

Figure S6

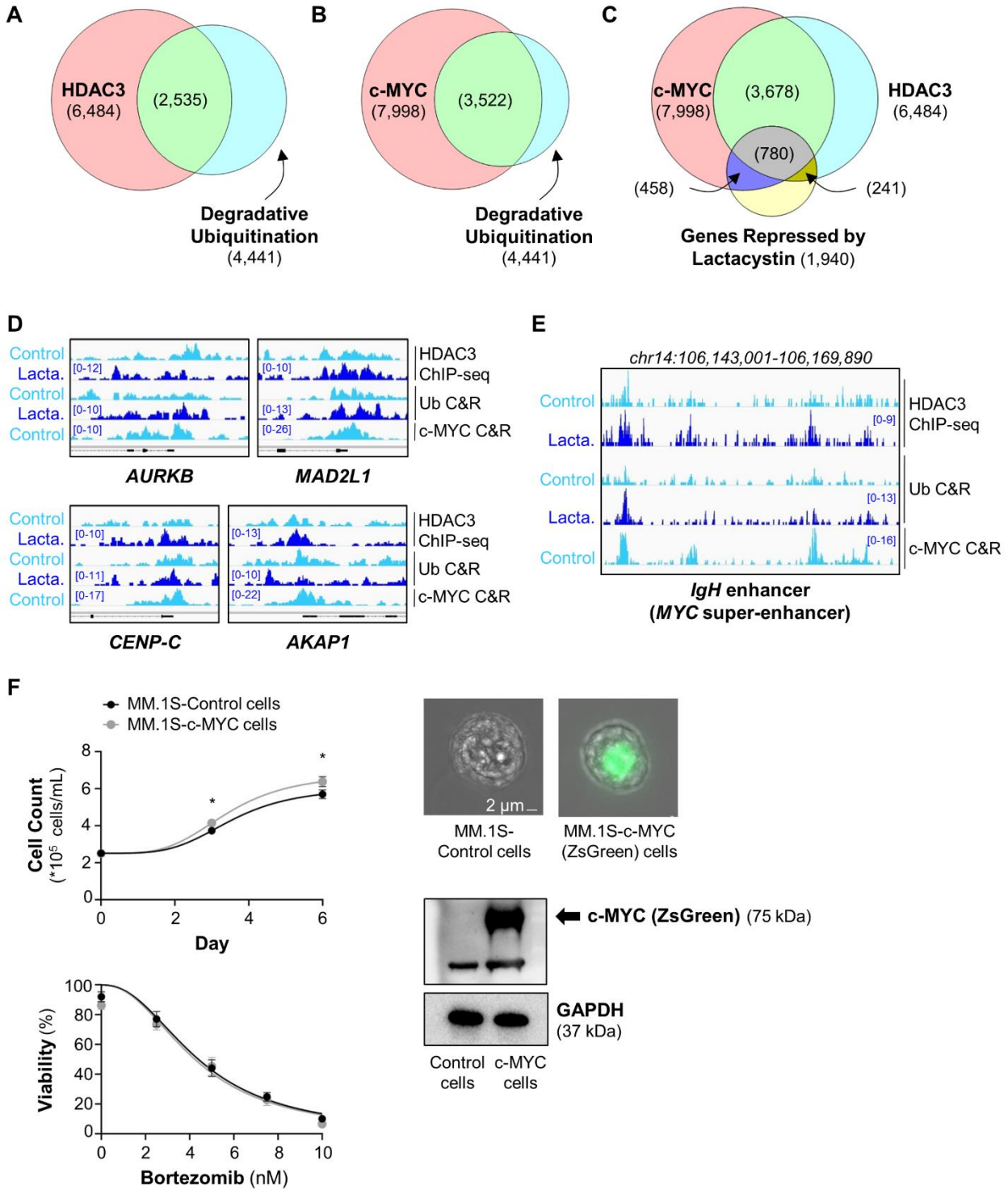


Figure S6. Association of c-MYC and HDAC3 DNA occupancy with protein turnover at the promoters of c-MYC target genes and at the c-MYC super-enhancer.

A, Venn diagram showing the significant overlap between HDAC3-associated sites and degradative ubiquitination sites within 1 kb of transcription start site after treatment of MM.1S cells with lactacystin.

B, Venn diagram showing the 79.8% overlap between c-MYC binding sites and degradative ubiquitination peaks within 1 kb of transcription start site after treatment of MM.1S cells with lactacystin.

C, Venn diagram demonstrating significant overlap in c-MYC binding sites, HDAC3-associated sites and gene transcripts repressed by proteasome inhibition within 1 kb of transcription start site in MM.1S cells.

D, Representative IGV tracks of ubiquitin (Ub) binding sites, HDAC3 and c-MYC DNA occupancy show that protein turnover associates with HDAC3 DNA occupancy and c-MYC binding at the promoters of cell cycle and mitochondrial genes following lactacystin exposure of MM.1S cells. All IGV tracks in a given comparison are represented at the same scale (numbers in brackets at the y-axis). The gene structure is shown in black at the bottom of each panel. The genomic region on the x-axis spans 2.5 kb for all the regions. HDAC3 ChIP-seq snapshots are representative of two independent experiments and ubiquitin and c-MYC CUT & RUN tracks were the result of a single experiment. C&R, CUT & RUN; Lacta., Lactacystin.

E, Gene tracks of ubiquitin (Ub) binding sites, HDAC3 and c-MYC DNA occupancy show that protein turnover associates with HDAC3 DNA occupancy and c-MYC binding at the c-MYC super-enhancer following lactacystin exposure of MM.1S cells. All IGV tracks in a given comparison are represented at the same scale (numbers in brackets at the y-axis). The genomic region on the x-axis spans 25 kb of the c-MYC super-enhancer. HDAC3 ChIP-seq snapshots are representative of two independent experiments and ubiquitin and c-MYC CUT & RUN tracks were the result of one experiment. C&R, CUT & RUN; Lacta., Lactacystin.

F, Cell growth curves show that c-MYC (ZsGreen)-overexpressing MM.1S cells proliferate faster than control cells over a period of 6 days (upper left panel). However, c-MYC overexpression was not sufficient to overcome proteasome inhibition-mediated cell death (bottom left panel). Nuclear c-MYC expression was confirmed by microscopy (N-terminal ZsGreen fluorophore) and total c-MYC expression was an estimated 3.1-fold higher than in control MM.1S cells based on densitometry (right panels). * $p < 0.05$, MYC group compared to control group, determined by unpaired Student's two-tailed t-test.