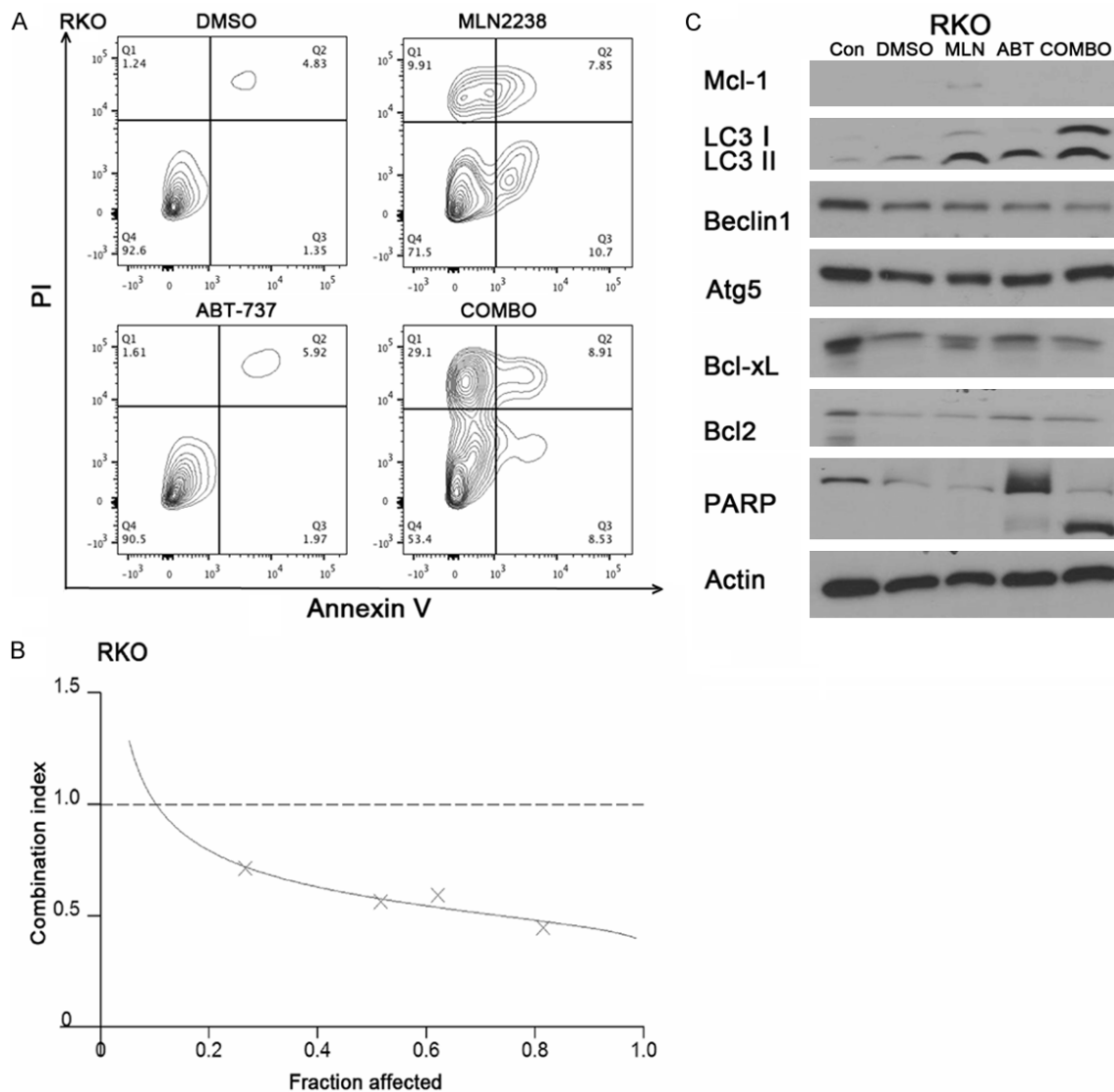


# ABT-737 sensitizes colorectal cancer cells to ixazomib

## Supplementary method

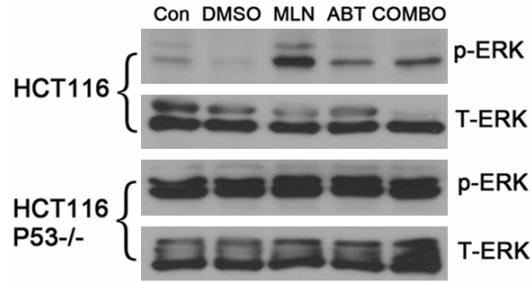
### Colony formation

Colony formation in HCT116, LOVO, SW480, HT-29 clones on culture plates. Each bar represents the average number of colonies formed by the different cell lines. For analyses of colony formation on culture plates,  $1.0 \times 10^3$  cells were plated into each well of a six-well plate and exposed previously to DMSO (Ctrl), MLN2238, and/or ABT-737 for 24 h. Cell lines were treated with MLN2238 (200 nM) and/or ABT-737 (2  $\mu$ M). These cells allowed to grow for 14-20 days. Colonies of cells were stained with 0.1% crystal violet and the total number of colonies per well were counted using Colony 1.1 software (Fujifilm, Tokyo, Japan).

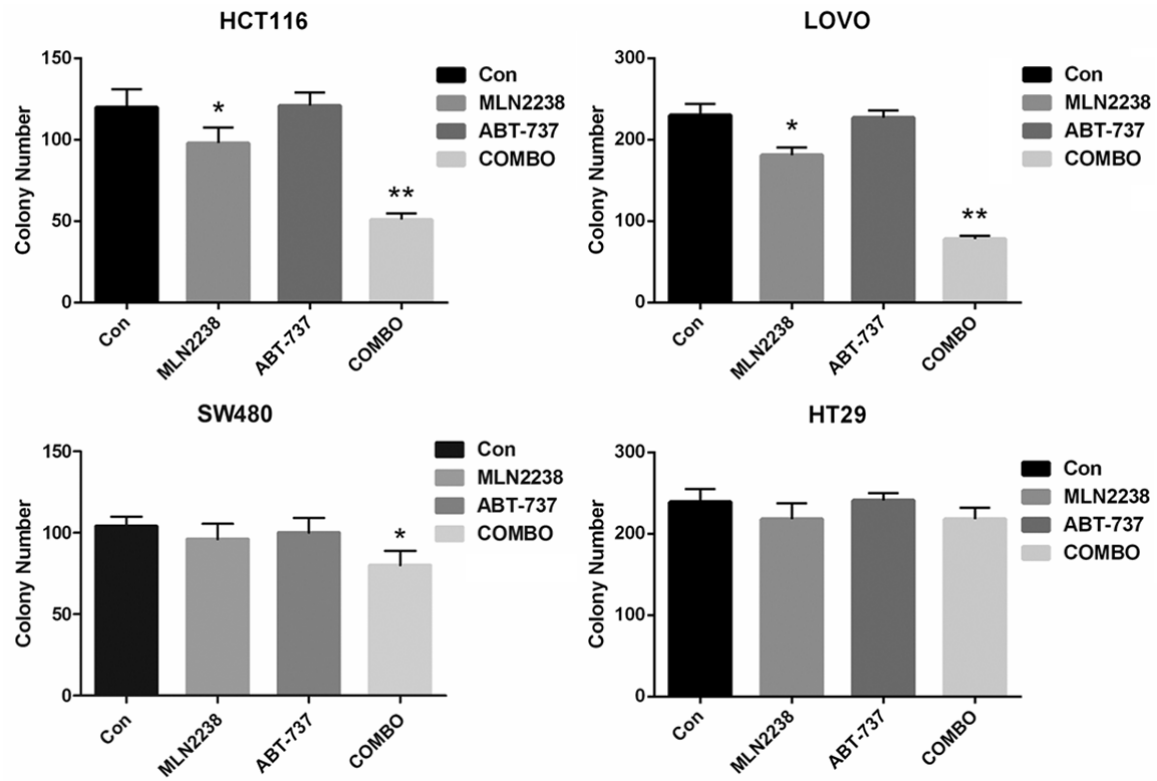


**Supplementary Figure 1.** ABT-737 sensitizes RKO cells to MLN2238. Cultured RKO cells were treated with doses of MLN2238 (200 nM), ABT737 (2  $\mu$ M), or the combination for 24 h. A. Flow cytometry analysis of annexin-V and propidium iodide (PI) staining of apoptotic cells following the treatment. B. Protein lysates were harvested and western blots performed using antibodies for Mcl-1, LC3, Beclin1, Atg5, Bcl-xL, Bcl2, PARP and Actin. C. The CI was calculated after treatment of MLN2238 and ABT-737 at a fixed ratio (1:10). Isobologram showing CI < 1 indicates synergy.

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**Supplementary Figure 2.** Combination induced p-ERK expression decrease is P53 related. Cultured HCT116 and HCT116 P53<sup>-/-</sup> cells were treated with doses of MLN2238 (200 nM), ABT737 (2  $\mu$ M), or the combination for 24 h. Protein lysates were harvested and western blots performed using antibodies for p-ERK and T-ERK.



**Supplementary Figure 3.** ABT-737 sensitizes cells to MLN2238. Cell lines were treated with doses of MLN2238 (200 nM), ABT737 (2  $\mu$ M), or the combination for 24 h. Colonies of cells were counted after 14-20 days. Mean  $\pm$  SD. \*,  $P < 0.05$ , \*\*,  $P < 0.005$ , relative to Control.