

## Supplementary Data

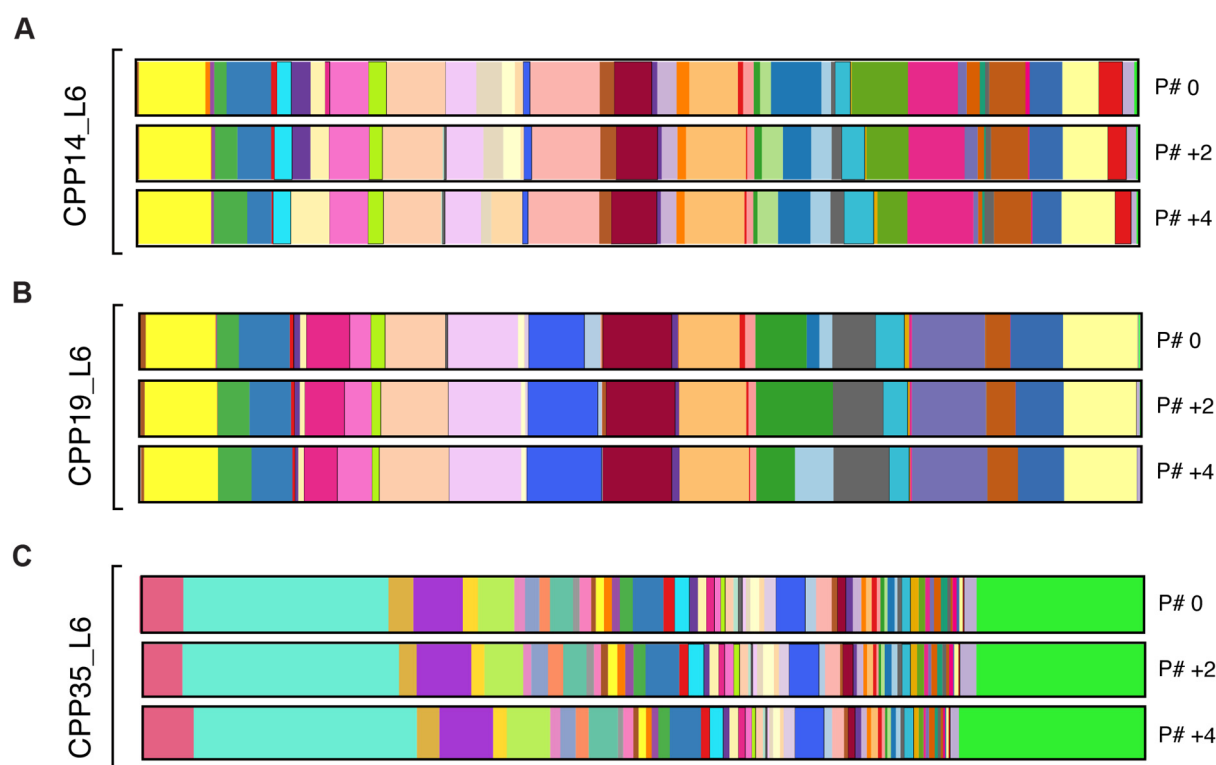
Table S1 | Doubling time estimation of wildtype and optically barcoded cell lines

Cell Line	Variant	Doubling time (hours) ± 95% CI
CPP14	WT	31.7 ± 8.4
	L6	29.8 ± 9.6
CPP19	WT	39.7 ± 8.7
	L6	36.9 ± 4.9
CPP35	WT	40.3 ± 15.9
	L6	39.5 ± 12.05

Table S2 | Profiling of MEK-inhibitor responses in wildtype and optically barcoded cell lines

Cell Line	Variant	Drug	LogIC <sub>50</sub> ± 95% CI	Fold Change
CPP14	WT	Cobimetinib	-6.90 ± 0.56	0.95
	L6		-7.28 ± 0.30	
	WT	Selumetinib	-6.66 ± 0.62	0.94
	L6		-7.10 ± 0.46	
CPP19	WT	Cobimetinib	-7.24 ± 0.54	0.99
	L6		-7.29 ± 0.63	
	WT	Selumetinib	-7.33 ± 0.6	1.02
	L6		-7.22 ± 0.72	
CPP35	WT	Cobimetinib	-6.70 ± 0.84	1.06
	L6		-6.35 ± 0.98	
	WT	Selumetinib	-5.69 <sup>^</sup>	1.34
	L6		-4.23 <sup>^</sup>	

<sup>^</sup> 95% CI unable to be determined as 50% cell death not achieved at this dose range



**Figure S1 | Optical barcoding of 2D CRC cell lines show limited change in cluster distribution upon serial passaging**

Barcharts illustrating cluster frequencies following serial flow cytometric analysis of three optically barcoded CRC cell lines; **(A)** CPP14-L6 **(B)** CPP19-L6 and **(C)** CPP35-L6.