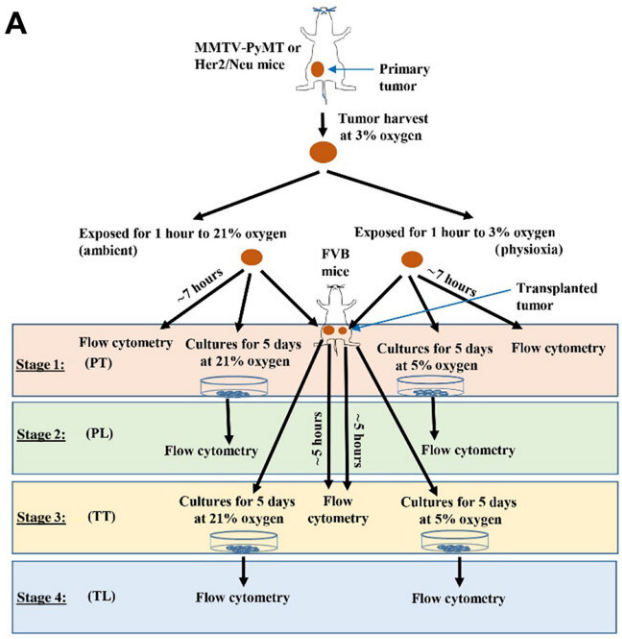
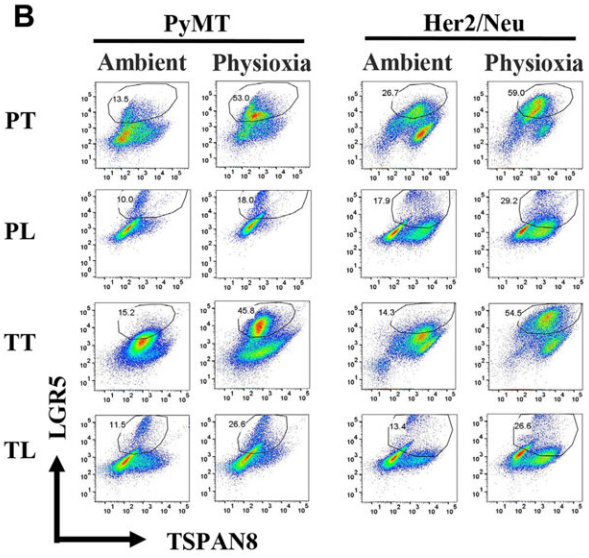
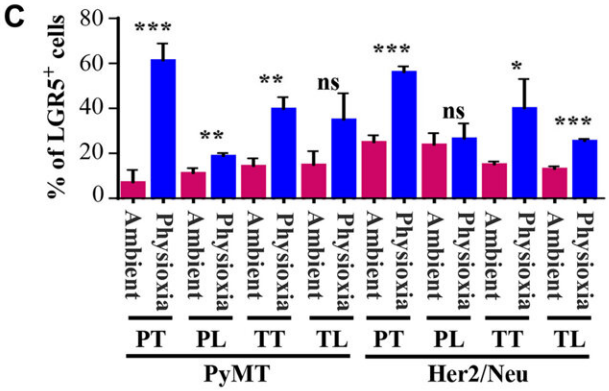
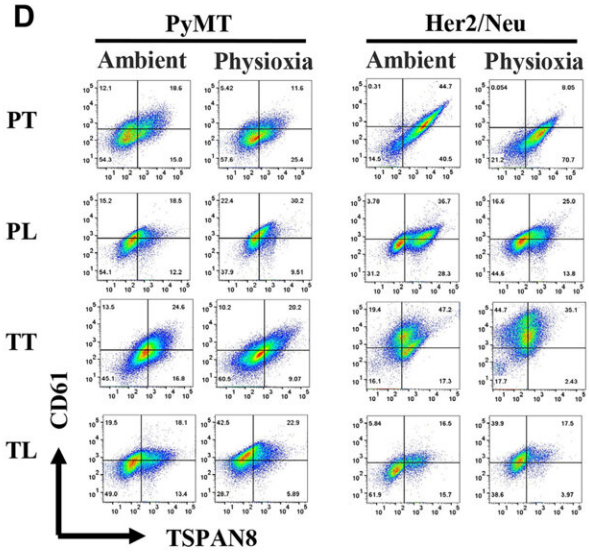
Important new findings about how to test cancer-fighting drugs

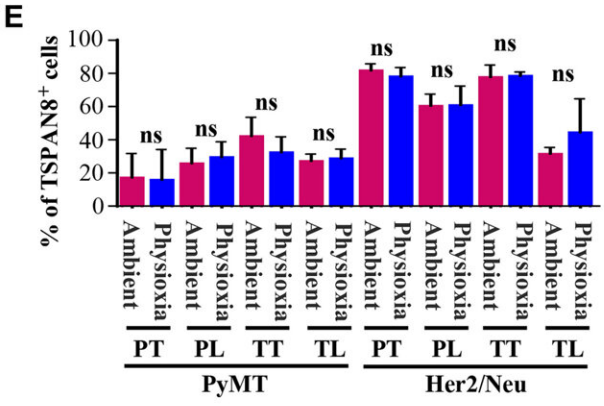
January 17 2022, by Christina Griffiths

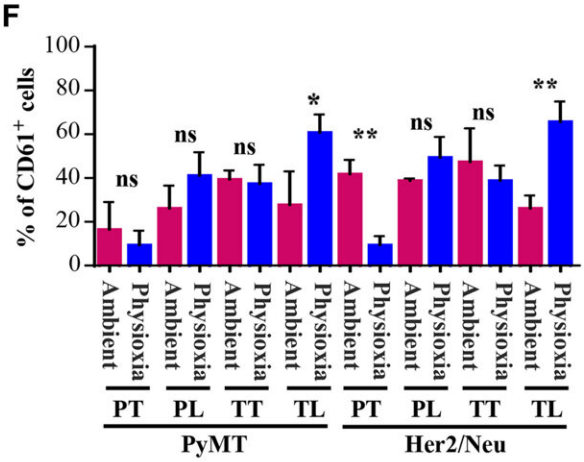

A
MMTV-PyMT or Her2/Neu mice
Primary tumor
Tumor harvest at 3% oxygen
Exposed for 1 hour to 21% oxygen (ambient)
Exposed for 1 hour to 3% oxygen (physioxia)
FVB mice
~7 hours
~7 hours
Transplanted tumor
Stage 1: (PT)
Flow cytometry
Cultures for 5 days at 21% oxygen
Cultures for 5 days at 5% oxygen
Flow cytometry
Stage 2: (PL)
Flow cytometry
~5 hours
~5 hours
Flow cytometry
Stage 3: (TT)
Cultures for 5 days at 21% oxygen
Flow cytometry
Cultures for 5 days at 5% oxygen
Stage 4: (TL)
Flow cytometry
Flow cytometry


B
PyMT
Her2/Neu
Ambient
Physioxia
PT
PL
TT
TL
LGR5
TSPAN8


C
% of LGR5+ cells
Ambient
Physioxia
PT
PL
TT
TL
PyMT
Her2/Neu


D
PyMT
Her2/Neu
Ambient
Physioxia
PT
PL
TT
TL
CD61
TSPAN8


E
% of TSPAN8+ cells
Ambient
Physioxia
PT
PL
TT
TL
PyMT
Her2/Neu


F
% of CD61+ cells
Ambient
Physioxia
PT
PL
TT
TL
PyMT
Her2/Neu

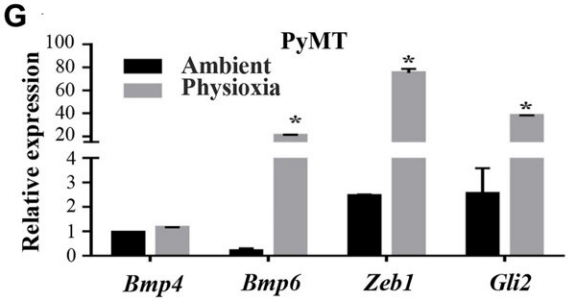

G
PyMT
Ambient
Physioxia
Relative expression
Bmp4
Bmp6
Zeb1
Gli2

Fig. 1. Effects of ephoss on csc-associated cell surface marker levels and stemness-associated gene expression. (A) Schematic view of the experimental design. Stage 1 (PT, primary tumor) involved processing of tumors under physioxia and ambient air, flow cytometry characterization of cells, cell propagation, and reimplantation of cells into female FVB/N mice. Stage 2 (PL, primary line) involved flow cytometry characterization of cultured cells of stage 1. Stage 3 (TT, Transplanted Tumor) involved recharacterization of tumors obtained from cell implantation of stage 1. Stage 4 (TL, transplanted line) involved flow cytometry characterization of cells grown from stage 3 tumor. PT, Primary Tumor; PL, Primary Line; TT, Transplanted Tumor; TL, Transplanted Line. (B) Representative flow cytometry profile of tumor cells stained for antibodies against LGR5 and TSPAN8. Antibodies against lineage markers CD31-PE (phycoerythrin)/Cy7, CD45-PE/Cy7, and CD140a-PE/Cy7 were used to label endothelial cells, hematopoietic cells, and fibroblasts, respectively, and only lineage-negative cells were included in the analysis. (C) Quantitation of LGR5+ cells. Differences in LGR5+ cells between ambient air and physioxia are significant [n = 3 to 6, one-way analysis of variance (ANOVA)]. (D) CD61/TSPAN8 staining patterns of tumor cells (n = 3 to 6, one-way ANOVA). (E) Quantitation of TSPAN8+ cells. (F) Quantitation of CD61+ cells. (G) Tumor cells collected and processed at physioxia express higher levels of stemness-associated genes compared to ambient air (n = 3, one-way ANOVA, P = 0.0004). *P

Researchers from Indiana University School of Medicine are discovering new ways to find out how effective a drug might be against cancer. Their findings are detailed in a new paper published by *Science Advances*.

"This paper completely changes the way we need to collect tumor tissues and test for drug sensitivity," said Harikrishna Nakshatri, Ph.D., a senior author of the paper. Nakshatri is the Marian J. Morrison professor of breast cancer research at IU School of Medicine and a researcher with the Vera Bradley Foundation Center for Breast Cancer Research at the Indiana University Melvin and Bren Simon Comprehensive Cancer Center. Hal Broxmeyer, Ph.D., a distinguished

professor at IU School of Medicine who passed away in December 2021, also contributed to this study.

Typically, tumors are collected and exposed to room oxygen, which is about 21 percent. However, different organs in the body have different oxygen levels. For example, the brain has 4.4 percent oxygen, blood 5.3 percent, and liver 5.4 percent. When cancer drugs are used on tumors in the clinical setting, they're still in a patient's body and are not exposed to ambient air.

"The oxygen level in our different parts of the body is almost half of what we find in ambient air," Nakshatri said. "Oxygen can have a different effect on the function of different proteins in the tumors. They may get activated, lose their activity level, get degraded or get stabilized. We wanted to test the tumors in a way that more closely resembles how they are in the body, so we know more about what drugs to use."

Researchers tested three different drugs on two different types of tumors. They split the tumors in half and tested one part in 5 percent oxygen, since that is an average oxygen level in the body, and exposed the other part to room oxygen before testing. They looked at the difference in the cancer stem cells, signaling pathways and how drugs behaved in the different oxygen levels. They found the sensitivy level of the tumor cells was different in 5 percent oxygen versus room oxygen.

"This is a study that is now raising more questions we need to answer," Nakshatri said. "Why do the cells react differently? Are we screening the drugs against cancer cells the right way? If we screen for drugs at the physiologic oxygen level, are we going to find different drugs that we may have missed all these years by doing the experiments at 21 percent oxygen?"

In the future, researchers hope to study the different reactions tumors have to other various oxygen levels, like 1 percent or 20 percent. Nakshatri explained this kind of testing could act as another method of screening to determine a drug 's efficacy.

"Suppose we identify a drug with the way we are doing right now in room oxygen, then add another layer of testing in the lab where we keep the cells at the

physiologic oxygen level and compare whether the drug is working or not," Nakshatri said. "If it works, then we can move forward to the clinical setting and it increases the chances of the drug being successful."

More information: Brijesh Kumar et al, Tumor collection/processing under physioxia uncovers highly relevant signaling networks and drug sensitivity, *Science Advances* (2022). DOI: 10.1126/sciadv.abh3375

Provided by Indiana University

Citation: Important new findings about how to test cancer-fighting drugs (2022, January 17) retrieved 8 May 2024 from https://medicalxpress.com/news/2022-01-important-cancer-fighting-drugs.html

This document is subject to copyright. Apart from any fair dealing for the purpose of private study or research, no part may be reproduced without the written permission. The content is provided for information purposes only.