Welcome to the second activity of our escape game!

You have identified your protein of interest – Huntingtin – and now you wish to obtain a pure sample of this protein to study further. Our lab has expressed one isoform of the Huntingtin gene in insect cells using Baculovirus.

Your overall goal is to find a strategy to purify this protein. To help you, our lab has created a Python script called **PurifyMe.py**.

Unfortunately, a previous intern got into the files and messed up the location of some other scripts that PurifyMe needs to run.

Your first task, therefore, is to search the lab's server for the necessary files and move them to the same folder as the PurifyMe script (**~/HEAD/05-PurifyMe**)

The files needed are:

1. **affinity.py**
2. **anion_xchange.py**
3. **cation_xchange.py**
4. **SEC.py**
5. **HTT_plasmid.png**
6. **HTT-2D-PAGE.png**

Here are some Linux commands in case you aren't familiar with the system:

Command syntax	Purpose
pwd	To see your current address
cd my_folder1/my_folder2	To change your address to the one you specify (ex. move to my_folder2 inside my_folder1)
ls	List the contents of the current folder
mv my_file my_folder1/	Move a file or folder to another location (ex. move my_file to my_folder1)
.	Your current directory when specifying a path
..	The directory just above when specifying a path

Once you have moved all 6 files to the same folder as **PurifyMe.py**, you can launch the Python script. Make sure your current directory is the folder containing **PurifyMe.py** then use the following code:

python.exe ./PurifyMe.py

You are now ready for part 2 of this activity!

The PurifyMe programme will first allow you to explore different chromatography techniques available in our lab. You can read a brief description of the principles of these techniques if you are not already familiar with them.

Once you are ready, follow the instructions on the screen to proceed to the second section where you will choose 2 columns to purify the Huntingtin protein. You may find some useful information below.

Choose the correct columns with the correct strategy and you will purify your protein and obtain the secret code to help you in the escape game.

Biochemical information about Huntingtin:

- pI (isoelectric point) = 5.81
- Molecular weight = 348 kDa
- Number of amino acids = 3142, of which 70 are cysteines
- Extinction coefficient at 280 nm in water = 272 755 M^{-1} cm^{-1}

Information about the construct used to express HTT:

You previously recovered the file containing the image of the construct. To view the image, use the command **display HTT_plasmid.png** once you are inside the same folder.

Tip : You can do this in another tab or window so you don't have to close the PurifyMe programme.

Additional information about the cell extract from insect cells:

A previous intern performed a 2D-PAGE using a similar expression system. You previously recovered the file containing an image of the gel. To view the image, use the command **display HTT-2D-PAGE.png** once you are inside the same folder.

The 2D-PAGE could give you some clues about the native proteins contaminating your sample. You can assume all the proteins are monomeric.