nature portfolio

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Reporting Summary

Nature Portfolio 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 Portfolio policies, see our <u>Editorial Policies</u> and the <u>Editorial Policy Checklist</u>.

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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
	\square 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. <i>F</i> , <i>t</i> , <i>r</i>) with confidence intervals, effect sizes, degrees of freedom and <i>P</i> value noted <i>Give P values as exact values whenever suitable.</i>
\boxtimes	For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
\boxtimes	For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
\boxtimes	Estimates of effect sizes (e.g. Cohen's <i>d</i> , Pearson's <i>r</i>), 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

Image data were visualised and analysed in Leica LAS X software (Version 3.7.2.22383) and FIJI (ImageJ 2.00-rc-59/1.53c). Movies were produced in Imaris 9.0.0 (Bitplane AG).

Data analysis

Statistical analysis was carried out using GraphPad Prism 9.0. Depending on different experiments, ordinary one-way ANOVA and Tukey's multiple comparisons test, and two-way ANOVA together with Dunnett's multiple comparisons test were used.

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 Portfolio <u>guidelines for submitting code & software</u> for further information.

Data

Policy information about availability of data

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

- Accession codes, unique identifiers, or web links for publicly available datasets
- A description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our policy

Microscopy data reported in this paper and any information required to re-analyse the data reported in this paper are presented in the Extended Figures or are available from the lead contact upon reasonable request.

Human rese	arch part	icipants			
Policy information	about <u>studies</u>	involving human research participants and Sex and Gender in Research.			
Reporting on sex	and gender	N/A			
Population chara	cteristics	N/A			
Recruitment		N/A			
Ethics oversight		N/A			
Note that full informa	ation on the app	proval of the study protocol must also be provided in the manuscript.			
Field spe	ocific ra	phorting			
Field-spe		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			
		h all sections, see nature.com/documents/nr-reporting-summary-flat.pdf			
Life scier	nces st	udy design			
		e points even when the disclosure is negative.			
Sample size	Sample sizes r figure legends	may be chosen by using a target of power of a statistical test to be applied once the sample is collected and described in the			
Data exclusions	No data were	ata were excluded from the analyses.			
Replication		experiments were done as three independent experiments with three biological replicates analysed. All attempts correctly executed were used. All details were described in the figure legends.			
Randomization	Randomization involved selection of random samples within a experimental /treatment group by the investigators. Usually 10 randomly selected samples (i.e. cells, zebrafish embryos) were chosen and the parameters analysed. The investigators choosing the samples did not know about the particular treatment.				
Blinding	Samples were chosen randomly from a group without giving the investigators access to the treatment protocol.				
Reportin	g for s	pecific materials, systems and methods			
		s about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, o 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 Methods					
n/a Involved in th	n/a Involved in the study				
Antibodies					
Eukaryotic cell lines Flow cytometry Palaeontology and archaeology MRI-based neuroimaging					
Animals and other organisms					
☐ Clinical data					
Dual use research of concern					
Antibodies					

Antibodies used WNT5A-B, rabbit PolyAb, ProteinTech, 55184-1-AP, 1:50; ROR2 (D3B6F), rabbit mAb, Cell Signalling Technology, 88639S, 1:50; antirabbit antibody Alexafluor 488, ab150077, Abcam, 1:1000; donkey anti-goat antibody Alexafluor 647, ab150135, Abcam, 1:1000

Validation All primary antibodies have been validates by IHC and WB as stated in the manuscript.

Eukaryotic cell lines

Policy information about <u>cell lines and Sex and Gender in Research</u>

PAC2 zebrafish fibroblasts provided by Nicholas Foulkes (KIT, Germany) and Reinhard Koester (TU Braunschweig, Germany) -Cell line source(s)

No commercial source. AGS (Gastric cancer cells)- American Tissue Culture Collection, ATTC, Wesel, Germany

Authentication None of the cell lines were authenticated.

Mycoplasma contamination The cell line was tested for Mycoplasma contamination every three months and confirmed mycoplasma-free - overseen by experimental officer Dr Francesca Carlie, LSI Tissue Culture Facility.

Commonly misidentified lines (See ICLAC register)

No commonly misidentified cell lines were used in this study.

Animals and other research organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research, and Sex and Gender in Research

Laboratory animals zebrafish, strain: WIK, sex: male/female, age: 8-18months

Wild animals No wild animals were used in the study.

Reporting on sex N/A

Field-collected samples

No field collected samples were used in the study.

Ethics oversight

WIK wild-type, Tg(-6gsc: EGFP -CAAX), and Ror2 (T13fs38X) zebrafish (Danio rerio) were maintained as previously described at 28°C and on a 14hr light/10hr dark cycle. Zebrafish care and all experimental procedures were carried out in accordance with the European Communities Council Directive (2010/63/EU) and Animals Scientific Procedures Act (ASPA) 1986. In detail, adult zebrafish for breeding were kept and handled according to the ASPA animal care regulations and all embryo experiments were performed before 120h post fertilization. Zebrafish experimental procedures were carried out under personal and project licenses granted by the UK Home Office under ASPA. The project has been ethically approved by the Animal Welfare and Ethical Review Body at the University of Exeter.

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