Small Molecule inhibitors of BRAF-V600E

BRAF

- BRAF is located at 7q34
- BRAF encodes for a protein related to cell division in the human body
- The normal function of BRAF includes sending signals inside the cell to promote cell division
- BRAF regulates the MAP kinase and EPK signaling pathway and affects cell division and differentiation..

Mutations in BRAF

- Mutations in BRAF result in the absence of the chemical signals that regulate cell division, causing uncontrolled cell growth.
- Mutated forms of the gene and protein are found in many types of cancers
  - Melanoma, non-Hodgkin lymphoma, colorectal cancer, thyroid carcinoma
- V-600E is the most common BRAF mutation and a class one mutation that causes a range of diseases

BRAF V600E

- The V600E mutation is caused by single nucleotide variant and indel (insertion/deletions) mutations
- Mutation occurs when Valine (V) replaces a Glutamic Acid (E) at amino acid 600

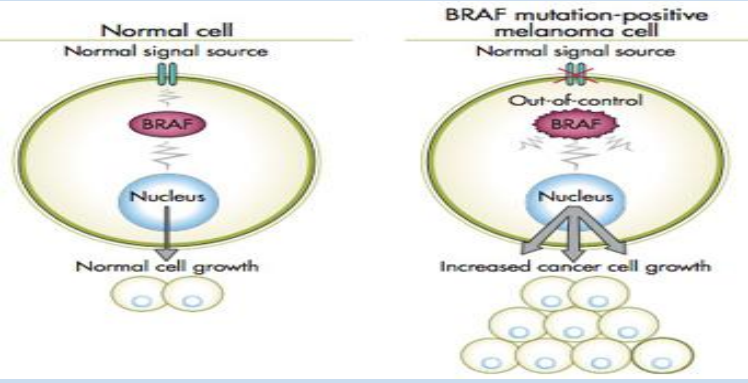


Fig 1: BRAF cell function and cell mutation

K601E	2	High	1 (1.8)
G469A	2	High	1 (1.8)
G469R	2	Intermediate	1 (1.8)
G466E	3	Low	1 (1.8)
G466A	3	Low	1 (1.8)
D594N	3	None	1 (1.8)
D594G	3	None	2 (3.4)
T599I	/	Unknown	1 (1.8)

Fig 2: BRAF mutation classes

BRAF Mutation Classes

Class 1: related to codon 600, kinase activated, RAS-independent (RAS WT), BRAF monomers. Constitutively active extracellular signal related kinases (ERK) by phosphorylation, allows BRAF to act as a monomer.
Class 2: non-V600 mutant, strong kinases activation is regulated by dimers of mutant BRAF, independent on RAS (RAS WT)
Class 3: Kinase impaired or inactive and consists of BRAF non-600 mutant and CRAF wild type as a heterodimer. Is dependent on RAS, causing signals to be transferred downstream.

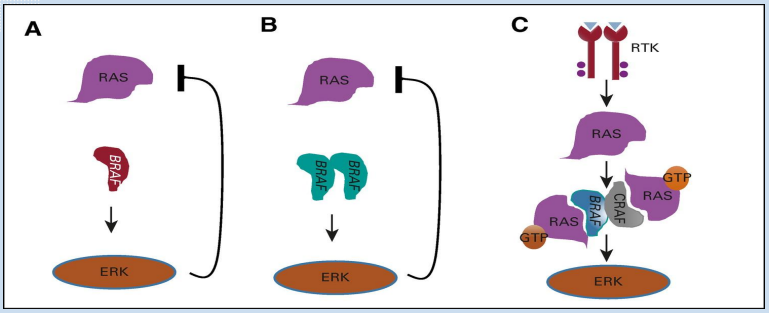
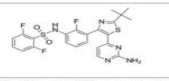
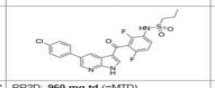
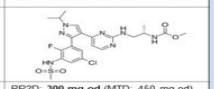
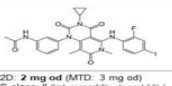
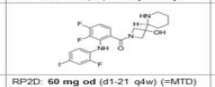
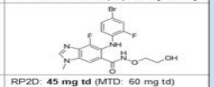


Fig 3: Oncogenic signaling of different classes of BRAF mutation, Class 1(A), Class 2(B), Class 3(C)

BRAF Inhibitors

- BRAF inhibitors such as Vemurafenib and Dabrafenib target the mutated BRAF protein, resistance often develops.
- BRAF inhibitors are drugs designed to target and block the activity of the mutated BRAF protein, especially in cancers with the V600E mutation.

	Dabrafenib / Trametinib	Vemurafenib / Cobimetinib	Encorafenib / Binimetinib
BRAF Inhibitors **Dab.** (GSK2118436) / **Vem.** (PLX4032,RG7204) / **Enc.** (LGX818)			
	RP2D: **150 mg td** (MTD not reached) * BCS class: II (high permeability, low solubility) Food effect: Intake 1h prior or 2 h after meal Absorption (t_{max}): **1.9 h** Time to steady-state ($t_{max,ss}$): **14 d** $AUC_{0-24,ss}$: **4.3 h*µg/mL** (38 %CV_S) $C_{max,ss}$: **1478 ng/mL** (37 %CV_S) Clearance (CL/F): **17.3 L/h** (nc) Elimination half-life (t½): **8.4 h** (nc)	RP2D: **960 mg td** (=MTD) BCS class: IV (low permeability, low solubility) Food effect: none (intake with/without food) Absorption (t_{max}): **~ 4 h** Time to steady-state ($t_{max,ss}$): **15.22 d** $AUC_{0-8,ss}$: **380.2 h*µg/mL** (38 %CV_S) $C_{max,ss}$: **56,700 ng/mL** (38 %CV_S) Clearance (CL/F): **1.2 L/h** (32 CV_{ss}%) Elimin. half-life (t½): **56 h** [30-120]	RP2D: **300 mg od** (MTD: 450 mg od) BCS class: *nr* Food effect: None (intake with/without food) Absorption (t_{max}): **2.0 h** Time to steady-state ($t_{max,ss}$): **15 d** $AUC_{0-24,ss}$: **12.3 h*µg/mL** (med.) $C_{max,ss}$: **3100 ng/mL** (med.) Clearance (CL/F): **24.4 L/h** (med.) Elimination half-life (t½):**6.3 h** [3.7-8.1]
MEK Inhibitors **Tra.** (GSK1210212) / **Cob.** (RG7420) / **Bin.** (MEK162)			
	RP2D: **2 mg od** (MTD: 3 mg od) BCS class: II (high permeability, low solubility) Food effect: Intake 1h prior or 2 h after meal Absorption (t_{max}): **1.5 h** Time to steady-state ($t_{max,ss}$): **15 d** $AUC_{0-24,ss}$: **0.4 h*µg/mL** (22 %CV_S) $C_{max,ss}$: **22 ng/mL** (28 %CV_S) Clearance (CL/F): **5.4 L/h** (nc) Elimination half-life (t½): **90 h** [58-183]	RP2D: **60 mg od** (d1-21 q4w) (=MTD) BCS class I: (high permeability, high solubility) Food effect: none (intake with/without food) Absorption (t_{max}): **2.4 h** Time to steady-state ($t_{max,ss}$): **10 d** $AUC_{0-24,ss}$: **4.3 h*µg/mL** (61 %CV_S) $C_{max,ss}$: **273 ng/mL** (60 CV_S%) Clearance (CL/F): **13.8 L/h** (61% CV) Elimination half-life (t½): **44 h** [23-69]	RP2D: **45 mg td** (MTD: 60 mg td) BCS class: *nr* Food effect: none (intake with/without food) Absorption (t_{max}): **2.0 h** (1.5 h at 60 mg td) Time to steady-state ($t_{max,ss}$): **15 d** $AUC_{0-8,ss}$: **1.5 h*µg/mL** (nc) $C_{max,ss}$: **273 ng/mL** (65 %CV_S) Clearance (CL/F): *nr* Elimination half-life (t½): **8.7 h** (nc)

* no-dose-limiting toxicity recorded at 300 mg td

Fig 4: Table of BRAF mutation inhibitors

Drug treatments for BRAF V600E

Trametinib: Used in patients with unresectable or metastatic melanoma with BRAF V600E or V600K mutations, or metastatic non-small cell lung cancer with BRAF V600E mutation. Acts by blocking MEK molecules within the MAP kinase signaling pathway, which mediates cell proliferation and survival, and is often deregulated in cancer cells.
Encorafenib: Used in combination with binimetinib, for the treatment of patients with unresectable or metastatic melanoma with a BRAF V600E or V600K mutation, or for the treatment of adult patients with metastatic non-small cell lung cancer (NSCLC) with a BRAF V600E mutation.

Drug Treatments for BRAF V600E

Binimetinib: Used in combination with encorafenib, for the treatment of patients with unresectable or metastatic melanoma with a BRAF V600E or V600K mutation, or for the treatment of adult patients with metastatic non-small cell lung cancer (NSCLC) with a BRAF V600E mutation.

Vemurafenib: a kinase inhibitor used to treat patients with unresectable or metastatic melanoma in cases where the BRAF V600E mutation is found.

Trametinib

Molecular weight: 615.394g/mol
Chemical Formula: C26H23FIN5O4

- Kinase activated small molecule used to treat BRAF mutations
- FDA Approved since 2013
- Can be an irritant and hazardous to health, reproductive toxicity and harmful to organs

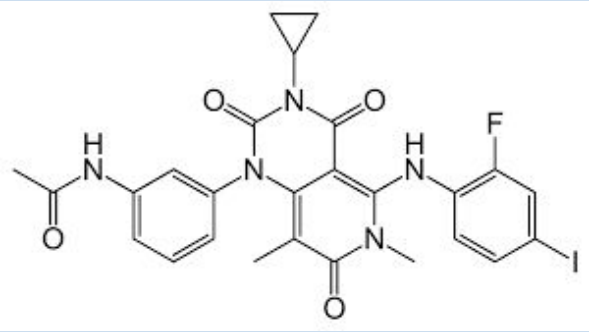


Fig 5: Trametinib chemical structure

Encorafenib

Molecular weight: 540.01g/mol
Chemical Formula: C22H27ClFN7O4S

- Kinase inhibiting small molecule, used to treat some types of skin cancer
- FDA approved since 2018 for use in combination with Binimetinib
- Can be hazardous to health, dangerous to reproductive health, can cause damage to organs

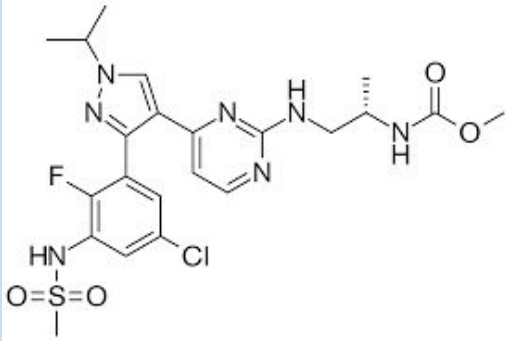


Fig:6 Encorafenib chemical structure

Binimetinib

Molecular weight: 441.233 g/mol
Chemical Formula: C17H15BrF2N4O3

- Used to treat metastatic melanoma with specific mutations
- FDA approved since 2018 for use in combination with Encorafenib

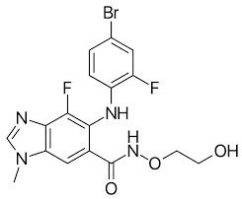


Fig 7 : Binimetinib chemical structure

Vemurafenib

Molecular weight: 489.9 g/mol
Molecular Formula: C23H18ClF2N3O3S

- A competitive kinase inhibitor with activity against BRAF kinase mutations such as V600E
- FDA approved since 2011 for treatment of metastatic melanoma with mutation on BRAF in the valine located in exon 15 at codon 600
- Can be an irritant

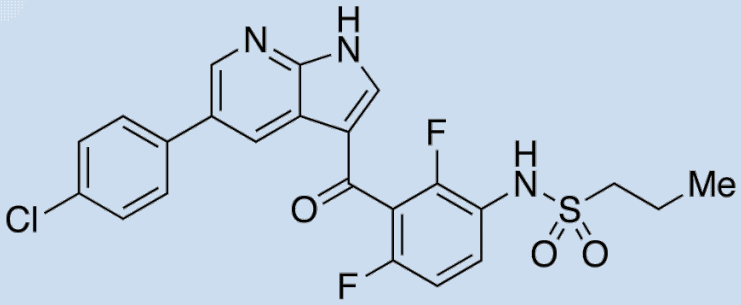


Fig 8: Vemurafenib chemical structure

Dataset

- Dataset consisted of CSV of chemical compounds that contain features and information for each and whether or not they inhibit BRAF
- Our dataset was given by the professor, with some information taken from PubChem
- Rows represent a chemical compound
- Columns represent features of each compound
- "Class" Column indicates if chemical is a BRAF inhibitor or not, will be used to train SVM classifier model so it can predict whether or not a compound is an inhibitor
- "1" = inhibitor "0"= not inhibitor

```
# view dataset
chemical_compounds_df
```

	CID	Class	PUBCHEM_XLOGP3_AA	PUBCHEM_EXACT_MASS	PUBCHEM_MOLECULAR_WEIGHT	PUBCHEM_CACTVS_TPSA	PUBCHEM_MONOISOTOPIC_WEIGHT	PUBCHEM_TOTAL_CHARGE
0	11574718	0	3.1	393.16885	393.43573	81.699997	393.16885	0
1	11588394	0	2.0	353.13754	353.37186	81.199997	353.13754	0
2	11595532	0	2.9	353.13754	353.37186	85.699997	353.13754	0
3	11603925	0	3.5	409.16376	409.43512	91.800003	409.16376	0
4	11624721	0	3.0	362.13788	362.38193	89.300003	362.13788	0
...	...	...	...	...	...	...	...	...
238	86766662	1	4.0	453.11584	453.48599	109.000000	453.11584	0
239	86766668	1	4.9	379.11322	379.35950	67.300003	379.11322	0
240	86766679	1	4.6	367.14331	367.40329	75.699997	367.14331	0
241	86766683	1	4.6	344.11609	344.36334	64.500000	344.11609	0
242	86766693	1	5.5	383.06369	383.77859	58.000000	383.06369	0

243 rows x 357 columns

Fig 9: A section of our dataset

Models and Methods

Models

- Currently have 2 models
- Model 1: does bare-bone preprocessing, including removing columns that are singular value or provide no variance, handles missing values, splits the data into train and test set.
- Model 2: model that does standardized scaling and steps

Methods

- Performed EDA on the dataset to understand it, this needed to be done so we could figure out what kind of preprocessing steps we needed to take
- We split the data into train and test set using 80% for train and 20% for the test set
- Created an interface to take a CSV input and describe the expected input

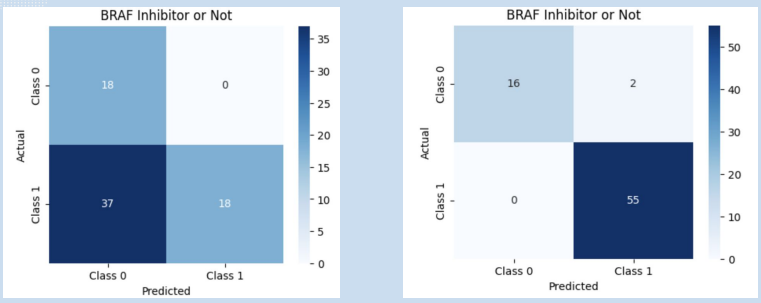


Fig 10: Our results for model 1(left) and model 2(right)

References

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