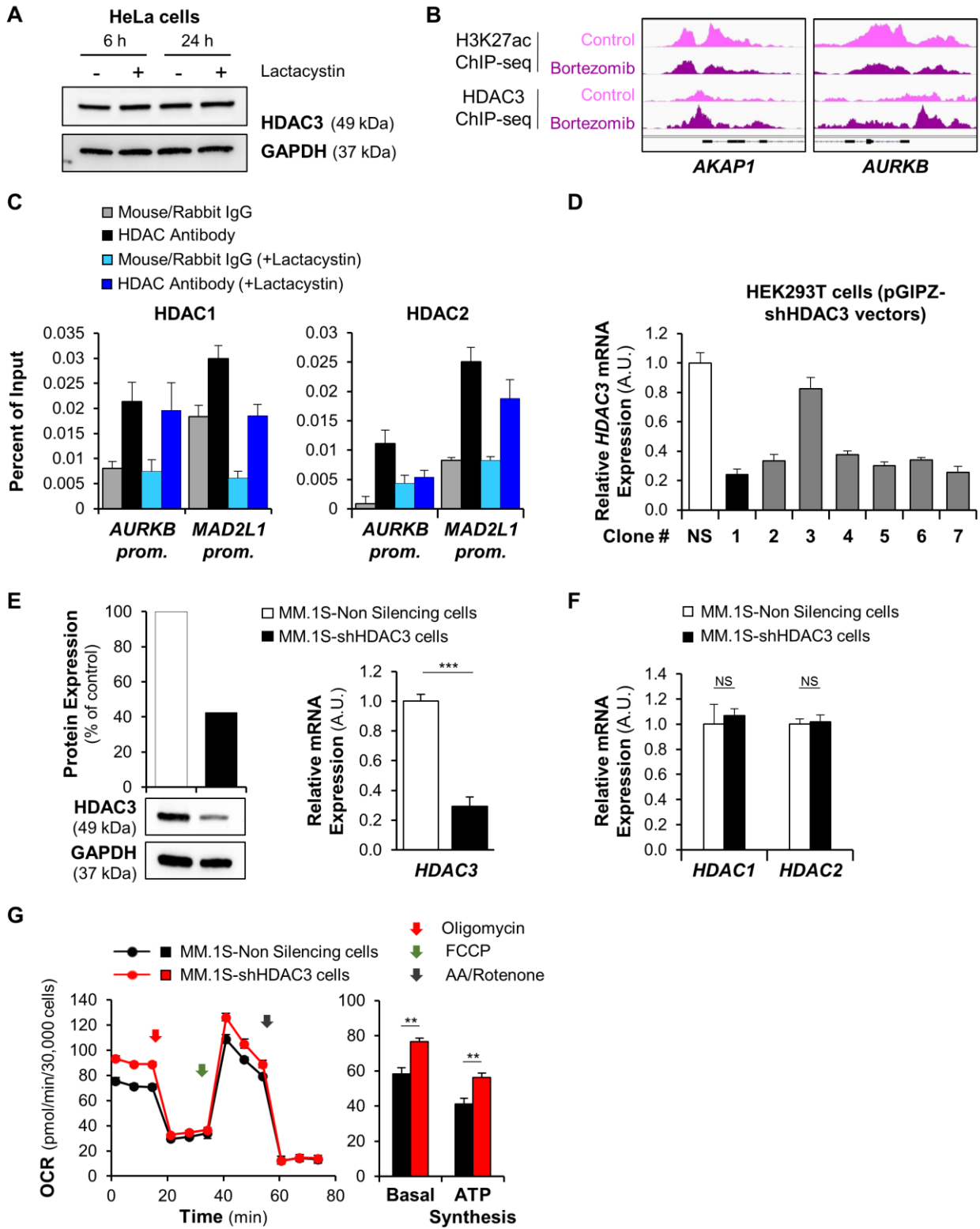


Figure S4



**Figure S4. DNA site-specific stabilization of HDAC3 by proteasome inhibition in MM cells.**

**A**, Western blot analysis of HDAC3 expression levels in HeLa cells after treatment with 6  $\mu$ M or 0.5  $\mu$ M lactacystin for 6 h or 24 h, respectively. GAPDH was used as an internal control.

**B**, ChIP-seq tracks of H3K27ac and HDAC3 occupancy on *AKAP1* and *AURKB* promoters following treatment of MM.1S cells with bortezomib. The gene structure is shown in black at the bottom of each panel. IGV browser images are representative of two independent experiments.

**C**, Proteasome inhibition does not modify the abundance of HDAC1 and HDAC2 at the promoters of cell cycle genes. ChIP-qPCR analysis of HDAC1 and HDAC2 DNA occupancy at selected promoters following proteasome inhibition with lactacystin in MM.1S cells. The presented data were the result of one experiment.

**D**, Screening for effective HDAC3 knockdown clones in HEK293T cells. HDAC3 expression levels were measured by RT-qPCR to determine knockdown efficiency. Clone#1 showed the best knockdown efficiency and was selected for further knockdown experiments. Clone #7 was excluded due to cell toxicity. The Open Biosystems nomenclature and clone sequences were the following:

Clone#1: Open Biosystems Clone ID#200204565, Sequence 5'- GCCTGACAATGGTACCTAT-3'

Clone#2: Open Biosystems Clone ID#200222927, Sequence 5'- ATAGCTATCTGGGACATTA-3'

Clone#3: Open Biosystems Clone ID#200222927, Sequence 5'- ATAGCTATCTGGGACATTA-3'

Clone#4: Open Biosystems Clone ID#200222927, Sequence 5'- ATAGCTATCTGGGACATTA-3'

Clone#5: Open Biosystems Clone ID#200243533, Sequence 5'- AACCAGGTAGTGGACTTCT-3'

Clone#6: Open Biosystems Clone ID#200247565, Sequence 5'- CCACTGTTTTTATCCAATA-3'

Clone#7: Open Biosystems Clone ID#200251116, Sequence 5'- AGAGCTTCAATATCCCTCT-3'

**E**, Validation of HDAC3 knockdown in MM.1S cells (Clone#1). HDAC3 expression levels were measured by Western blot (left panel) or RT-qPCR (right panel) to determine knockdown efficiency. The relative expression of HDAC3 protein was quantified in Image J and normalized to that of GAPDH protein. \*\*\* $p < 0.001$  determined by unpaired Student's two-tailed t-test.

**F**, RT-qPCR analysis of HDAC1 and HDAC2 transcripts after 6 h treatment of MM.1S cells with 6  $\mu$ M lactacystin. Inhibition of the proteasome activity did not modulate the mRNA expression levels of HDAC1 and HDAC2 genes. NS, not significant determined by unpaired Student's two-tailed t-test.

**G**, Measurement of mitochondrial respiration in MM.1S-shHDAC3 knockdown cells. Basal OCR values and ATP synthesis were measured as described in the **Fig. S3D** legend. \*\* $p < 0.01$  determined by unpaired Student's two-tailed t-test.