Intermittent Treatment of BRAFV600E Melanoma Cells Delays Resistance

Melanoma patients receiving drugs targeting BRAFV600E and MEK1/2 invariably develop resistance and continue progression. Based on preclinical studies, intermittent treatment involving alternating periods of drug challenge and withdrawal has been proposed as a method to delay the onset of resistance. The beneficial effect of intermittent treatment has been attributed to drug addiction, where drug withdrawal reduces the viability of resistant cells due to MAP kinase pathway hyperactivation. However, the mechanistic basis of the intermittent effect is incompletely understood. We show that intermittent treatment with the BRAFV600E inhibitor, LGX818/encorafenib, suppresses growth compared to continuous treatment in human melanoma cells engineered to express BRAFV600E. Analysis of the BRAFV600E-overexpressing cells shows that growth suppression in an intermittent treatment schedule is best explained by resensitization of cells during periods of drug removal rather than drug addiction. Cells treated intermittently reveal a subset of transcripts that exhibit reversible transcriptional profiles, and include mediators of cell invasiveness and the epithelial to mesenchymal transition. These transcripts change during periods of drug removal by adaptive switching, rather than selection pressure. Resensitization occurs against a background of sustained expression of melanoma resistance genes, producing a transcriptome distinct from that of the initial drug-naive cell state. We conclude that phenotypic plasticity leading to drug resensitization can underlie the beneficial effect of intermittent treatment.

### Impact and Future Directions

- Previously described benefits of intermittent drug treatment in melanoma can be modeled in vitro
- Intermittent drug treatment produces a unique transcriptional profile
- Transcriptional adaptation can be associated with intermittent drug treatment
- Identifying genes involved in resensitization of cells may help identify therapeutic targets to combat resistance
- Further use of an in vitro system to model optimal schedules of intermittent treatment may identify additional targets

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