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Reporting Summary

Nature Research wishes to improve the reproducibility of the work that we publish. This form provides structure for consistency and transparency in reporting. For further information on Nature Research policies, see our Editorial Policies and the Editorial Policy Checklist.

Statistics

For all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.

n/a	Confirmed	
☐	☒	The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement
☐	☒	A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☐	☒	The statistical test(s) used AND whether they are one- or two-sided *Only common tests should be described solely by name; describe more complex techniques in the Methods section.*
☒	☐	A description of all covariates tested
☒	☐	A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
☐	☒	A full description of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
☐	☒	For null hypothesis testing, the test statistic (e.g. *F*, *t*, *r*) with confidence intervals, effect sizes, degrees of freedom and *P* value noted *Give P values as exact values whenever suitable.*
☒	☐	For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
☒	☐	For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
☐	☒	Estimates of effect sizes (e.g. Cohen's *d*, Pearson's *r*), indicating how they were calculated

Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection

Paired-end HiC sequencing reads were mapped using BWA-MEM to the reference genome (hg19) in single-end mode with default parameter setting for each of the two ends separately.

For RNAseq, quality of sequencing reads was assessed using FastQC (Babraham Bioinformatics, version 0.11.2) and aligned to a reference genome (hg19, UCSC Genome Browser) using TopHat. Sequencing yielded on average 46.3 million reads per sample with a ~71% mapping rate to generate ~33 million reads per sample uniquely mapped to the reference genome.

For whole genome sequencing, mapping to the human genome was performed using the BWA algorithm BWA-MEM (Version 0.7.8) and genome build hs37d5.

Data analysis

For HiC sequencing, the independently mapped ends were then paired-up and the read pairs were kept if both ends uniquely mapped to the genome (MQAL>10). Next, read pairs were further removed if either end was mapped more than 500bp apart away from the closest MboI cutter site. Read pairs were next sorted based on genomic coordinates followed by PCR duplicate removal using samtools rmdup. Processed Hi-C data is converted to .hic file and is visualized using juicebox (version 1.5.2) without normalization.

For RNAseq, Cufflinks was used to generate transcript abundance as fragments per kilobase of transcript per million mapped reads (FPKM), and statistical analysis of FPKM values was calculated using R (version 3.5.0).

For whole genome sequencing, copy number was called using the ascatNgs algorithm. The variant calling pipeline of the Cancer Genome Project, Wellcome Sanger Institute was used to call somatic mutations. The following algorithms with standard settings and no additional post-processing were used: CaVEMan for substitutions and the BRASS algorithm for rearrangements.

Statistical analyses were performed with GraphPad Prism software (version 4.0)
Live imaging software: CQ1 software version 1.05.01.02

ImageJ version 1.51f used for image analysis
FACS Diva version 8.0.1 for flow cytometry analysis

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Research guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a data availability statement. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A list of figures that have associated raw data
- A description of any restrictions on data availability

Data reported in this study are available upon request.

Paired-end whole genome sequencing data is available at ENA (European Nucleotide Archive), accession number ERP107458 https://www.ebi.ac.uk/ena/data/view/PRJEB25535

In-situ Hi-C sequencing data is available at GEO (Gene Expression Omnibus), accession number GSE119825..

RNA sequencing data is available at GEO (Gene Expression Omnibus), accession number GSE119979.

TCGA database can be accessed in https://portal.gdc.cancer.gov/

Field-specific reporting

Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

☒ Life sciences ☐ Behavioural & social sciences ☐ Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see nature.com/documents/nr-reporting-summary-flat.pdf

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	Sample sizes were chosen based upon previous standards in the field.
Data exclusions	No data was excluded from analysis.
Replication	All experiments were independently conducted and reproduced multiple times, as described in the figure legends. All P-values were derived from measurements obtained from experiments conducted independently at least three times.
Randomization	No experiments requiring randomization were performed (e.g. animal experiments). Human samples analyzed were chosen due to their amplification of BRAF.
Blinding	Whole genome sequencing was done blinded (using barcoded tubes). Data was not blinded for other experiments as per the standards in the field.

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

Materials & experimental systems

n/a	Involved in the study
☐	☒ Antibodies
☐	☒ Eukaryotic cell lines
☒	☐ Palaeontology and archaeology
☒	☐ Animals and other organisms
☐	☒ Human research participants
☒	☐ Clinical data
☒	☐ Dual use research of concern

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☐	☒ Flow cytometry
☒	☐ MRI-based neuroimaging

Antibodies

Antibodies used: Aurora B (Abcam, ab2254), Lamin B (Proteintech, 12987-1-AP).

Validation: Validation was done using immunofluorescence showing the expected cell localization and appearance.

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s):
HeLa S3 cervical adenocarcinoma cancer cells (Cleveland lab stock)
Colo320DM-GFP colorectal adenocarcinoma containing double minute labeled with GFP (gift from Prof. Noriaki Shimizu)
HT-29 colorectal cancer cells (ATCC® HTB-38™)
293T Human embryonic kidney cells (Cleveland lab stock)
DLD-1 (ATCC)
T-47D (ATCC)

Authentication:
Cells purchased through ATCC were not authenticated.
HeLa cells authenticated using our sequencing and cytogenetic analysis (showing identical genomic events to previous reports)
293T cells authenticated by their culture behavior (loosely attached), morphology under the microscope, and by their efficiency in transfection and virus production.

Mycoplasma contamination:
All cell lines used in this study were confirmed to be free of mycoplasma contamination, by routine (every 6 months) testing and by frequent DAPI staining.

Commonly misidentified lines (See ICLAC register):
No commonly misidentified cell lines were used in this study

Human research participants

Policy information about studies involving human research participants

Population characteristics:
Patient data was previously reported in: Yaeger, R. et al. Mechanisms of Acquired Resistance to BRAF V600E Inhibition in Colon Cancers Converge on RAF Dimerization and Are Sensitive to Its Inhibition. Cancer Res 77, 6513-6523, doi:10.1158/0008-5472.CAN-17-0768 (2017).

Recruitment:
Patient data was previously reported in: Yaeger, R. et al. Mechanisms of Acquired Resistance to BRAF V600E Inhibition in Colon Cancers Converge on RAF Dimerization and Are Sensitive to Its Inhibition. Cancer Res 77, 6513-6523, doi:10.1158/0008-5472.CAN-17-0768 (2017).

Ethics oversight:
Memorial Sloan Kettering (MSK) Cancer Center Institutional Review Board/Privacy Board biospecimen protocol 19-323 was used to analyze samples. Patients provided consent and signed MSK's tissue banking protocol (06-107). Samples were assigned a sample number with no patient identifying information before submission for DNA extraction and genomic analysis. No one performing genomic analysis had access to the patients' identifying information.

Note that full information on the approval of the study protocol must also be provided in the manuscript.

Flow Cytometry

Plots

Confirm that:

☒ The axis labels state the marker and fluorochrome used (e.g. CD4-FITC).

☒ The axis scales are clearly visible. Include numbers along axes only for bottom left plot of group (a 'group' is an analysis of identical markers).

☒ All plots are contour plots with outliers or pseudocolor plots.

☒ A numerical value for number of cells or percentage (with statistics) is provided.

Methodology

Sample preparation	Non synchronized HeLa cells, or mitotic HeLa cells (obtained by mitotic shake-off following 4 hours treatment with 0.1ug/ml nocodazole) were ethanol fixed, stained with 10µg/ml propidium iodide, and treated with 50µg/ml RNase A.
Instrument	BD LSR II instrument (BD Biosciences)
Software	BD cellquest (version 5.1)
Cell population abundance	10,000 cells were analyzed. Non synchronized cells contained 20% G2/M cells. Mitotic enriched samples contained 75-88% G2/M cells.
Gating strategy	Dead cells were excluded using SSC/FSC gating. G1, S, and G2/M populations were gated using a histogram plot as shown in the figures.

☒ Tick this box to confirm that a figure exemplifying the gating strategy is provided in the Supplementary Information.