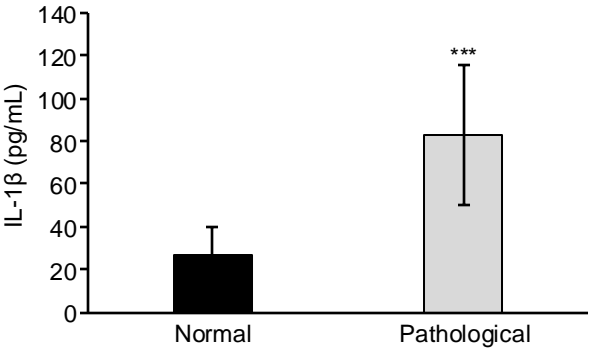
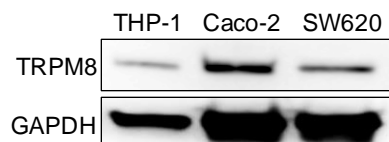
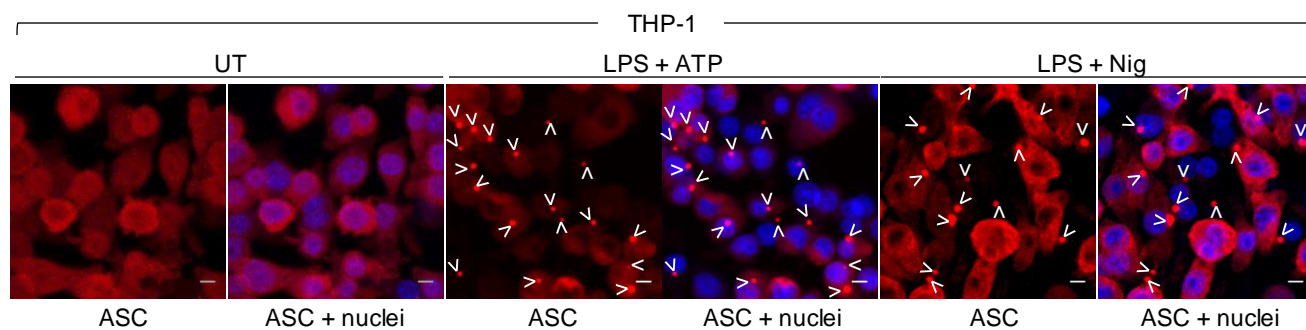
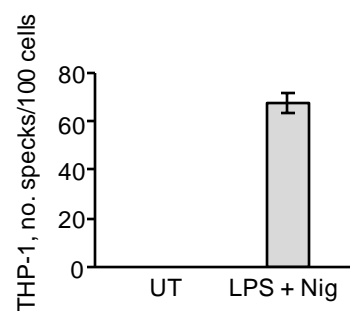
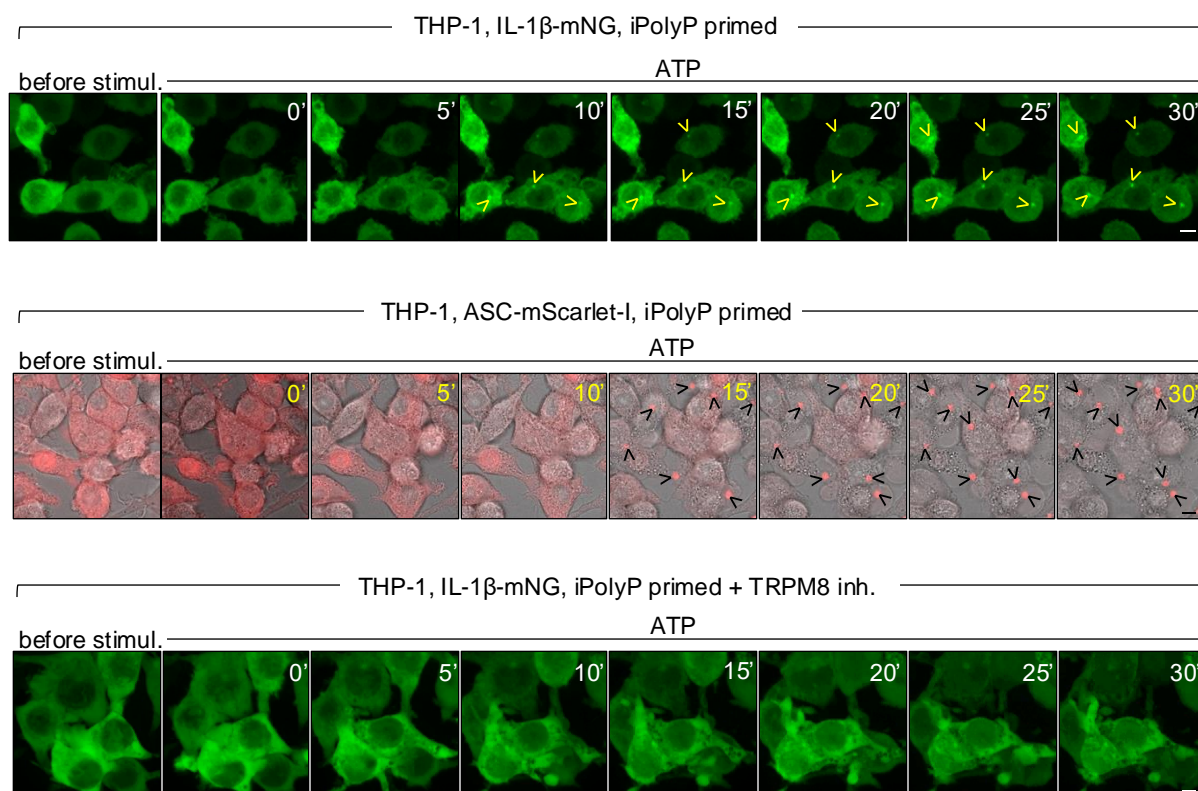


A



A**B****C****D**

Supplementary Figure legend

Supplementary Figure S1: iPolyP induces α -SMA expression and fosters Caco-2- and SW620-derived spheroids growth. **A.** Immunofluorescence images performed on HCEC-1CT- (upper panel), Caco-2- (middle panel) and SW620-derived (lower panel) spheroids for α -SMA detection upon treatment with iPolyP, TRPM8 inhibitor or both for 48 hours. Scale bar 100 μ m. **B.** Quantification relative to panel **A**. Fold changes versus control, untreated (UT). Statistical analysis performed by Student's t-test (**** $p < 0.0001$). **C.** Immunofluorescence images showing F-actin and Fibronectin level performed on HCEC-1CT (upper panel), Caco-2 (middle panel) and SW620 (lower panel) cell line upon treatment with iPolyP, TRPM8 inhibitor or both. **D.** Quantification relative to panel **C**. Fold changes versus control; untreated (UT). Statistical analysis performed by Student's t-test (**** $p < 0.0001$, ** $p < 0.01$). Cells were treated with iPolyP and TRPM8 inhibitor for 48 hours.

Supplementary Figure S2: A. CRC patients display elevated level of circulating IL-1 β . Evaluation of plasma-derived IL-1 β performed by ELISA on normal (n = 10) and CRC subjects (n = 10). Fold changes of Pathological subjects versus Normal individuals. Statistical analysis performed by Student's t-test (** $p < 0.01$, *** $p < 0.001$).

Supplementary Figure S3: iPolyP + ATP trigger the NLRP3 inflammasome, mimicking the canonical activation mediated by LPS and ATP. **A.** Cellular extracts from THP-1 monocyte, treated with LPS, iPolyP or iPolyP + TRPM8 inhibitor for 4 hours were analyzed by immunoblotting against pro-IL-1 β and TRPM8, normalized on GAPDH. **B.** Representative confocal micrographs on THP-1 cells treated with LPS (4 hours) + ATP (30 minutes) or LPS (4 hours) + Nigericin (30 minutes), used as positive control, untreated (UT). White angle brackets indicate the presence of ASC specks. Scale bar 10 μ m. Images are representative of three independent experiments. **C.** Quantification of the number of ASC specks, relative to panel **B**. **D.** Upper panel, confocal micrographs of time-lapse confocal microscopy performed on THP-1 cells, stably expressing IL-1 β -mNG, upon treatment with iPolyP (4 hours) + ATP (30 minutes). Yellow angle brackets indicate ASC specks; Middle panel, time-lapse experiment in bright-field merged with fluorescence channel on THP-1 cell, stably expressing ASC-mRuby3, upon treatment with iPolyP (4 hours) + ATP (30 minutes). Black angle brackets indicate ASC-mScarlet-I specks; Lower panel, time-lapse confocal microscopy performed on THP-1 cells, stably expressing IL-1 β -mNG, upon treatment with iPolyP (4 hours) + ATP (30 minutes) + TRPM8 inhibitor (4 hours). Scale bar 10 μ m. Data are presented as mean \pm SD for triplicate wells from three independent experiments.

Supplementary Videos: Time-lapse confocal microscopy on THP-1 cells treated with iPolyP. **Supplementary Video 1:** THP-1, IL-1 β -mNG, iPolyP (4 hours) + ATP (30 minutes); Scale bar = 10 μ m. **Supplementary Video 2:** THP-1 ASC-mScarlet-I, iPolyP (4 hours) + ATP (30 minutes); Scale bar = 5 μ m. **Supplementary Video 3:** THP-1, IL-1 β -mNG, iPolyP (4 hours) + ATP (30 minutes) + TRPM8 inhibitor (4 hours); Scale bar 10 μ m.