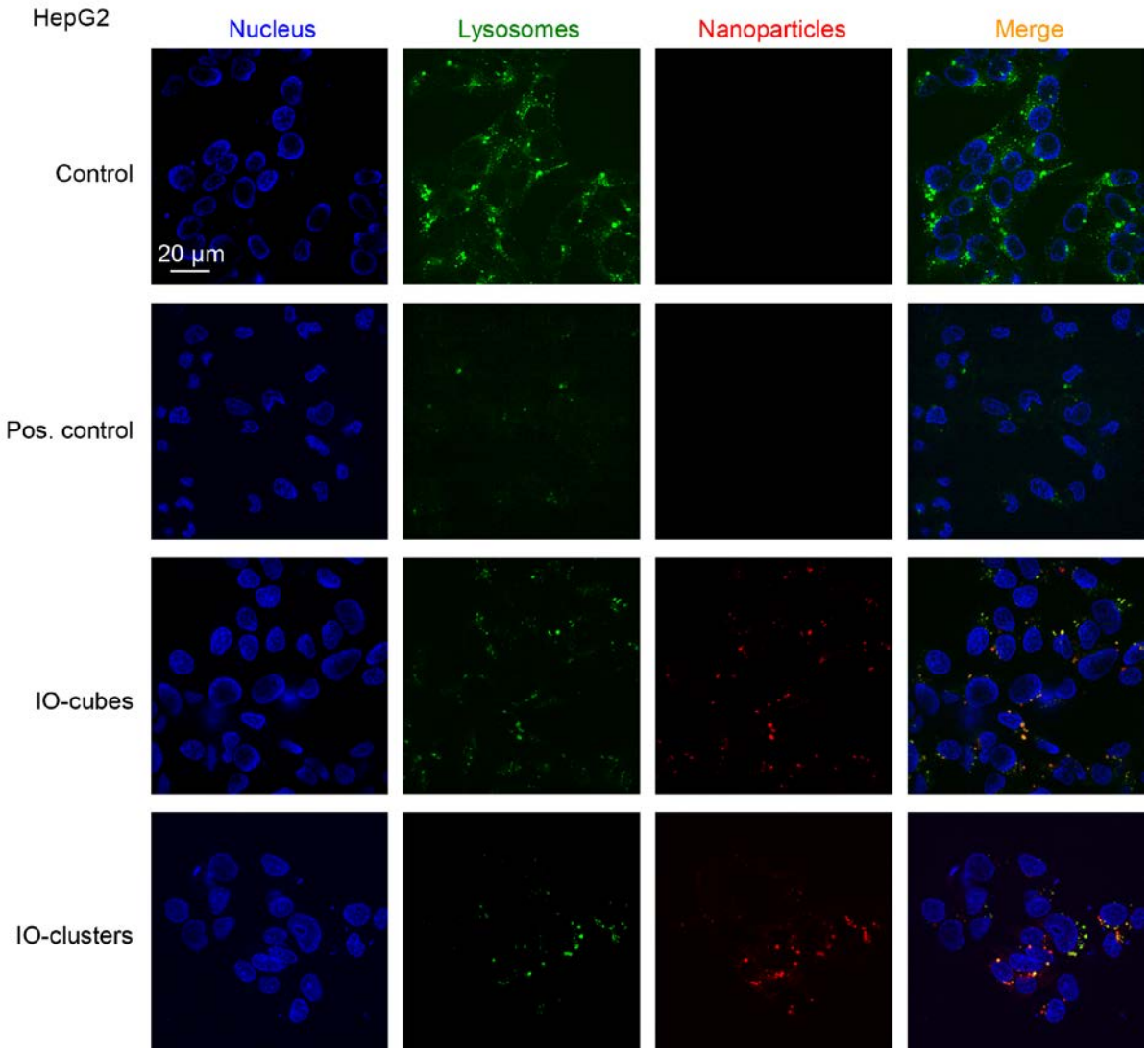

Figure S8. HepG2 cells were treated with fluorescently labeled (red) IO-cubes or IO-clusters (100 µg/mL) for 24 h and stained with LysoTracker (green), colocalization of fluorescently labeled nanoparticles with lysosomes (yellow). Positive control – 20 % ethanol for 20 min. Nuclei were labelled with hoechst 33342 nuclear stain (blue). Labeled cells were then imaged using spinning disk confocal microscopy.

Uncropped immunoblot scans