

Corresponding Author Name: Kiran K Velpula Manuscript Number: CDDIS-22-0465RR

# **Reporting Summary**

Springer Nature wishes to improve the reproducibility of the work that we publish. This checklist is used to ensure good reporting standards and to improve the reproducibility. Please respond completely to all questions relevant to your manuscript. For more information, please read the journal's Guide to Authors.

☑ Check here to confirm that the following information is available in the Material & Methods section:

- The exact sample size (n) for each experimental group/condition, given as a number, not a range
- A description of the sample collection allowing the reader to understand whether the samples represent technical or biological replicates (including how many animals, litters, culture, etc.)
- A statement of how many times the experiment shown was replicated in the laboratory
- **Definitions of statistical methods and measures**: For small sample sizes (n<5) descriptive statistics are not appropriate, instead plot individual data points
  - $\circ$  Very common tests, such as *t*-test, simple  $\chi^2$  tests, Wilcoxon and Mann-Whitney tests, can be unambiguously identified by name only, but more complex techniques should be described in the methods section
  - Are tests one-sided or two-sided?
  - o Are there adjustments for multiple comparisons?
  - **♂** Statistical test results, e.g., *P* values
  - Definition of 'center values' as median or mean;
  - o Definition of error bars as s.d. or s.e.m. or c.i.

Please ensure that the answers to the following questions are reported in the manuscript itself. We encourage you to include a specific subsection in the methods section for statistics, reagents and animal models. Below, provide the page number or section and paragraph number.

#### Statistics and general methods

 How was the sample size chosen to ensure adequate power to detect a pre-specified effect size? (Give section/paragraph or page #)

For animal studies, include a statement about sample size estimate even if no statistical methods were used.

- Describe inclusion/exclusion criteria if samples or animals were excluded from the analysis. Were the criteria pre-established? (Give section/paragraph or page #)
- 3. If a method of randomization was used to determine how samples/animals were allocated to experimental groups and processed, describe it. (Give section/paragraph or page #)

For animal studies, include a statement about randomization even if no randomization was used.

Reported in section/paragraph or page # 12				
n/a				
we used n=8 (both male and female) for control and treatments for statistical significance				
n/a				
n/a				
n/a				

 If the investigator was blinded to the group allocation during the experiment and/or when assessing the outcome, state the extent of blinding. (Give section/paragraph or page #)

For animal studies, include a statement about blinding even if no blinding was done.

5. For every figure, are statistical tests justified as appropriate?

Do the data meet the assumptions of the tests (e.g., normal distribution)?

Is there an estimate of variation within each group of data?

Is the variance similar between the groups that are being statistically compared? (Give section/paragraph or page #)

n/a	
n/a	
All experiments are repeated more than 3 times	
n/a	
n/a	
n/a	

### Reagents

Reported in section/paragraph or page # <sup>5</sup>
GAPDH (sc-47724), Gal-1(sc-166617), Anti-GST (A-6) (sc-374171), CA IX (H-11) (sc-365900), LDH-B

6. Report the source of antibodies (vendor and (sc-100/75), Casp-9 (sc-17784), and HIF-1α siRNA (h) (sc-35561) - Santa Cruz Biotechhology Gal-1 (NBP2-75499), CA IX (NB100-417), HIF-1 alpha (NB100-105), Goat anti-Mouse IgG Secondary Ab (NB7539), Goat anti-Mouse IgG Secondary Ab (HB764), NF-kB (#4764S), BAD (#9292) PDH (3205S), IDHA(C4BS) (3582P), and CA9 (D47G3) (5649S) -Cell Signaling Technology.

 Identify the source of cell lines and report if they were recently authenticated (e.g., by STR profiling) and tested for mycoplasma contamination

Cell line GSC33 is obtained from Dr. Kuo's laboratory (University of Wisonsin Madison). Cell lines GSC20, GSC28, GSC 262, GSC 268, GSC 6-27 were obtained from Dr. Bhat's laboratory (MD Anderson). Human astrocytes (#1800) were obtained from ScienCell Research Laboratories (Carlsbad, CA) Neural progenitor cells (NPC) were purchased from Lonza (PT-2599) (Basel, Switzerland). Mycoplasma testing is done for every three months.

#### **Animal Models**

- 8. Report species, strain, sex and age of animals
- For experiments involving live vertebrates, include a statement of compliance with ethical regulations and identify the committee(s) approving the experiments.

#### Reported in section/paragraph or page #12

4-week-old athymic nude mice (nude-Foxn1nu, 4 male and 4 female mice from Envigo)					
n/a					

10. We recommend consulting the ARRIVE guidelines (<u>PLoS Biol. 8(6)</u>, e1000412,2010) to ensure that other relevant aspects of animal studies are adequately reported.

#### **Human subjects**

- 11. Identify the committee(s) approving the study protocol.
- 12. Include a statement confirming that informed consent was obtained from all subjects.
- 13. For publication of patient photos, include a statement confirming that consent to publish was obtained.
- 14. Report the clinical trial registration number (at <u>ClinicalTrials.gov</u> or equivalent).

#### Reported in section/paragraph or page # 5

IRB #:85193	
The tissue utilized in this study is "excess tissue" that would have been remove subjects are recruited for this research and no patient contact took place and s required	
n/a	
n/a	

- 15. For phase II and III randomized controlled trials, please refer to the <u>CONSORT statement</u> and submit the CONSORT checklist with your submission.
- 16. For tumor marker prognostic studies, we recommend that you follow the REMARK reporting guidelines.

#### **Data deposition**

- 17. Provide accession codes for deposited data.

  Data deposition in a public repository is mandatory for:
  - a. Protein, DNA and RNA sequences
  - b. Macromolecular structures
  - c. Crystallographic data for small molecules
  - d. Microarray data

## Reported in section/paragraph or page #

n/a			

Deposition is strongly recommended for many other datasets for which structured public repositories exist; more details on our data policy are available in the Guide to Authors. We encourage the provision of other source data in supplementary information or in unstructured repositories such as <u>Figshare</u> and <u>Dryad</u>. We encourage publication of Data Descriptors (see <u>Scientific Data</u>) to maximize data reuse.

18. If computer code was used to generate results that are central to the paper's conclusions, include a statement in the Methods section under "Code availability" to indicate whether and how the code can be accessed. Include version information as necessary and any restrictions on availability.

n/a	à			