

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 [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
<i>Only common tests should be described solely by name; describe more complex techniques in the Methods section.</i> |
| ☒ | ☐ | 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
<i>Give P values as exact values whenever suitable.</i> |
| ☒ | ☐ | 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 R version 4.3.1, TCGAblinks package version 2.29.6

Data analysis R version 4.3.1 was used for all analysis.
Custom code is available at :
Zenodo: <https://doi.org/10.5281/zenodo.14602489>
GitHub: <https://github.com/SydneyBioX/HEtoSGEBench>

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 [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 description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our [policy](#)

All datasets used in this study are publicly available and were downloaded from the following links: (1) human HER2-positive breast cancer ST data reported in

Anderson et al. (<https://github.com/almaan/her2st>); (2) Human cutaneous squamous cell carcinoma reported in Ji et al. (<https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE144240>); (3) TCGA-BRCA data was requested using the TCGAbiolinks package, clinical information from Broad GDAC Firehose (<http://gdac.broadinstitute.org/>); (4) 10x Visium Human Breast Cancer (Visium-HER2+) Block A Section 1 and Section 2 were downloaded from (<https://www.10xgenomics.com/datasets/human-breast-cancer-block-a-section-1-1-standard-1-1-0>) and (<https://www.10xgenomics.com/datasets/human-breast-cancer-block-a-section-2-1-standard-1-1-0>); (5) 10x Visium Whole Transcriptome Human Breast Cancer (Visium-Hercep-Test2+) was downloaded from (<https://www.10xgenomics.com/datasets/human-breast-cancer-visium-fresh-frozen-whole-transcriptome-1-standard>); (6) The 10x Visium Human Healthy Kidney (Visium-Kidney) dataset was downloaded from (<https://www.10xgenomics.com/datasets/human-kidney-11-mm-capture-area-ffpe-2-standard>). Source data are provided with this paper.

Research involving human participants, their data, or biological material

Policy information about studies with [human participants or human data](#). See also policy information about [sex, gender \(identity/presentation\), and sexual orientation](#) and [race, ethnicity and racism](#).

Reporting on sex and gender	N/A
Reporting on race, ethnicity, or other socially relevant groupings	N/A
Population characteristics	N/A
Recruitment	N/A
Ethics oversight	N/A

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

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](https://www.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	Our study consists 36 samples of HER2+ breast tissue sections from 8 patients, 12 samples of CSCC from 4 patients, and 671 images from TCGA-BRCA, two adjacent slides from a HER2+ breast cancer patient (Visium-HER2+), one slide from a Hercep-Test2+ breast cancer patient (Visium-Hercep-Test2+) and one kidney healthy tissue from a patient (Visium-Kidney).
Data exclusions	We only considered TCGA-BRCA images that had one associated RNA-Seq entry and were either stage I, stage III, or stage IIA due to storage limitations.
Replication	The benchmarking study was conducted on 6 datasets to ensure robustness of results.
Randomization	This is not applicable to our study because we do not have experimental groups.
Blinding	This is not applicable to our study because we do not have experimental groups.

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	Involvement in the study
☒	☐ Antibodies
☒	☐ Eukaryotic cell lines
☒	☐ Palaeontology and archaeology
☒	☐ Animals and other organisms
☒	☐ Clinical data
☒	☐ Dual use research of concern
☒	☐ Plants

Methods

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

Plants

Seed stocks	N/A
Novel plant genotypes	N/A
Authentication	N/A