Researchers turn back the clock on cancer cells to offer new treatment paradigm

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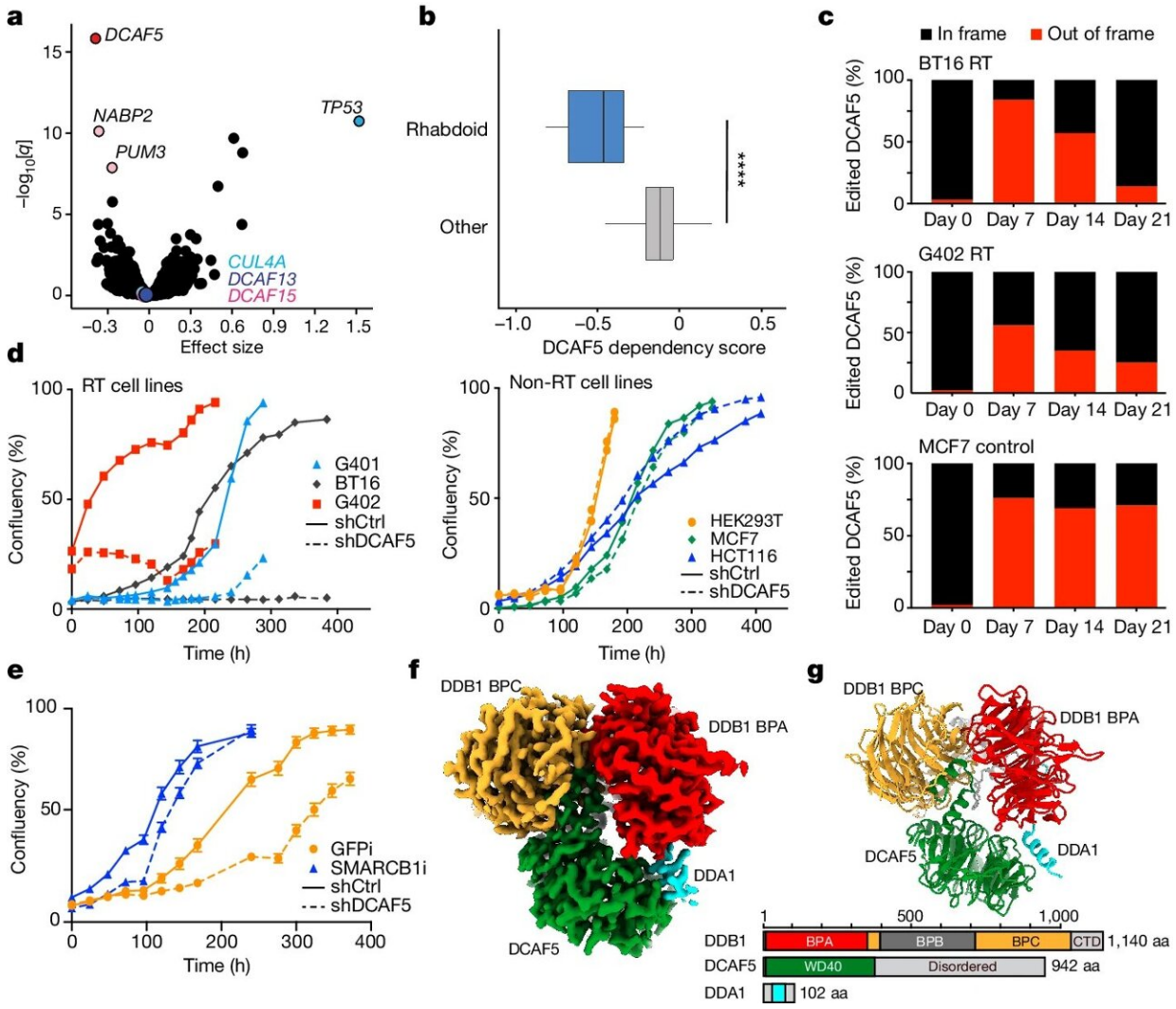

DCAF5 is a specific dependency in SMARCB1-mutant cancers. **a**, Comparison of $n = 14$ biologically independent RT cell lines to $n = 789$ biologically independent other cancer cell lines from DepMap (release CERES 21Q1). Each

circle represents a single gene. A negative effect size indicates that RT cells are preferentially dependent on that gene. $-\log_{10}[q]$ was calculated from empirical-Bayes-moderated *t*-statistics with Benjamini–Hochberg correction. **b**, Two-class comparison of $n = 14$ biologically independent RT cell lines to $n = 789$ biologically independent other cancer cell lines. Statistical analysis was performed using a two-tailed Student's *t*-test; $^{****}P = 8.21 \times 10^{-21}$. Release CERES 21Q1. The box plot shows the median (center line), the third and first quartiles (box limits) and 1.5 × interquartile range above and below the box (whiskers). **c**, Indel toxicity assay. *DCAF5* was targeted with a CRISPR guide to generate mutations. Then, selective pressure against out-of-frame mutations (containing *DCAF5* knockout) was measured over time in BT16 and G402 RT cells and control MCF7 cells. **d**, The effects of *DCAF5* shRNA knockdown on the proliferation of *SMARCB1*-mutant cell lines or *SMARCB1*-expressing control cell lines. The solid lines show shCtrl and the dotted lines show shDCAF5. Data are mean values from $n = 8$ technical replicates per cell line condition from one independent experiment. **e**, The proliferation of *SMARCB1*-knockout HEK293T cells after knockdown of *DCAF5* and re-expression of SMARCB1 or GFP (control). The solid lines show shCtrl and the dotted lines show shDCAF5. Data are mean values from $n = 16$ technical replicates per cell line condition from one independent experiment. **f**, Cryo-EM map (post-processed using deepEMhancer) of the DCAF5–DDB1(ΔB)–DDA1 complex segmented to indicate DDA1 (cyan), DCAF5 (green), DDB1 BPC (orange), DDB1 BPA (red) and DDB1 C-terminal domain (gray). **g**, Cartoon representation of the DCAF5–DDB1(ΔB)–DDA1 complex. Domain representation of the proteins present in the complex. Regions omitted from the constructs (BPB) are indicated in dark gray. aa, amino acids; CTD, C-terminal domain. Credit: *Nature* (2024). DOI: 10.1038/s41586-024-07250-1

St. Jude Children's Research Hospital scientists reversed an aggressive cancer, reverting malignant cells towards a more normal state. Rhabdoid tumors are an aggressive cancer which is missing a key tumor suppressor protein. Findings showed that with the missing tumor suppressor, deleting or degrading the quality control protein DCAF5 reversed the cancer cell state.

These results suggest a new approach to curing cancer—returning cancerous cells to an earlier, more normal state rather than killing cancer cells with toxic therapies—may be possible. The results were published today in *Nature*.

"Rather than creating a toxic event that kills rhabdoid cancer, we were able to reverse the cancer state by returning the cells toward normal," said senior author Charles W.M. Roberts, MD, Ph.D., Executive Vice President and St. Jude Comprehensive Cancer Center director. This approach would be ideal, especially if this paradigm could also be applied to other cancers."

"We found a dependency which actually reverses the cancer state," said first author Sandi Radko-Juettner, Ph.D., a former St. Jude Graduate School of Biomedical Sciences student, now a Research Program Manager for the Hematological Malignancies Program at St. Jude. "Standard cancer therapies work by causing toxicities that also damage healthy cells in the body. Here, it appears that we're instead fixing the problem caused by the loss of a tumor suppressor in this rhabdoid cancer."

Drugging the un-targetable

In many cancers, there is no easily druggable target. Often, these cancers are caused by a missing tumor suppressor protein, so there is nothing to target directly as the protein is missing. Loss of tumor suppressors is much more common than a protein gaining the ability to drive cancer.

Consequently, finding a way to intervene therapeutically in these tumors is a high priority. The researchers were looking for a way to treat an aggressive set of cancers caused by the loss of the tumor suppressor protein SMARCB1 when they found a new approach to treatment.

The St. Jude group found a little-studied protein, DCAF5, was essential to rhabdoid tumors missing SMARCB1. Initially, they identified DCAF5 as a target, using the Dependency Map (DepMap) portal, a database of cancer cell lines and the genes critical for their growth. DCAF5 was a top dependency in rhabdoid tumors. After the initial finding, the scientists genetically deleted or chemically degraded DCAF5. The cancer cells reverted to a non-cancerous state, persisting even in a long-term mouse model.

"We saw a spectacular response," Roberts said. "The tumors melted away."

Removing quality control to reverse cancer

Normally, SMARCB1 is an essential component of a larger chromatin-regulating complex of proteins called the SWI/SNF complex. Unexpectedly, the study found that in the absence of SMARCB1, DCAF5 recognizes SWI/SNF as abnormal and destroys the complex.

When DCAF5 degrades them, the researchers showed that SWI/SNF re-forms and maintains its ability to open chromatin and regulate gene expression. While the SWI/SNF activity level in the absence of SMARCB1 was to a lesser extent than usual, it was nonetheless sufficient to reverse the cancer state fully.

"DCAF5 is doing a quality control check to ensure that these chromatin machines are built well," Roberts said. "Think of a factory assembling a machine. You need quality checks to examine and find faults and to pull it off the line if it doesn't meet standards. DCAF5 is doing such quality assessments for the assembly of SWI/SNF complexes, telling the cell to get rid of complexes if SMARCB1 is absent."

"The mutation of SMARCB1 shuts off gene programs that prevent

cancer. By targeting DCAF5, we're turning those gene programs back on," Radko-Juettner said. "We're reversing the cancer state because the cell is becoming more 'normal' when these complexes aren't targeted for destruction by DCAF5."

Future therapeutic opportunities to reverse cancer

"From a therapeutic perspective, our results are fascinating," Radko-Juettner said.

"DCAF5 is part of a larger family of DCAF proteins that have been shown to be drug targetable. We showed that when DCAF5 is absent, mice had no discernable health effects, so we could potentially target DCAF5. This can kill the cancer cells but shouldn't affect healthy cells. Targeting DCAF5 thus has the potential to avoid the off-target toxicity of radiation or chemotherapy, making it a promising therapeutic avenue to pursue."

Beyond DCAF5, the findings could have implications for other cancers driven by the loss of a tumor suppressor.

"We have demonstrated a beautiful proof of principle," Roberts said. "Myriad types of cancers are caused by tumor suppressor loss. We hope we may have opened the door to thinking about new ways to approach targeting at least some of these by reversing, instead of killing, cancer."

More information: Charles Roberts, Targeting DCAF5 suppresses SMARCB1-mutant cancer by stabilizing SWI/SNF, *Nature* (2024). DOI: 10.1038/s41586-024-07250-1.
www.nature.com/articles/s41586-024-07250-1

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