**NB204 Principal Component Analysis (PCA)** *in class exercise*

Your job is to develop some insight into how this group of 58 neurons are responding to an odor stimulus. First we will explore the data a bit by plotting the responses. We will try to describe the type of information that is conveyed by the neurons. We will determine whether these neurons fall into categories (clusters). We will perform PCA to reduce the dimensionality of the data set and examine the output of this analysis tool. By the end of this in class assignment we hope that you will have developed some intuition about dimensionality reduction techniques and specifically understand the inputs and outputs of PCA.

You are provided with the responses of 58 simulated neurons to a set of stimuli consisting of steps and ramps of stimulus intensity. We provide the responses to these stimuli as peristimulus time histograms (PSTHs). Your goal is to use PCA to reduce the dimensionality of the data from 58 neurons to a smaller number of ‘principal component neurons’ which capture most of the variance of the original data. You will do this by completing some code that we have started for you. Follow along in the code corresponding to each section of questions.

You are provided with three files:

**• pca\_neurons.m** This is the main script for performing PCA on the neurons and producing figures of your results. There are detailed instructions of how to complete the code provided in the file. As indicated you will need to complete code in places marked “Insert/Modify code here”. There are examples of how to load the data, plot and save figures.

**• pca\_data.mat** contains the data you will perform PCA on. A description of the data format is provided in the pca\_neurons.m script.

*Getting started*

• Open up MATLAB.

• Set the current folder to include the .m and .mat files we sent you.

• Open pca\_neurons.m

• Read the description at the beginning of the script and then run the ‘Load data’, ‘Plot data...’ and ‘Save

figure’ parts of the code and make sure you understand them.

• Take a look at the data that is plotted.

*Dimensionality Reduction Exercise*

**PART I.** *Initial exploration and visualization of data*

1. Plot the stimulus vs. time. What are the key features of the stimulus?
2. Plot the responses of some of the neurons. What type of responses do you observe? Are there repeating patterns across neurons?
3. Think of ways to plot the data for all 58 neurons at once. Does this reveal more about what types of responses are present and if there are categories (or clusters) of neurons with similar response types? (hint, you can try using the imagesc function)
4. What types of information could you read out from this population?

**PART II.** *Clustering*

1. How similar or different are each of these neurons’ responses? Are there pairs of neurons that have highly similar response profiles? What is an analysis we could do to address this question? What are some similarity metrics that we could use to compare neurons? Explore approaches to address this topic based on your ideas.
2. One method to compare how similar neurons are to one another is to look at the neuron-neuron covariance matrix. If you are not familiar with covariance, please read about it with a quick internet search. Then look at the code to see the introduction to the covariance analysis and perform the covariance steps in the code.
3. Does there appear to be clustered groups with different types of responses?
4. Now let’s perform clustering on the covariance matrix using k-means clustering. Read the Matlab documentation for the kmeans function. Use as input to the kmeans function the covariance matrix.
5. How many clusters will you specify as an input? Can you think of iterative approaches to determine how many clusters to use?
6. Plot the responses of all neurons in a given cluster. Does the clustering seem to have done a good job? Compare these responses to the stimulus vs. time plot. What does each cluster of neurons appear to represent regarding the stimulus.

**PART III.** *Perform PCA*

1. Describe in detail the input and output of PCA, specifying what the rows and columns of each input (data) and output (coeff, score, latent, tsquared, explained, mu) variable correspond to. Use the pca help function.
2. Show a plot of the percentage of the variance in the data explained by each PC, versus PC number; this is sometimes called a “scree plot”. Take an educated guess: how many PCs account for a statistically significant percentage of the variance in the data? We are asking you to make an estimate based on looking at the scree plot. Please explain your reasoning. What do the non-significant PCs represent?
3. What might be a principled way to determine the number of PCs that are statistically significant? (Hint: think about how you might be able to create a fake data set that embodies a useful “null hypothesis” that you could compare your data set to. It is not necessary for you to implement this suggestion; we just want you to describe what you would do in a clear and detailed manner.)

**PART IV.** *Interpreting the output of PCA*

1. Show a plot of the first few PC scores versus time. Plot them side by side with the stimulus as a function of time. Describe each of these PC scores. What part(s) of the stimulus does each PC correspond to? Try to give each one a categorical descriptive label.
2. Show a plot of the covariance matrix of the PC scores. Briefly describe this matrix. What can you conclude from the appearance of this matrix?
3. Show a 3D plot of each neuron’s loadings onto the first three PCs. Do different neurons seem to cluster? What does this structure mean? (Hint: look back at the PC scores and think about your descriptive labels for each one.)

**PART V.** *Trajectories in PC space*

1. We have used PCA to look for groups of neurons and key response features in the population. In various lectures, we discussed the idea of looking at neural trajectories, that is time-varying activity pattern trajectories in a population of neurons. With 58 neurons (and thus 58 dimensions) it is difficult to visualize how the pattern of activity in the population changes over time. But, with PCA, we can look at three PCs to see how those vary across time as a dimensionality-reduced visualization of population activity trajectories.
2. We will plot a ‘movie’ of how the stimulus evolves over time and how the 3-dimensional neural activity in PC space evolves over time. Code is written to show the evolution of the stimulus over time. Now fill in the code to make a 3-D plot of the first 3 PCs iteratively over time. This will allow you to compare how neural population activity evolves relative to stimulus presentation. What do you observe from the neural activity trajectories?
3. Does the stimulus history affect the instantaneous stimulus response? In other words, when the fly is exposed to the exact same odor concentration in the middle of a ramp up as during a ramp down, do the neurons respond the same way?
4. Are there ‘fixed points’ in this space? If so, where are they?

*Extension problems (optional)*

1. The first step in performing PCA is almost always to standardize the data – i.e., to make different samples more comparable to each other by making them conform to some standard format. The MATLAB ‘pca’ function centers the data (by subtracting the mean), but it doesn’t perform any additional standardization before performing PCA. Sometimes, however, we might want to perform an additional layer of standardization by scaling each vector by its own standard deviation. (In other words, we would be z-scoring the data before performing PCA.) Here, this would amount to dividingeach neuron’s response by its own standard deviation (computed across all time points). Briefly describe the pros and cons of z-scoring in the case of the data set you are working with in this exercise. Can you think of a case in neuroscience where z-scoring is important – i.e., a case where the results of PCA are pretty meaningless unless you z-score before performing PCA?
2. Implement the proposal you made in problem 8 (part 3) above to determine the number of PCs that are statistically significant. Describe and show the result. Was the result what you expected?
3. What happens if you only use one of the “ramping” portions of the stimulus response to calculate the PCs? Why might this be?