MAX PLANCK INSTITUTE
FOR MOLECULAR GENETICS

MethyLYZR: Live brain tumor diagnosis from sparse epigenomic data

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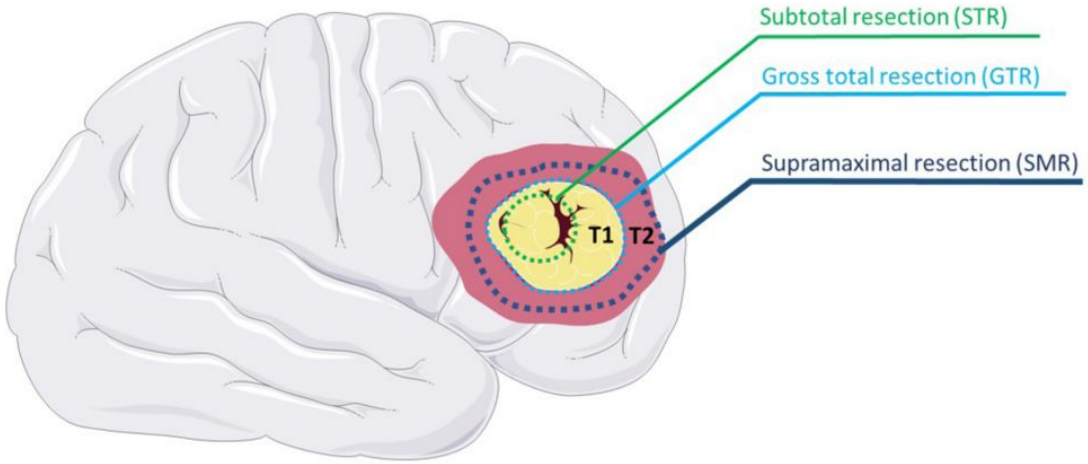
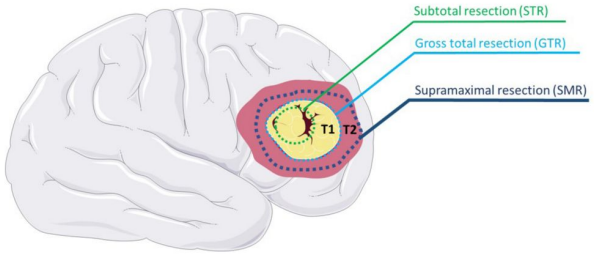
Brain tumor resection



Willman et al., 2023
Badza et al., 2020

Standard practice for CNS tumors



1 Preparation and begin 60 mins

2 Incision 30 mins

3 Resection 60 min

4 End 30 min

5 Standard molecular diagnostics 4-7 days

Classification

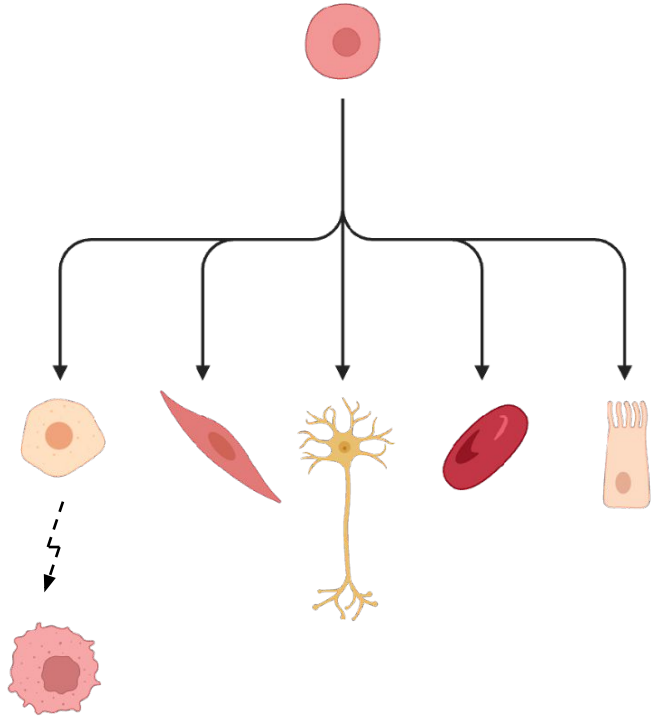
- sequencing
- Immunohistochemistry
- **methylation microarrays**

aim: intra-operative DNA methylation-based classification

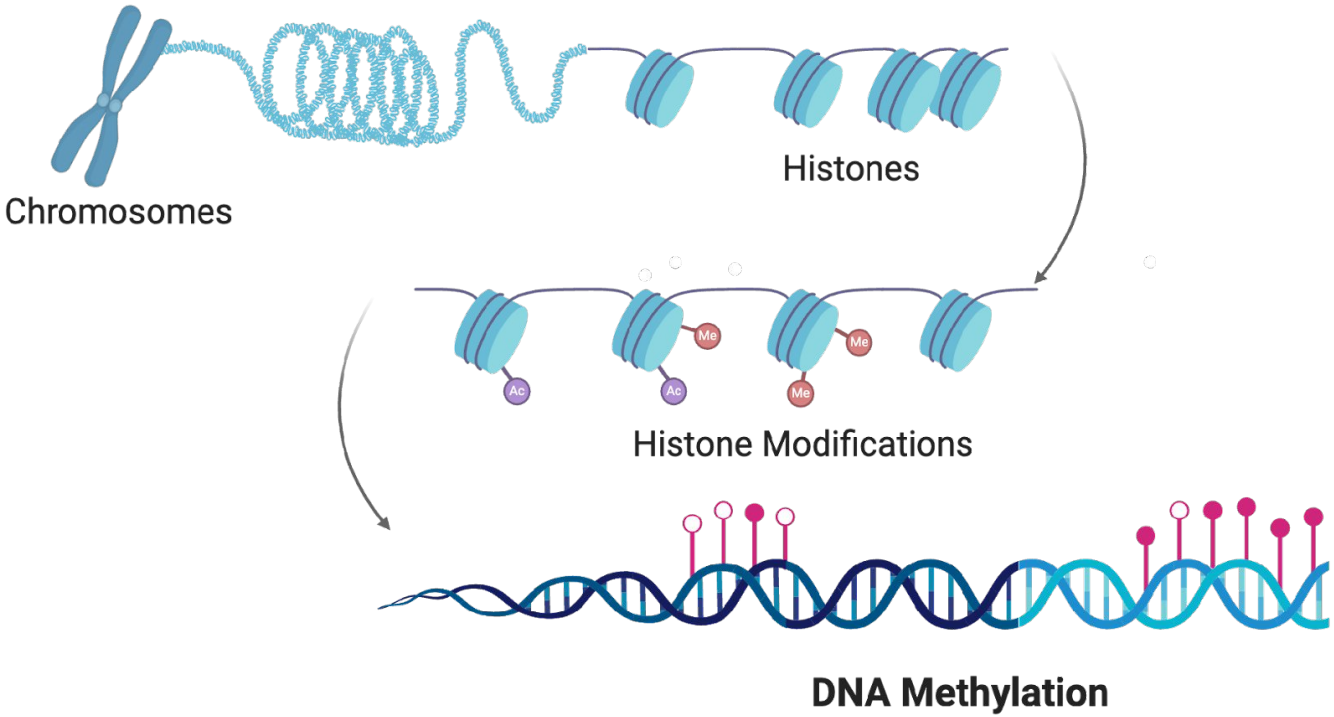
Senders et al., 2018
Sturm et al., 2023

DNA Methylation as biomarker

One genotype
– multiple phenotypes –

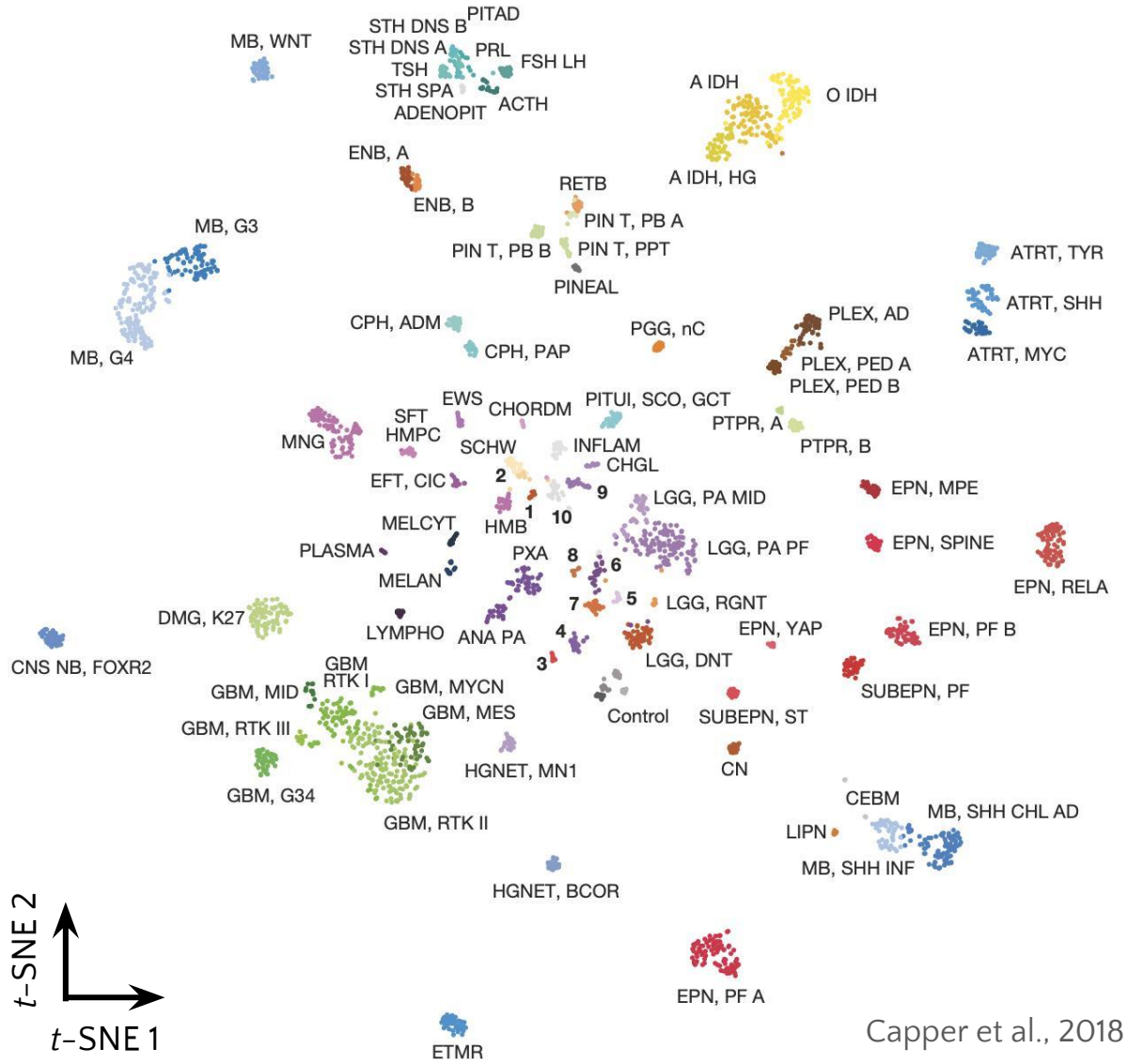


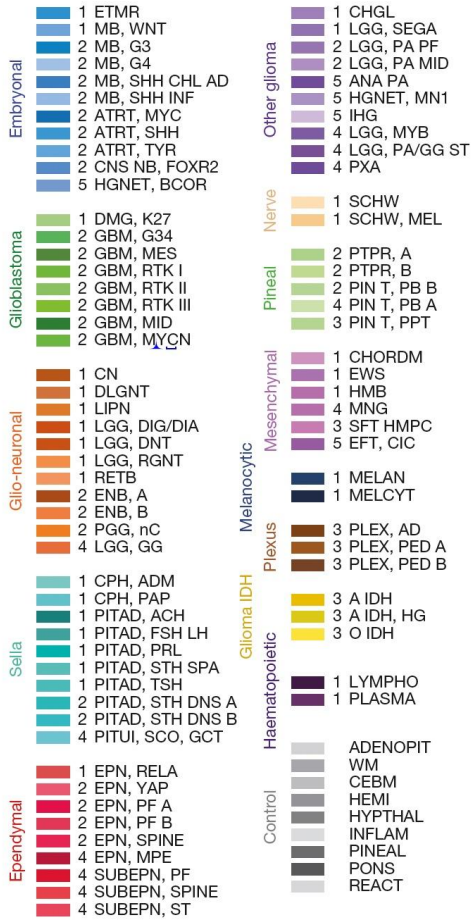
Epigenome
– 2nd layer of information –



Comprehensive training dataset

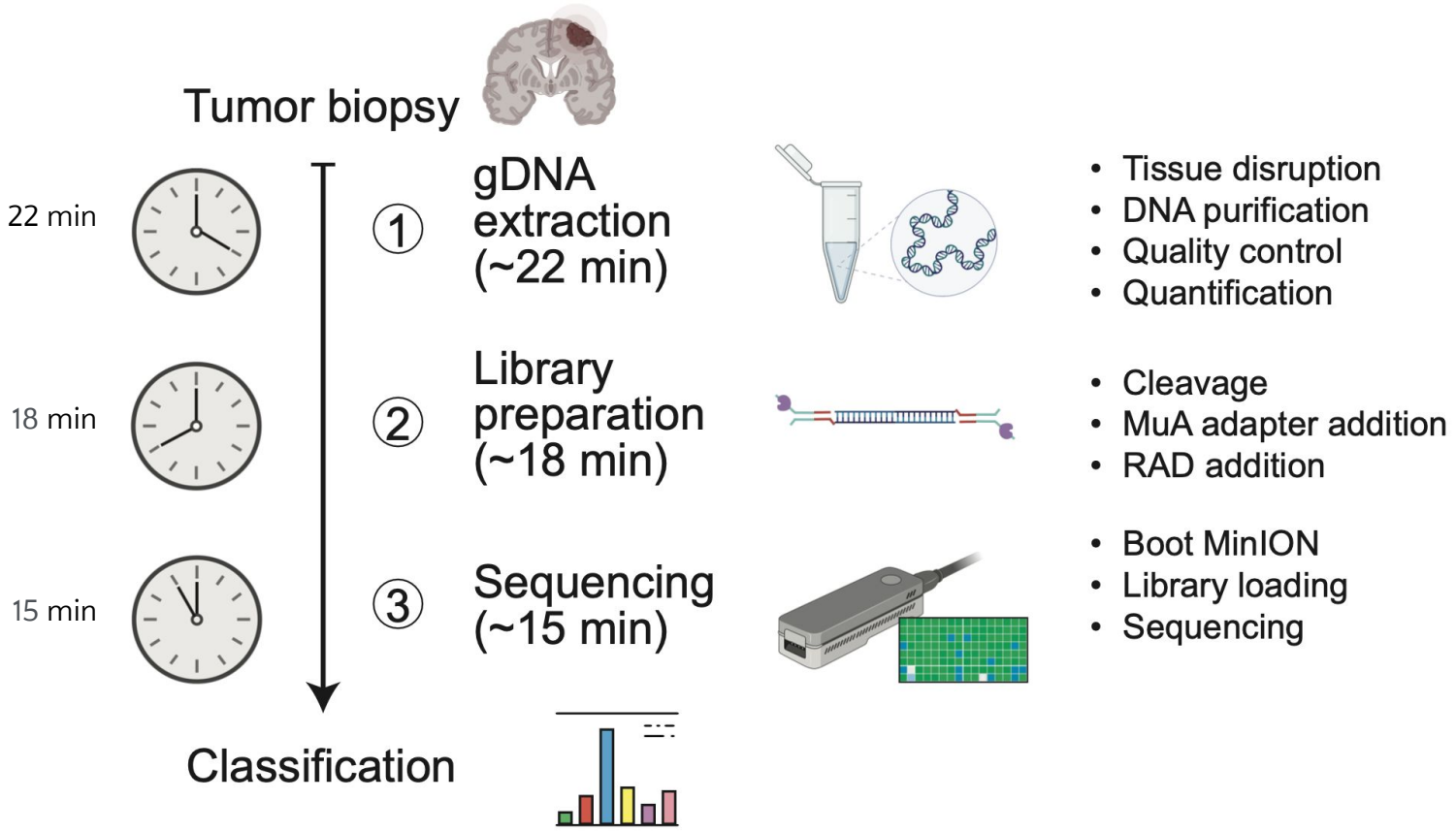
More than 2,800 samples covering 91 tumor classes





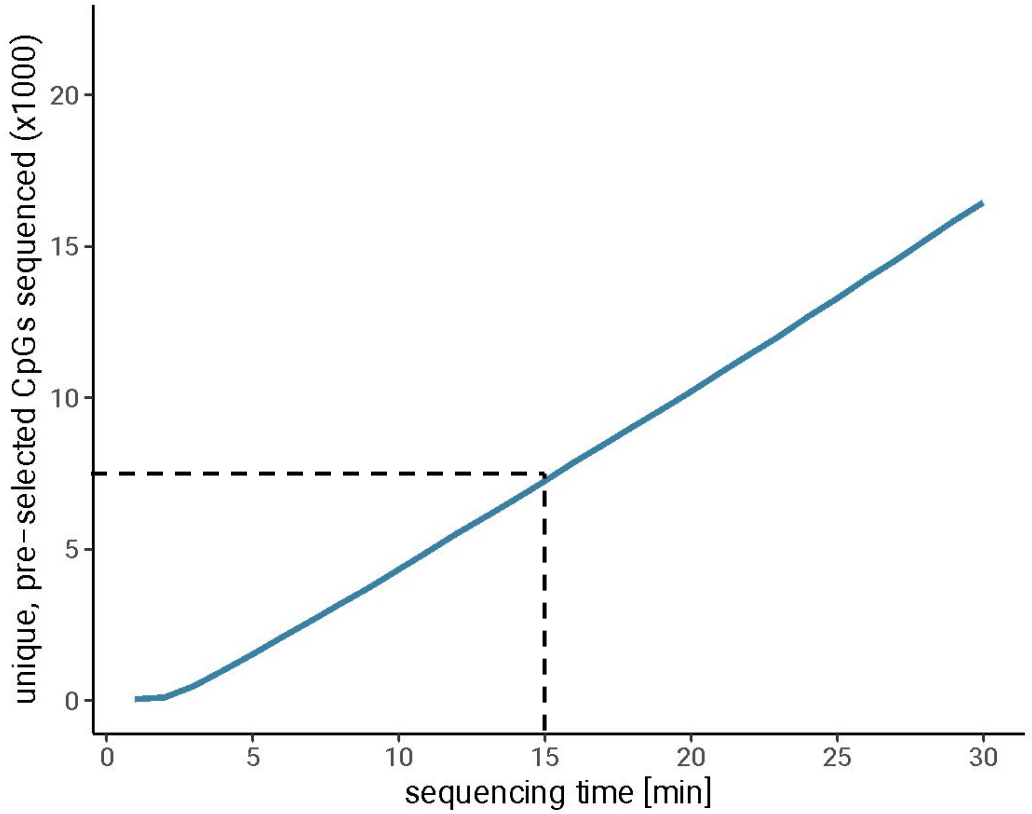
Capper et al., 2018

Intra-operative classification approach



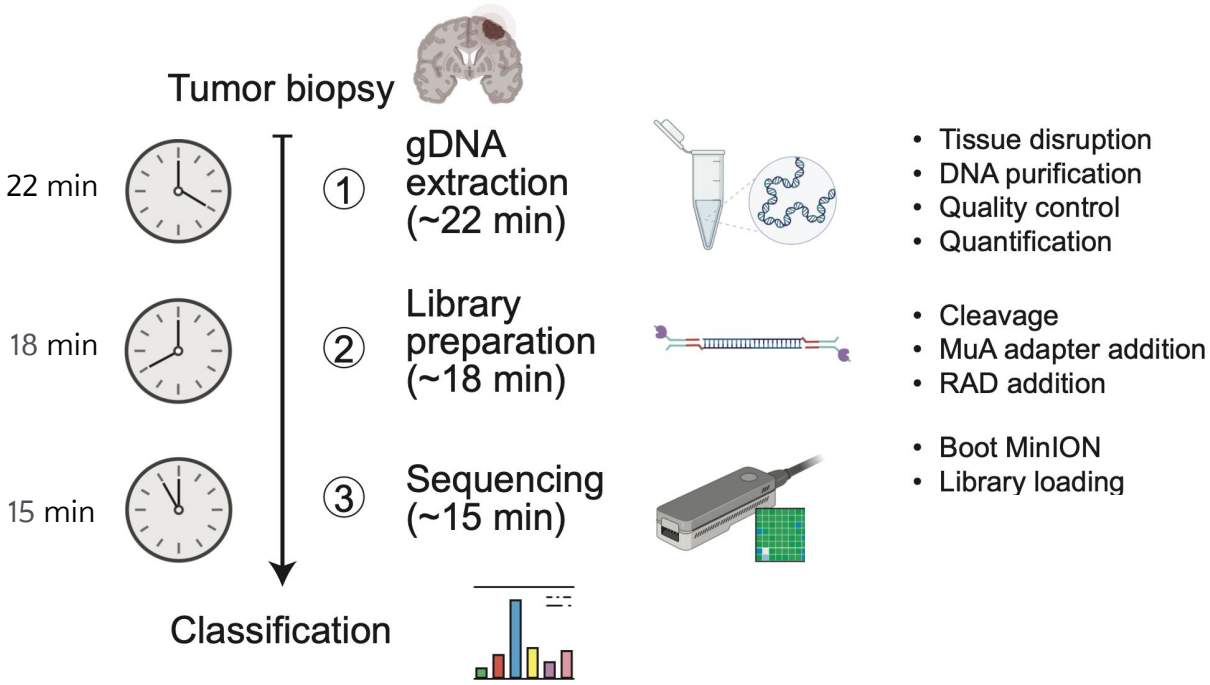
Intra-operative classification approach

Throughput (CpGs covered by Illumina 450k BeadChip)



~7,500 CpGs in 15 minutes

98% missing values



Naïve Bayes

Bayes' Theorem

likelihood

class prior

$$P(C_j|X) = \frac{P(X|C_j)P(C_j)}{P(X)}$$

posterior

predictor prior

class C_j

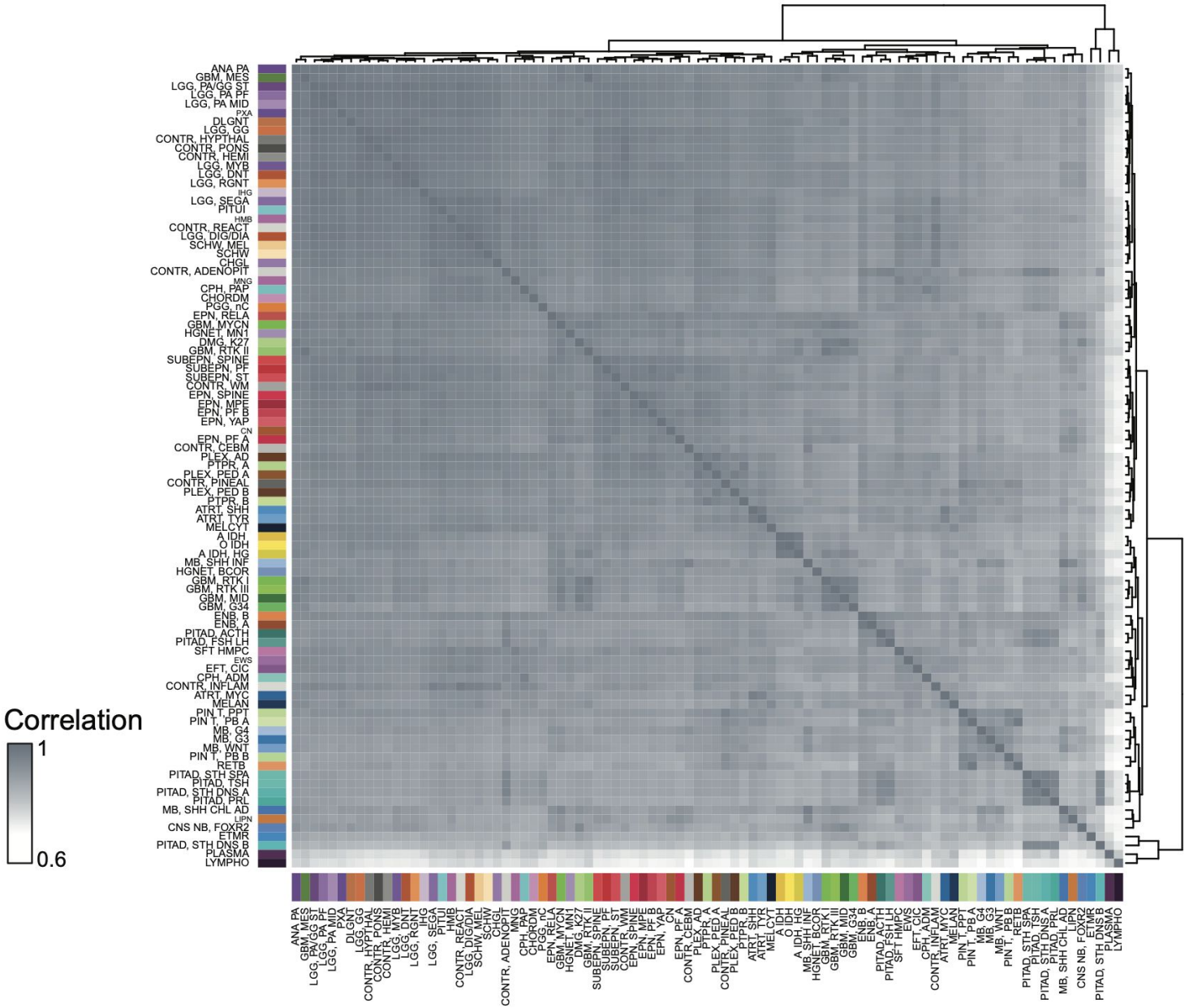
observation X

Independence assumption

$$P(X|C_j) = P(x_1, \dots, x_p|C_j) = \prod_{i=1}^{p} P(x_i|C_j)$$

$$X = \{x_1, x_2, \dots, x_p\}$$

Methylation profiles are highly correlated



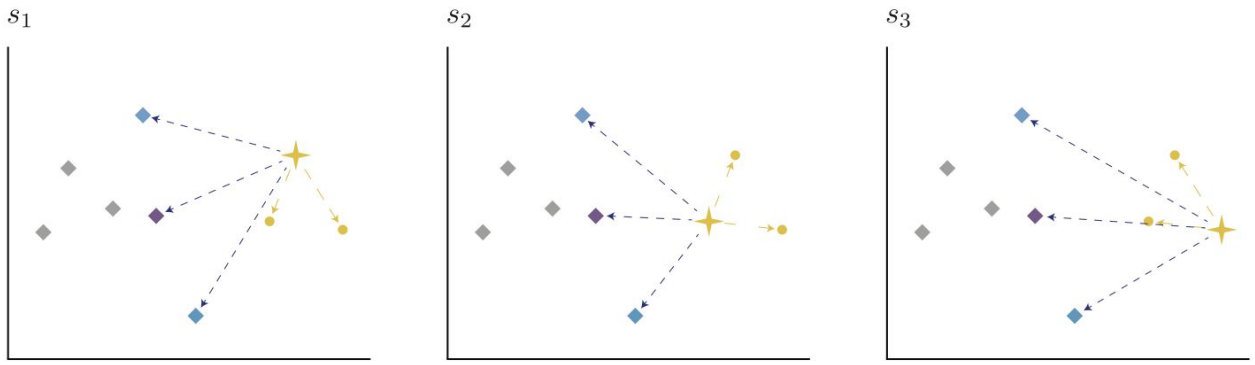
Feature weighting to discern informative and non-informative features

ReliefF-based feature weights

Case I: ▲ inter-class distance ▼ intra-class distance ⇒ $\omega_{i,j} > 0$

$$\omega_{i,j} = \sum_{s \in C_j} \{\text{mean distance to misses} - \text{mean distance to hits}\}$$

for each instance in class C_j

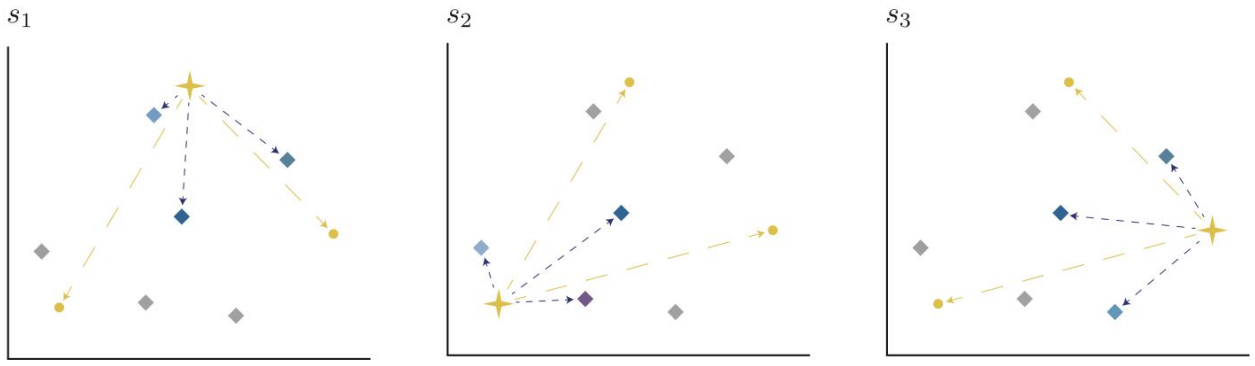


$k = 3$ misses

◇ class centroid
◆ miss
◆ not considered
✦ target instance
● non-target instance
→ distance
⇢ ... to miss
⇢ ... to hit

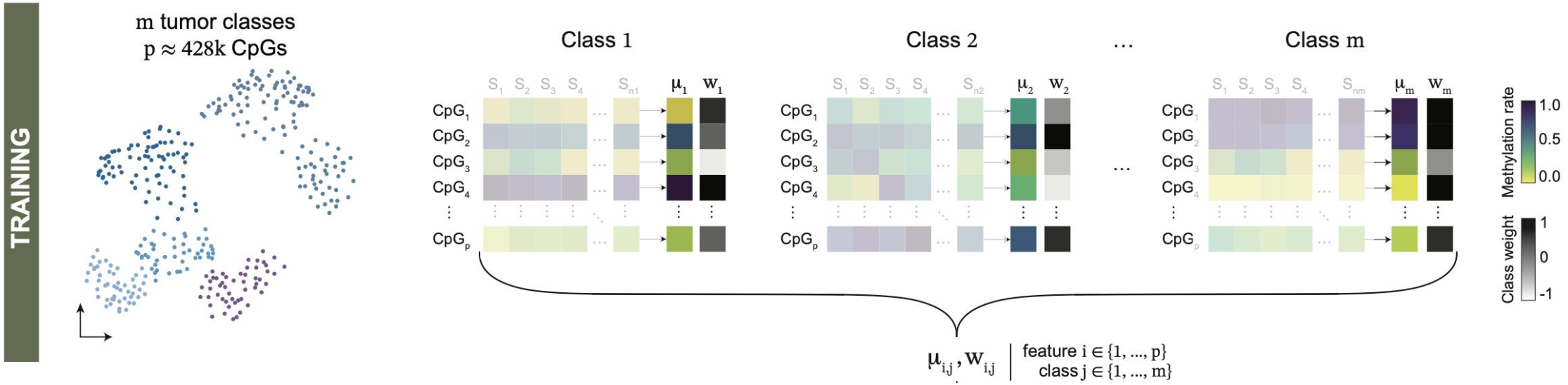
Case II: ▼ inter-class distance ▲ intra-class distance ⇒ $\omega_{i,j} < 0$

for each instance in class C_j



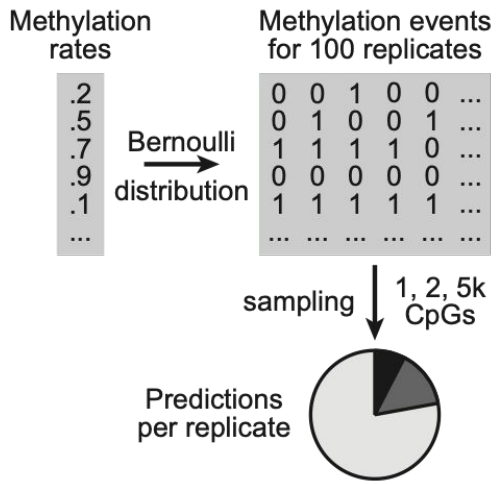
Kira and Rendell, 1992
Kononenko, 2005

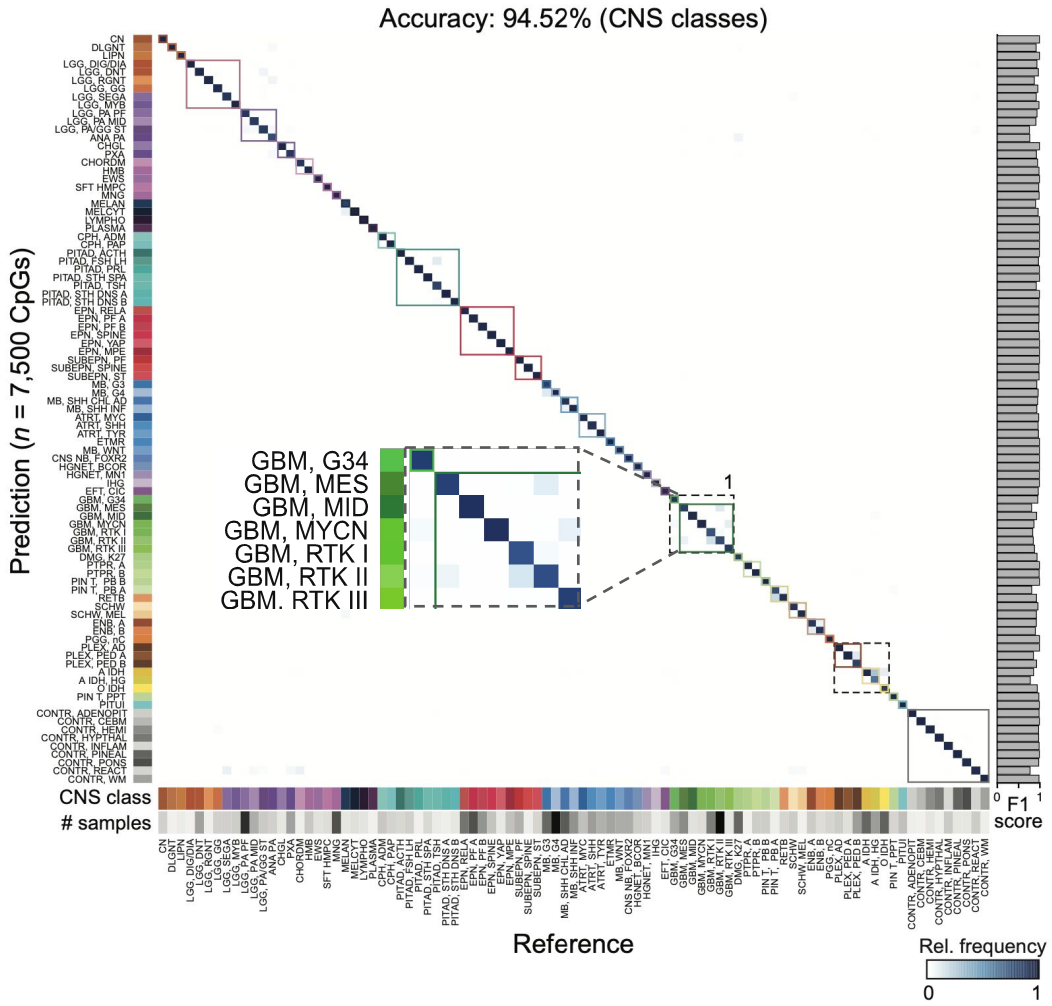
MethyLYZR Framework



Evaluation – Synthetic Data

Using 450k data to simulate low-coverage Nanopore sequencing

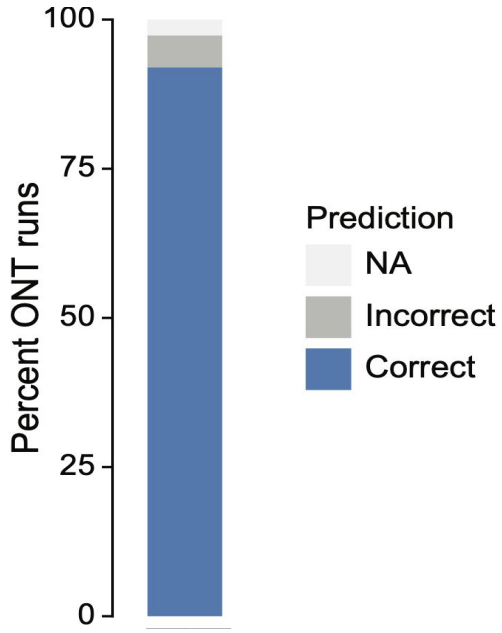


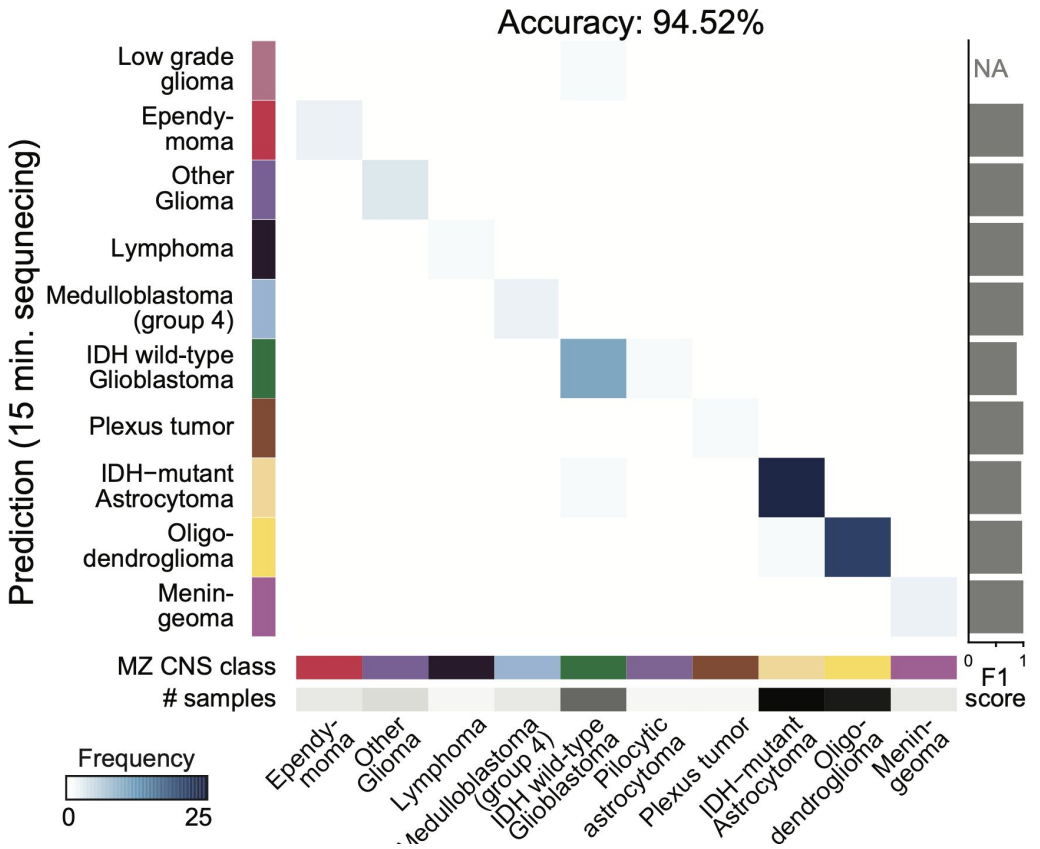


Broad level accuracy: 97.72%

Evaluation – Nanopore data

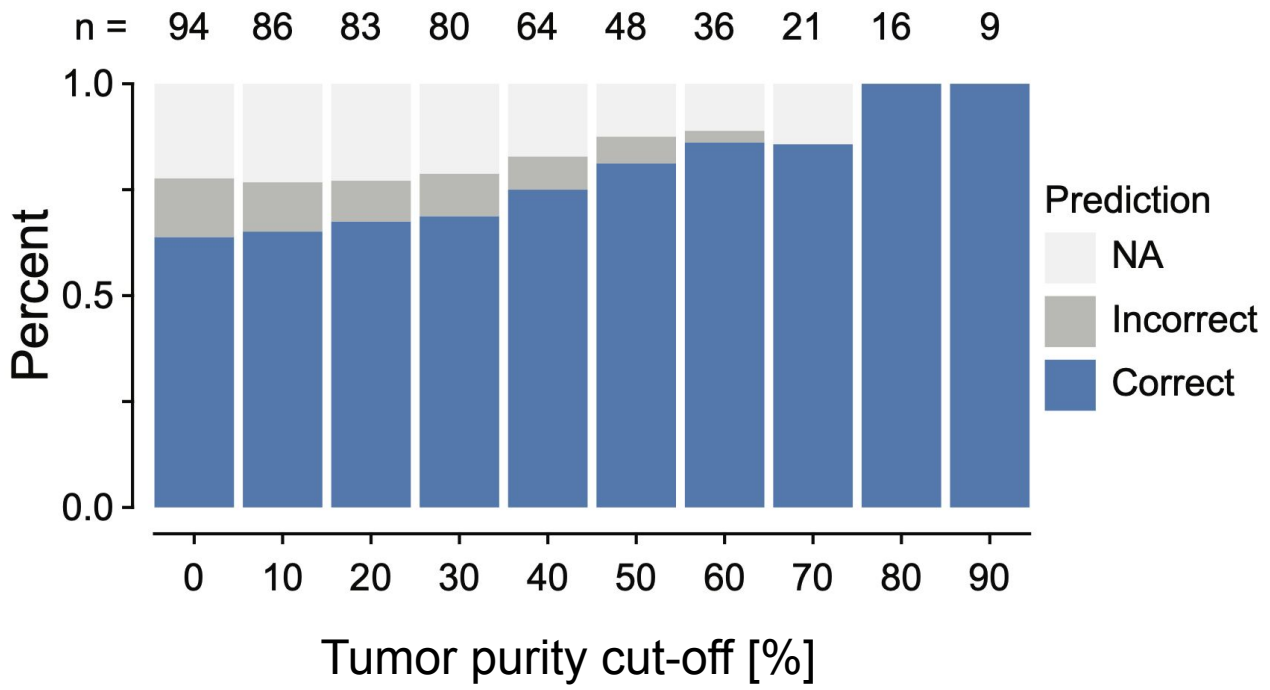
$n = 75$ Nanopore-sequenced samples





Feasibility – Tumor Purity

$n = 94$ external Nanopore-sequenced samples



Acknowledgments

Helene Kretzmer

Franz-Josef Müller

Alena van Bömmel

Björn Brändl

Carolin Kubelt

Michael Synowitz

Christian Rohrandt

Bernhard Schuldt

Gaojianyong Wang

Romulas Smičius

Maximilian Evers

Stephen Yip

Ole Ammerpohl

UKSH UNIVERSITÄTSKLINIKUM Schleswig-Holstein

FACHHOCHSCHULE KIEL University of Applied Sciences

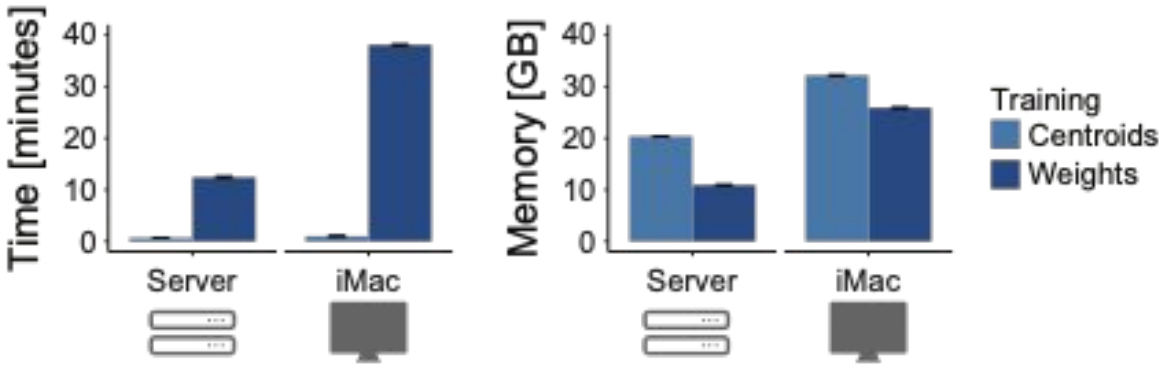
MAX PLANCK INSTITUTE FOR MOLECULAR GENETICS

altona DIAGNOSTICS

Federal Ministry of Education and Research

Funding: IntraEpiGliom
FKZ 13GW0347

Training Naïve Bayes model



Complexity

- p: number of features, i.e., CpGs
- m: number of classes
- n_j: number of samples in class C

  $\rightarrow$ such that $N = \sum_j n_j$ is the number of all samples

centroids

$$\mathcal{O}(p \cdot N)$$

weights

$$\mathcal{O}(m \cdot N \cdot p) + \mathcal{O}(p \cdot (n_1^2 + n_2^2 + \ldots + n_m^2))$$

Model Scalability

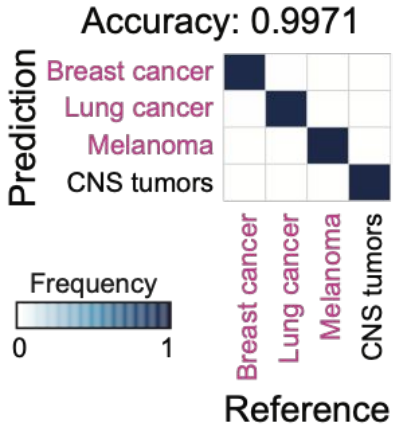
Metastases

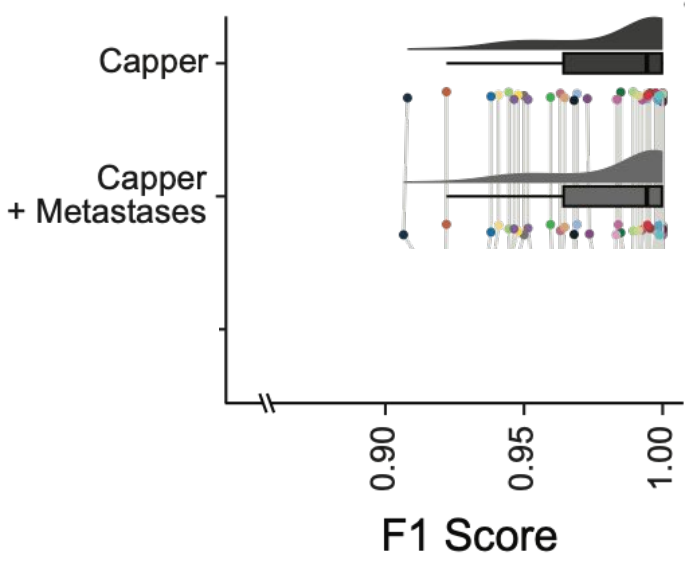
- Breast cancer (n = 30)
- Lung cancer (n = 18)
- Melanoma (n = 37)

Sarcomas

- m = 65 classes
- n = 1077 samples

Synthetic data





core MethyLYZR classes

Salomon et al., Scientific data (2018)
Koelsche et al., Nat. Commun. (2021)

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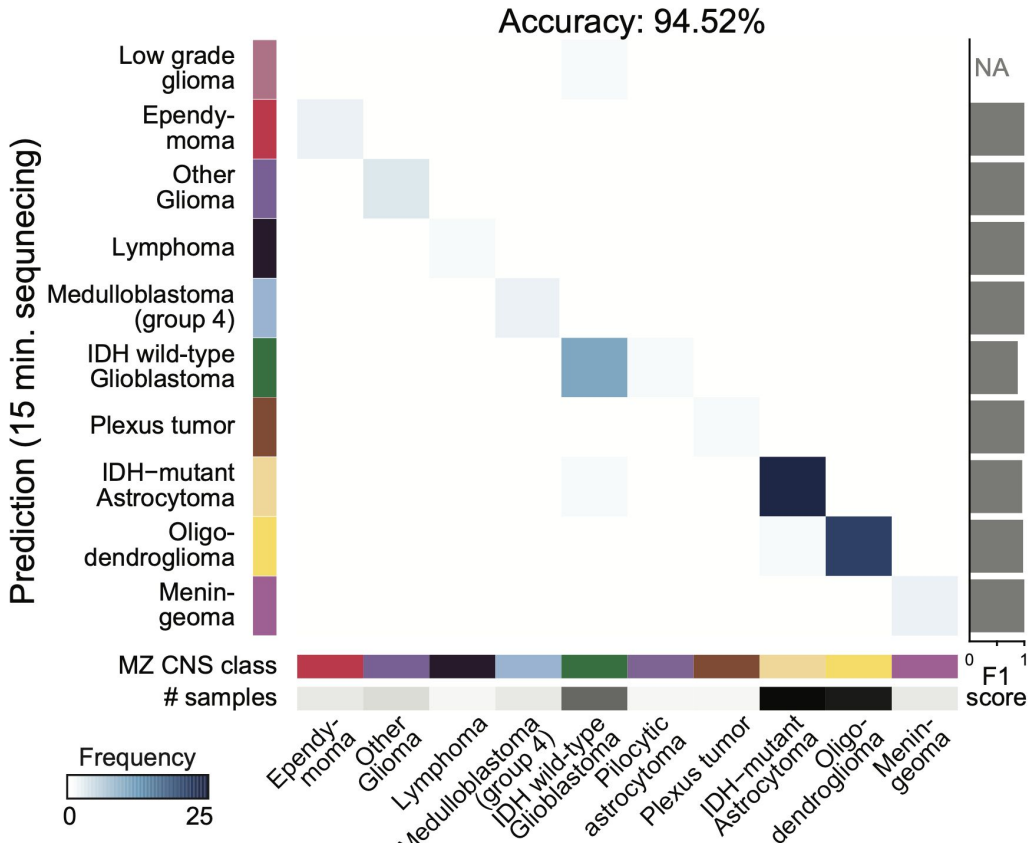
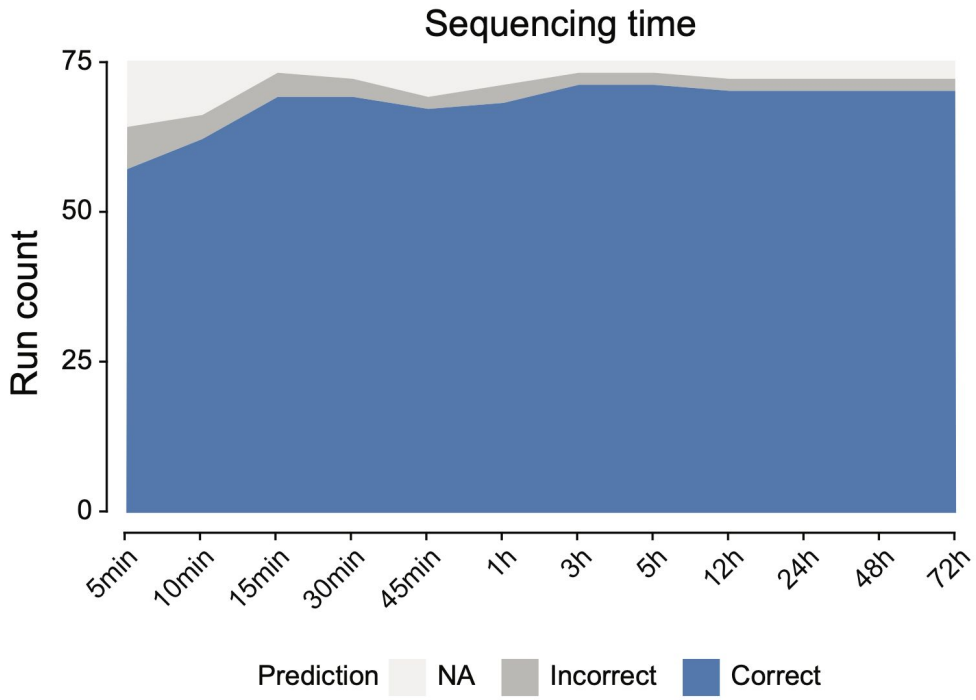
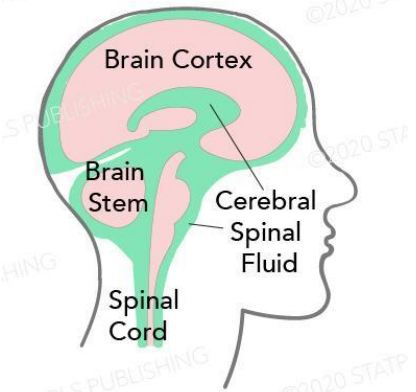
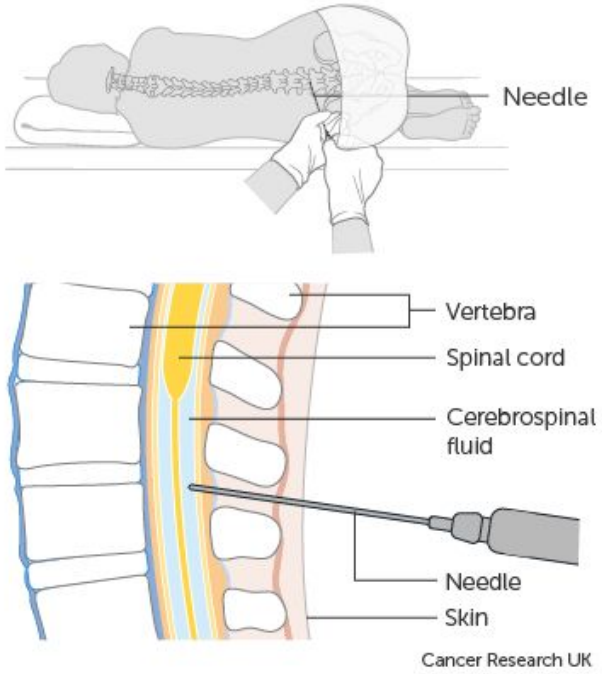




Feasibility – Liquid biopsy

Cerebrospinal Fluid (CSF) from Lumbar Puncture





Afflerbach et al., 2023

Feasibility – Liquid biopsy

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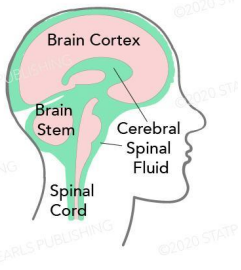
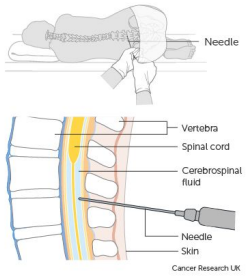
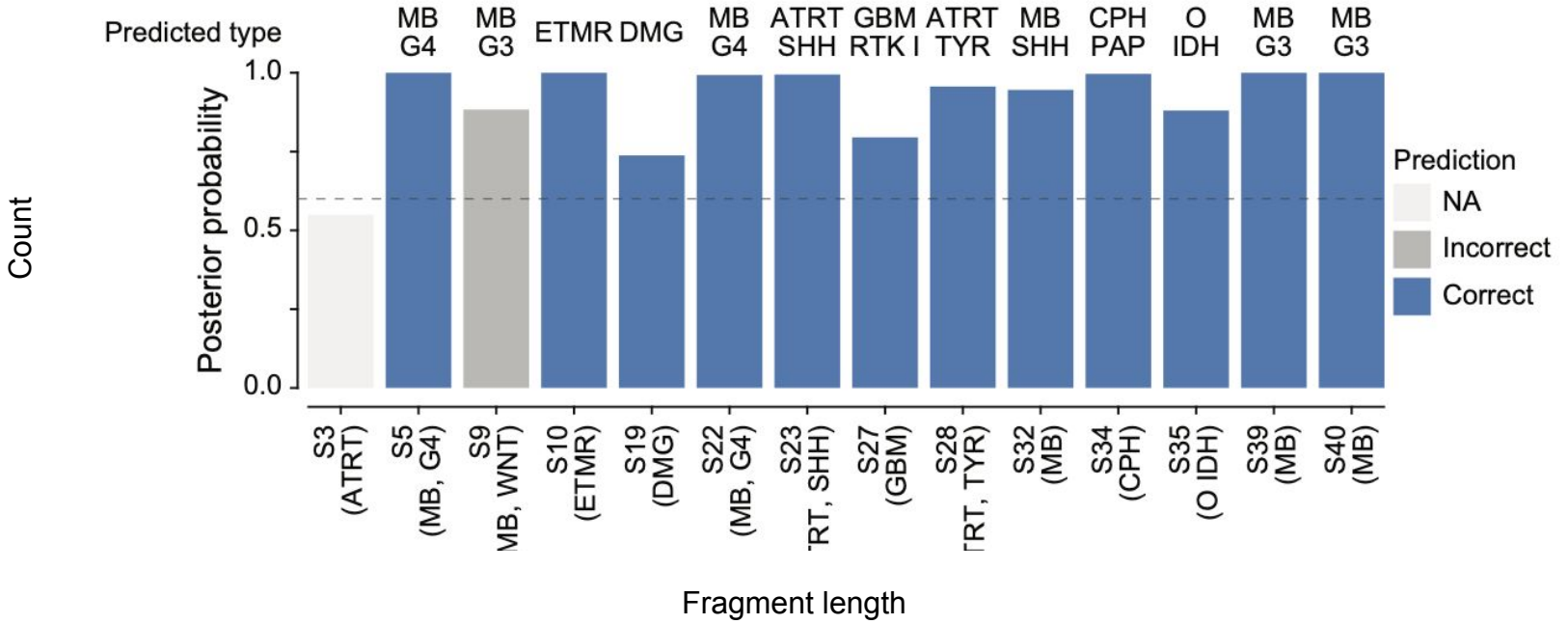






Afflerbach et al., 2023

Evaluation – Nanopore cohort

Comparing Performance to Sturgeon

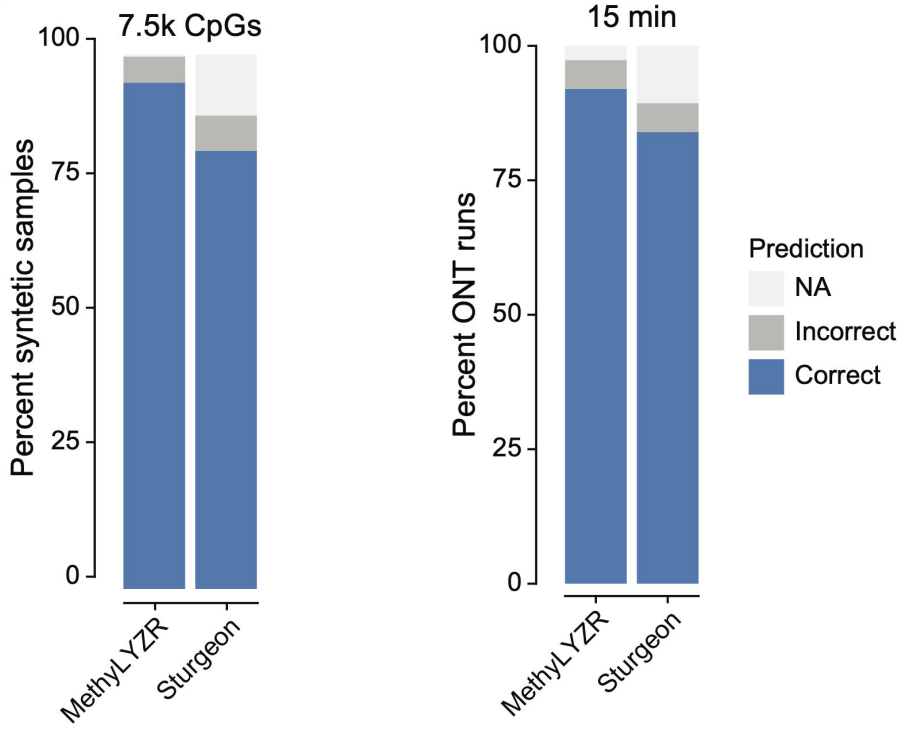
Article

Ultra-fast deep-learned CNS tumour classification during surgery

https://doi.org/10.1038/s41586-023-06615-2
Received: 10 February 2023
Accepted: 6 September 2023

C. Vermeulen[1,2,6], M. Pagès-Gallego[1,2,6], L. Kester[3], M. E. G. Kranendonk[3], P. Wesseling[3,4], N. Verburg[5], P. de Witt Hamer[5], E. J. Kooi[4], L. Dankmeijer[4,5], J. van der Lugt[3], K. van Baarsen[3], E. W. Hoving[3], B. B. J. Tops[3]✉ & J. de Ridder[1,2]✉

- Data augmentation to train neural network
  - >35 million simulated runs from 2800 arrays
- 18/25 classified samples in under 90 minutes
  - 20–40 minutes of sequencing



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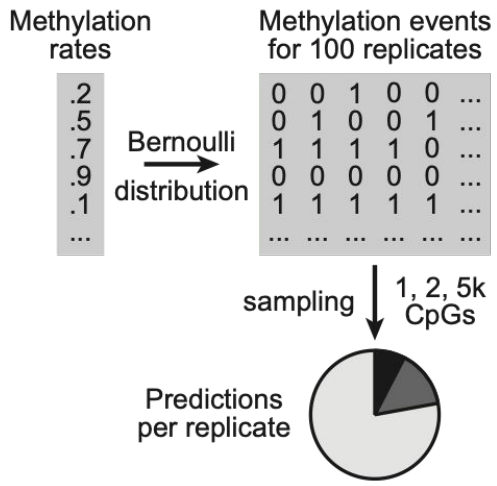
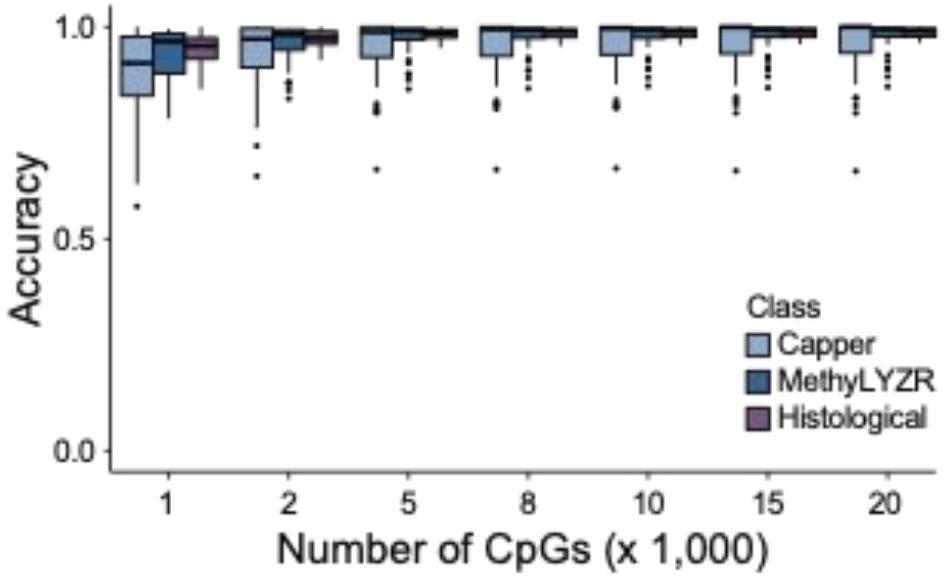




Histological classes (m = 8) defined by Capper et al. (2018)

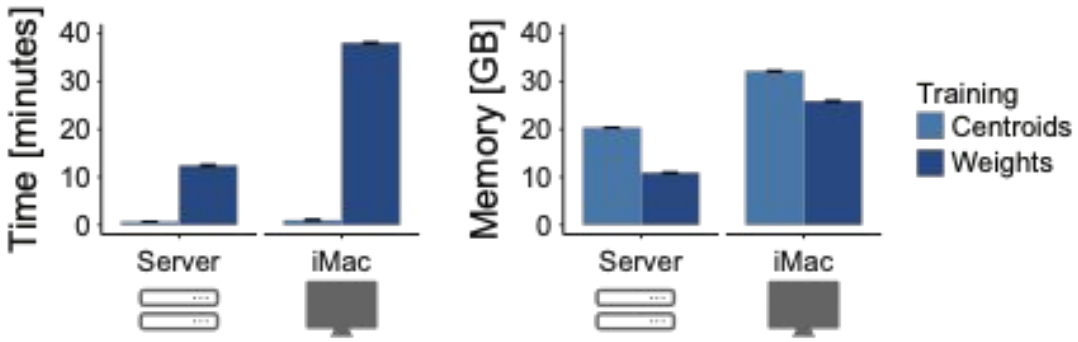
"histologically and biologically closely related tumour classes, the distinction of which is currently without clinical impact"

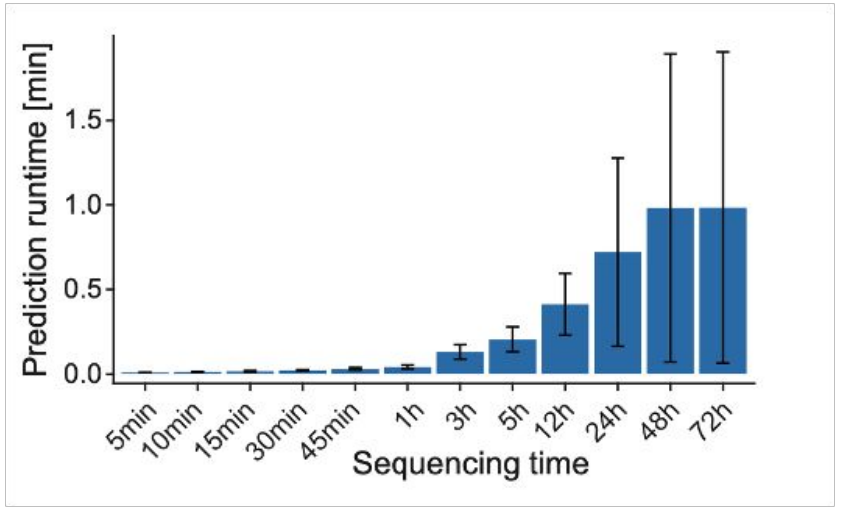
Benchmarking

Training

Prediction

MethyLYZR





Alternative Approaches

- ad hoc RF
  - no pre-training possible
- neural network
  - days – weeks
  - hundreds of GB to TB memory

- ad hoc RF
  - 17-60 min
- neural network
  - few seconds

Djirackor et al., 2021
Vermeulen et al., 2023

RELIEF-based feature weighting

$w_{i,j}$ feature weight for class C_j and feature i

$$w_{i,j} = \sum_{x \in C_j} \left\{ \frac{\sum_{\substack{m \in KNN(x) \\ l(m) \neq C_j}} |x_i - m_i|}{k} - \frac{\sum_{\substack{h \neq x \\ h \in C_j}} |x_i - h_i|}{|C_j| - 1} \right\}$$

, where $KNN(x)$: k-nearest centroids of x,
$l: X \rightarrow C$: maps centroid to class

distance to misses $\boldsymbol{m}$ distance to hits $\boldsymbol{h}$

- $w_{i,j} > 0$: informative feature
- $w_{i,j} < 0$: uninformative feature

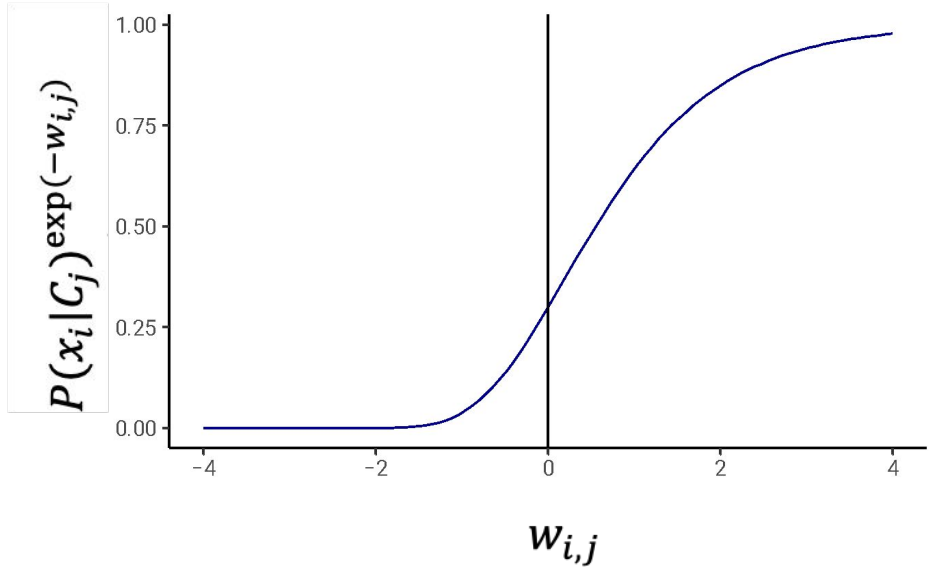
Kira and Rendell, 1992
Kononenko, 2005
Urbanowicz et al., 2018

Feature weighting in Naïve Bayes

proposed by Foo et al., 2021:

$$P(x_i|C_j)^{\exp(-w_{i,j})}$$

Example: $\boldsymbol{P(x_i|C_j) = 0.3}$



Zaidi et al., 2013
Xiang et al., 2015
Jiang et al., 2018