Potential preclinical activity of the EP300/CBP bromodomain inhibitor CCS1477/inobrodib in multiple myeloma

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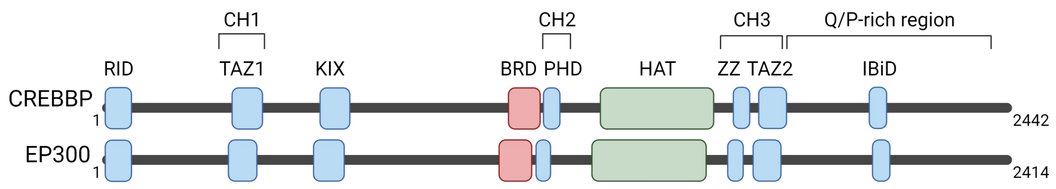
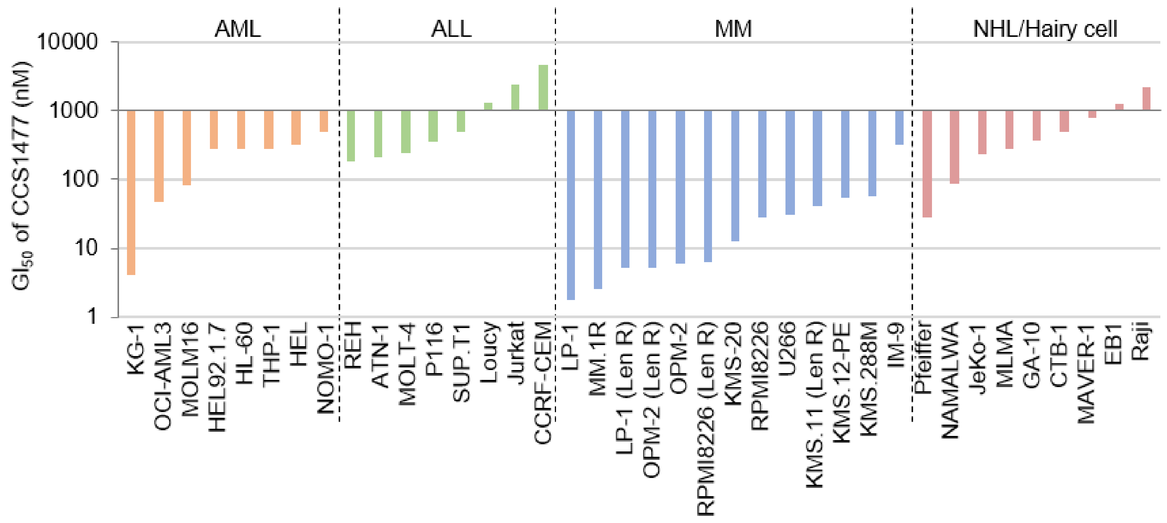
Introduction

- EP300/CBP are two paralogous acetyltransferases involved in transcriptional activation.
- EP300/CBP act as co-activators of cancer associated transcription factors including IRF4, MYC, MYB, and androgen receptor.
- EP300/CBP are attractive therapeutic targets in cancer in view of their critical role in promoting cellular growth and cell cycle progression.
- In haematological malignancies, there is ample evidence that pharmacologic targeting of the EP300/CBP bromodomain may be a useful therapeutic strategy.



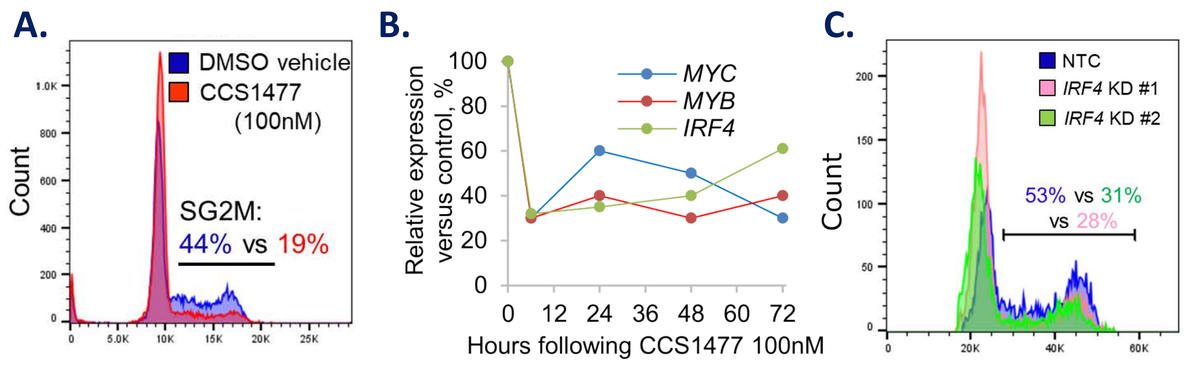
- CCS1477 (inobrodib) is a first-in-class oral EP300/CBP bromodomain inhibitor currently in early phase clinical trials.
- Potent and highly selective versus other bromodomains.

Result 1: Sensitivity of haematological models to CCS1477



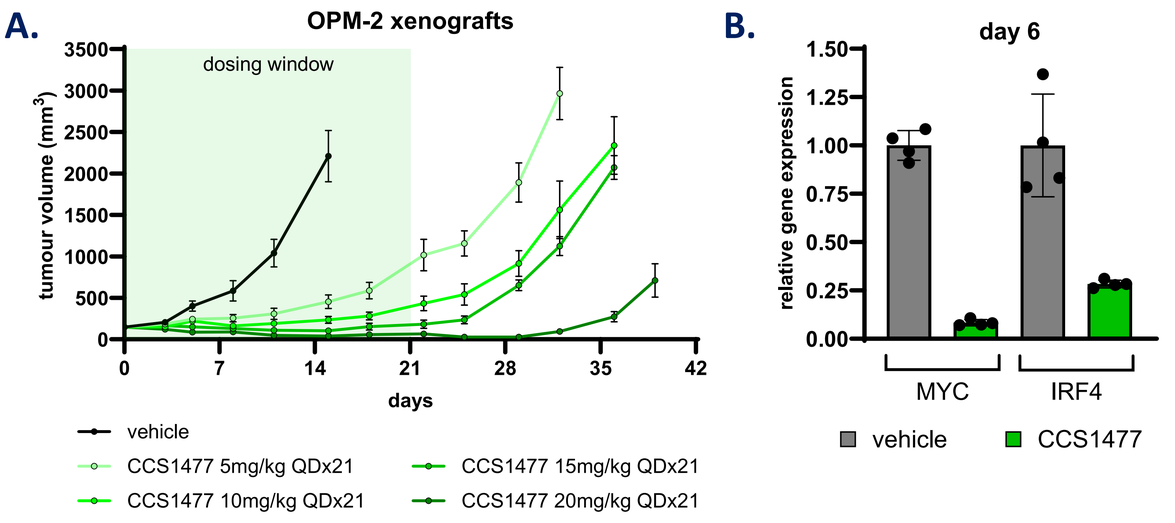
A panel of hematological cell lines grown in duplicate were exposed to a nine-point dosing of CCS1477 for 5 days and assessed by CellTiter-Glo to establish GI_{50} values. AML, acute myeloid leukemia; ALL, acute lymphoblastic leukemia; MM, multiple myeloma; NHL, non-Hodgkin lymphoma; Len R, cells resistant to lenalidomide.

Result 2: CCS1477 targets IRF4 to induce a cell cycle arrest



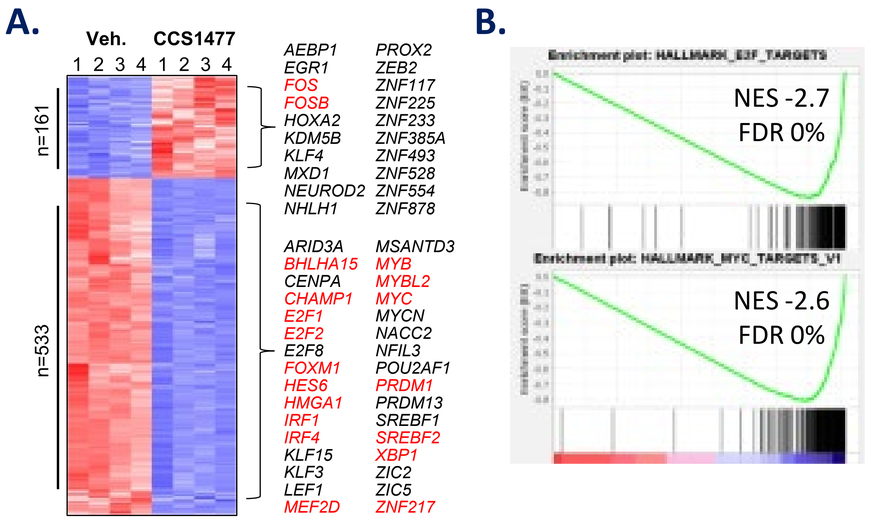
A) OPM2 cells exposed to 100nM CCS1477 were stained with propidium iodide and assessed via flow cytometry to determine cell cycle profiles. **B**) Indicated genes were quantified (relative to GAPDH) by qRTPCR at indicated timepoints in OPM2 cells treated with 100nM CCS1477. **C**) OPM2 cells were infected for 72 hours with lentiviruses targeting IRF4 (70-80% knockdown, data not shown) or a non-targeting control and the cell cycle was profiled as described above.

Result 3: CCS1477 elicits dose-dependent efficacy in vivo



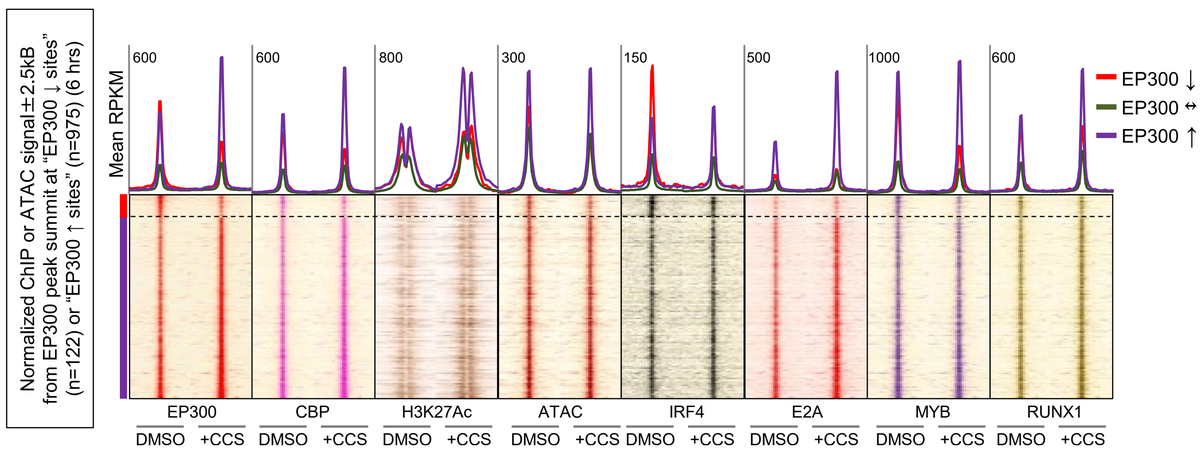
A) Mice harbouring OPM2 xenografts were treated with indicated doses of CCS1477 for 3 weeks and tumour dimensions were quantified every 3 days. B) Tumour RNA from vehicle- and CCS1477-treated mice was subjected to RNAseq profiling and relative expression of MYC and IRF4 was quantified.

Result 4: CCS1477 alters key transcriptional programs in multiple myeloma



A) Heatmap of differentially expressed genes in OPM2 xenograft tumours after six days of treatment with 20mg/kg CCS1477. Transcription factor genes are highlighted; those in red have high expression values, among the top 6.5% of all protein coding genes (i.e. ≥6 FPKM). **B**) Gene set enrichment plots. **C**) Immunoblot of indicated proteins following treatment of OPM2 cells with 100nM CCS1477 for the indicated times.

Result 5: CCS1477 induces bromodomain-dependent EP300/CBP genomic redistribution



ATAC or ChIPseq signal ±2.5kB from the summit of EP300/CBP binding peaks identified in OPM2 cells treated for 6 hours with 100nM CCS1477 for the indicated factors in the indicated conditions. EP300 binding sites are grouped in ranked order of loss ("EP300 ↓" sites) or gain ("EP300 ↑" sites) of EP300 ChIP signal from those sites after 6 hours of 100nM CCS1477 treatment. Graphs above heatmaps show mean ChIP signal for the indicated peak group.

Conclusions

- CCS1477/inobrodib demonstrates promising monotherapy efficacy in preclinical models of multiple myeloma.
- CCS1477 targets key oncogenic drivers, including IRF4 and MYC, to elicit a potent anti-proliferative effect.
- CCS1477-mediated transcriptional rewiring is associated with genomic redistribution of EP300/CBP away from IRF4-occupied sites.
- These data support the inclusion of myeloma patients in our ongoing phase I/IIa clinical trial (NCT04068597).