Supplemental Information

Small Molecule Targeting of Oxysterol-Binding Protein (OSBP)-Related Protein 4 (ORP4) and OSBP Inhibits Ovarian Cancer Cell Proliferation in Monolayer and Spheroid Cell Models

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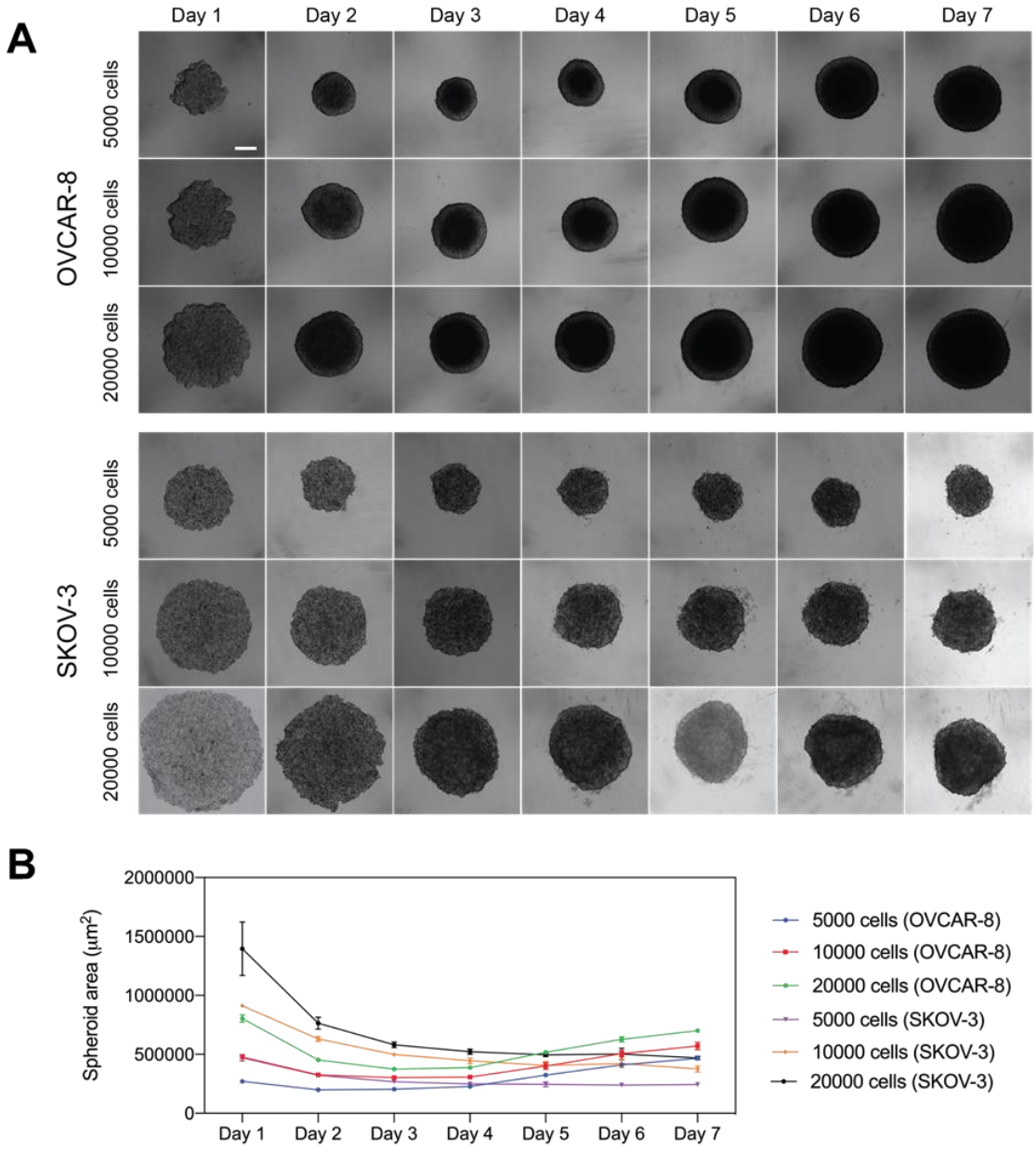

Figure S-1: OVCAR-8 and SKOV-3 spheroid morphology. A) OVCAR-8 and SKOV-3 spheroids seeded at the provided density for 7 days. B) Spheroid area measurement for either OVCAR-8 or SKOV-3 spheroids over the course of 7 days.

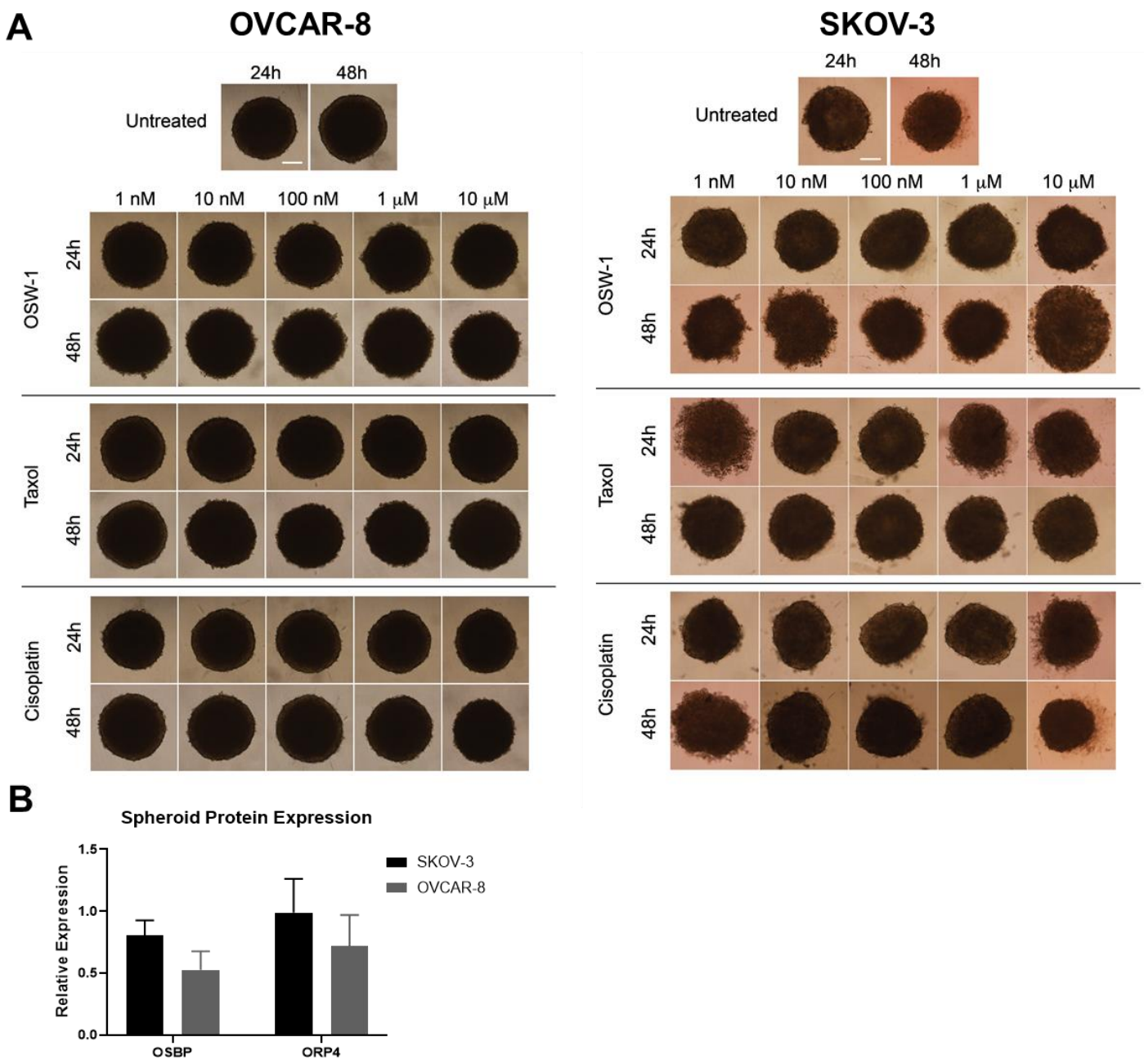

Figure S-2: Effects of compound treatment on OVCAR-8 and SKOV-3 spheroids. A) Images of 7-day-old SKOV-3 and OVCAR-8 spheroids treated with the indicated concentrations of compound at 24 and 48 h. B) Protein expression of OSBP and ORP4 in 7-day-old untreated spheroids relative to monolayer expression.

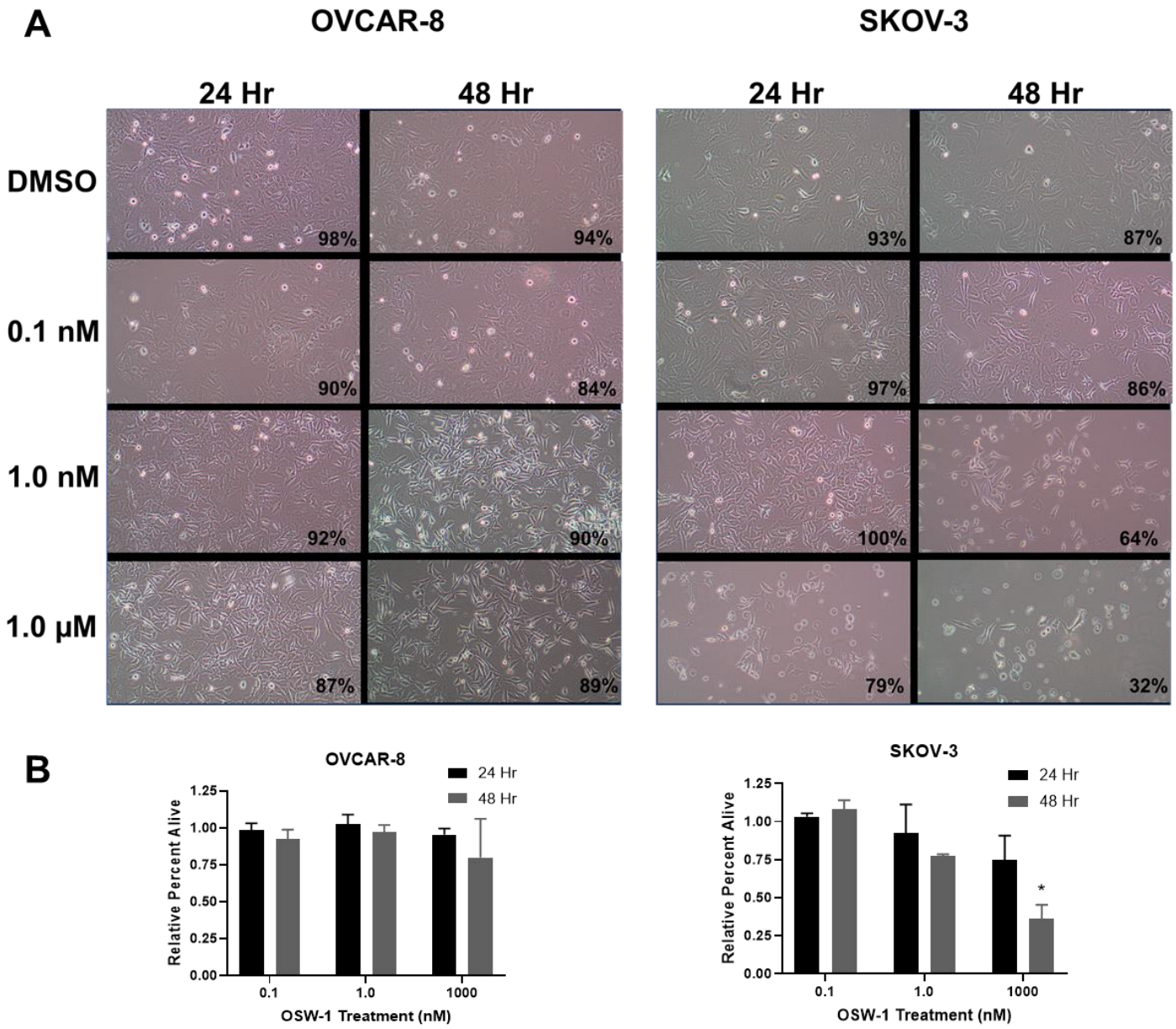

Figure S-3: Effects of OSW-1 on ovarian cancer cells morphology and viability. A) 40X light microscopy imaging of OVCAR-8 and SKOV-3 monolayer (2D) treated with either DMSO vehicle control or OSW-1 at the indicated concentrations, at 24 h and 48 h. B) Determination of viability of OVCAR-8 or SKOV-3 cells treated with in the indicated concentrations of OSW-1 at 24 h and 48 h, using Trypan Blue staining. * = p value ≤ 0.05.

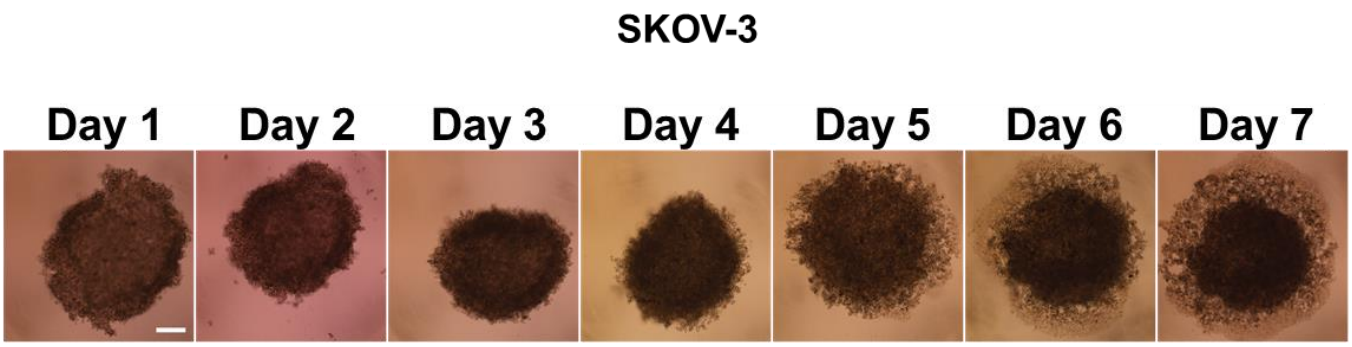

Figure S-4: SKOV-3 spheroid development with 10% FBS. 20,000 SKOV-3 cells culture as spheroids in the ultra-low attachment conditions failed to produce organized spheroids in the presence of FBS.

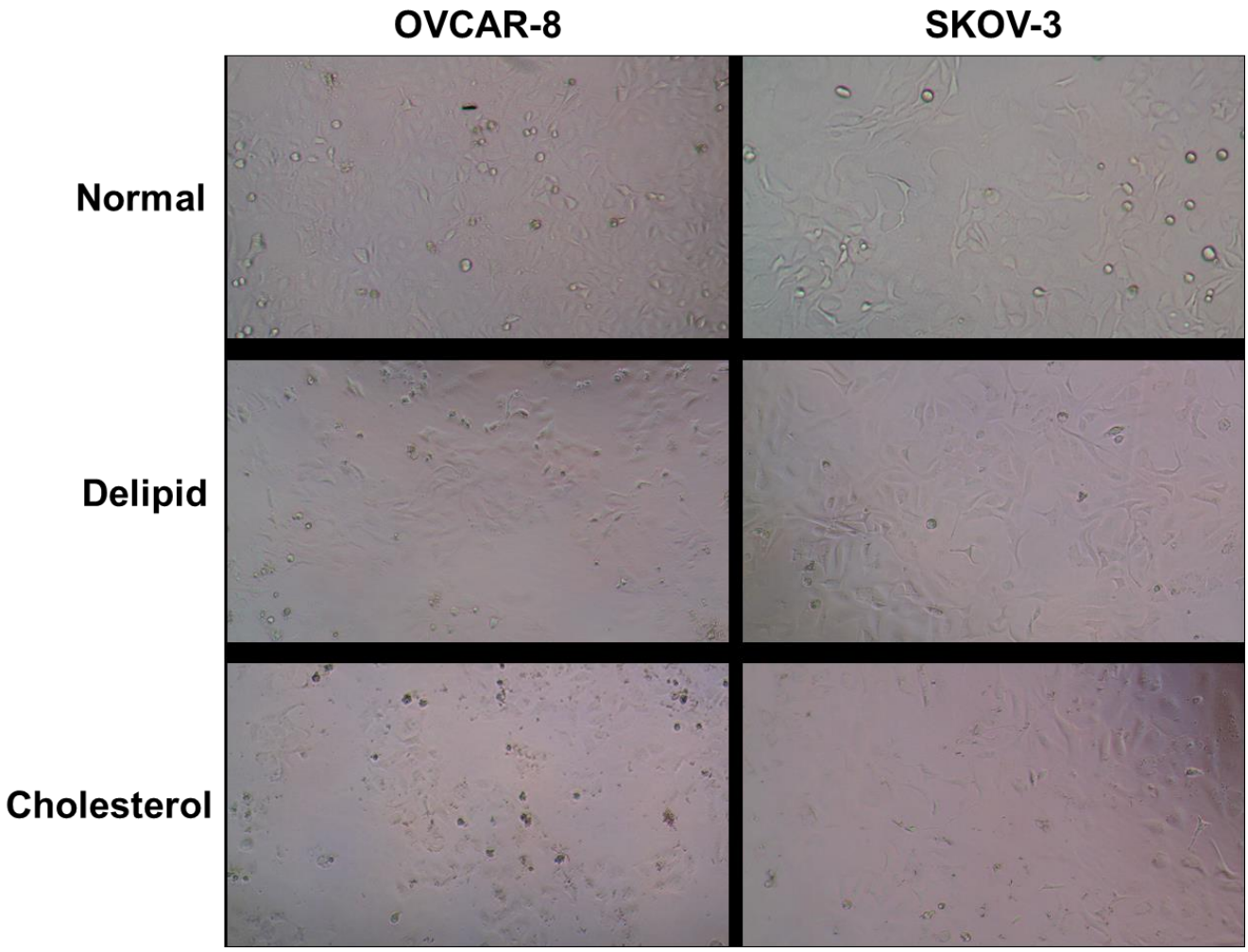

Figure S-5: Cell imaging of ovarian cancer cells grown under lipid depleted conditions: 40X magnification of OVCAR-8 and SKOV-3 2D cells grown in normal media (RPMI with 10% FBS), delipidated media (RPMI with 10% delipidated FBS), or cholesterol-supplemented delipidated media (RPMI with 10% delipidated FBS and 20 µg/mL cholesterol) for 96 h.

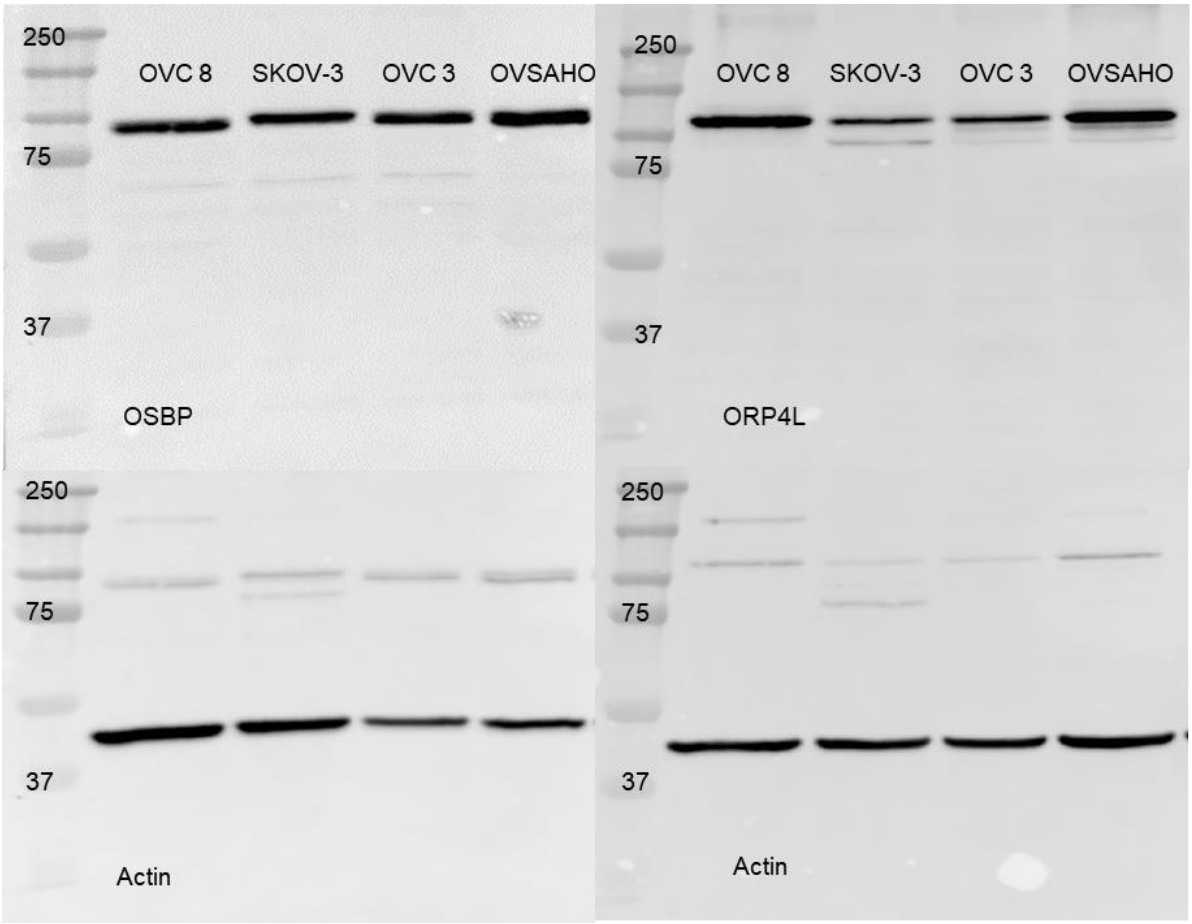

Figure S-6. Uncropped OSBP and ORP4 Western blots shown in Figure 1D.

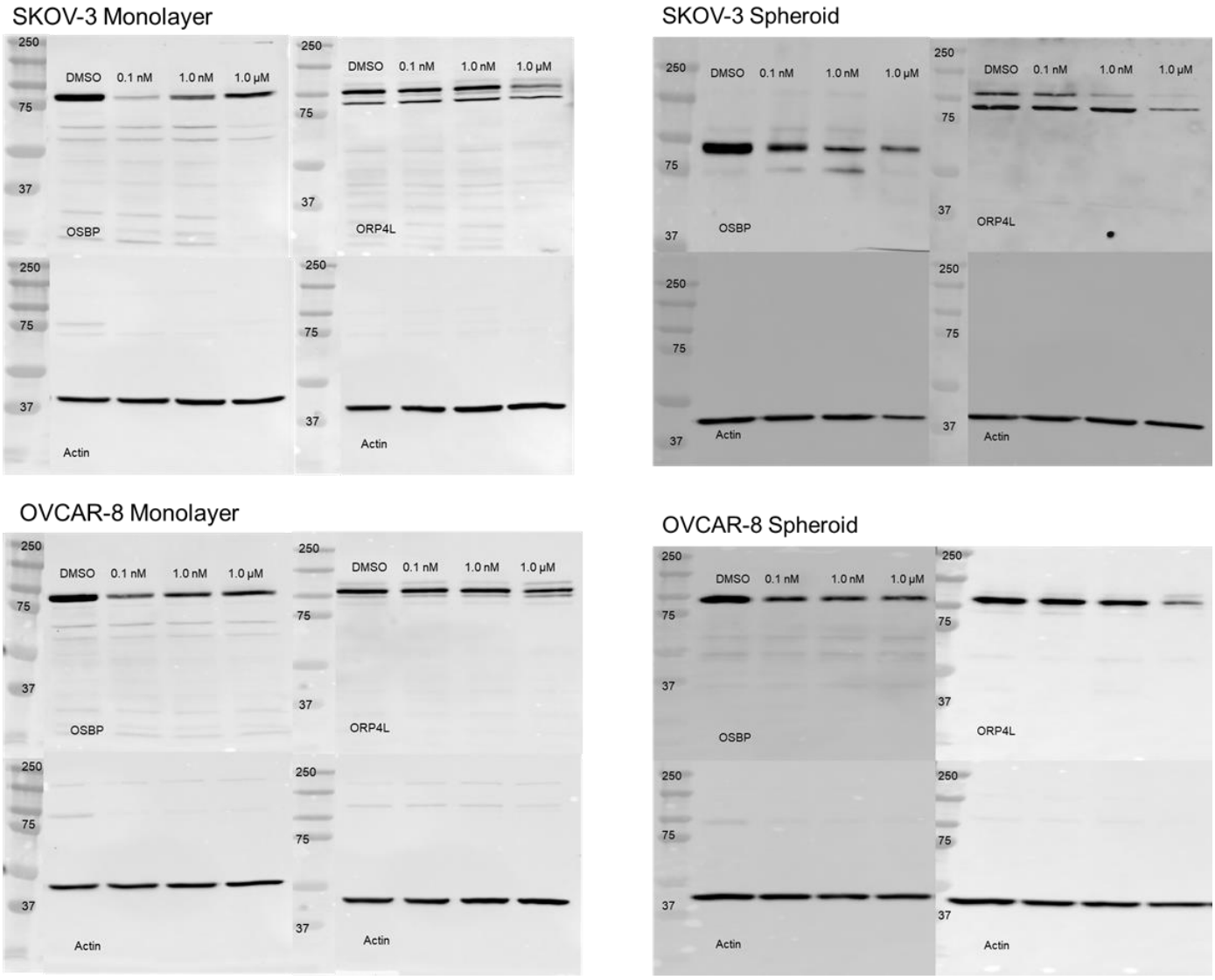

Figure S-7. Uncropped OSBP and ORP4 Western blots shown in Figure 5.