UNIVERSITÀ DEGLI STUDI
DI GENOVA

NEURAL SIGNAL ANALYSIS - Master in Bioengineering

ANALYSIS ON EXPERIMENTAL NEURAL DATASET

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PROJECT IDEA

The given data consists of 3 different samples coming from two different populations: control rats (SHAM) and injured rats (ET1). The rats are stimulated in the primary somatosensory area (S1) and activity is recorded in both S1 and rostral forelimb area (RFA). The lesion in ET1 rats is performed in the caudal forelimb area (CFA). This fact arises the following question: 'Which differences does the lesion induce between both populations?'. In terms of connectivity, we expect that the connection between the areas considered would be affected by the lesion. In particular, by analyzing the population activity we should detect a decrease in the activity of the not stimulated area. With that being said, one way of studying the connectivity could be to compare the differences between PSTH areas before and after the lesion in both populations, to visualize the difference between both mappings, and try to confirm the following hypothesis:

'The lesion produces a decrease in neural activity.'

This means that we will see some difference between pre and post lesion in ET1, but no in SHAM.

METHODOLOGY

As previously commented, we have a total of 6 samples. For each area in every rat, we analyze the activity coming from 16 channels. Before conducting the PSTH analysis, we have to perform the spike detection. The first step consists in filtering the signals: we used a band-pass filter with cutoff frequencies of 300 Hz and 3000 Hz, in order to attenuate brain oscillations. Moreover, the stimulation was delivered every 5 seconds, by adding artifacts to the signals. Thus, by detecting local maxima with a high threshold (2000 µV), the artifacts were detected, and the signal was set to zero for 5 ms starting from the detected artifact, in order to remove further oscillations. Then, we applied to each channel a differential threshold algorithm yielding the spike trains. After different attempts, the threshold was set to 6 times the standard deviation of each signal in the first 2.5 seconds, in order to catch only the background noise and spontaneous activity. The sliding window, instead, was set to 2 milliseconds.

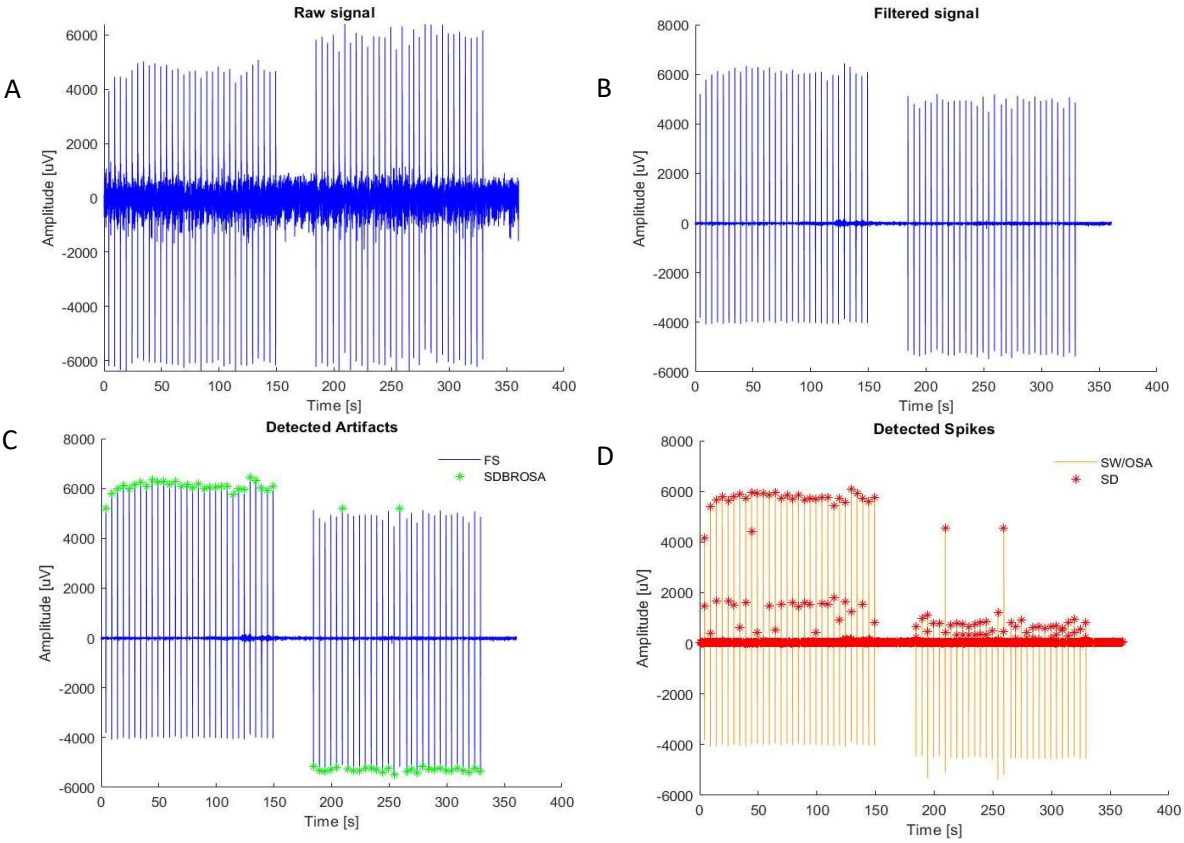

Fig. 1 - Different steps for processing one signal. (A) Raw signal. (B) Filtered Signal. (C) Artifact Detection. (D) Spike Detection

At this point, the PSTH can be computed. We chose a time window of 400 milliseconds and time bins of 4 milliseconds. Thus, in a time window after each stimulation (assumed to be each artifact) the number of spikes inside each bin is counted. Then, the spike count is averaged across the bin size and the number of trials. Once we have the PSTH plots, we compute for each channel the area under the curve. At this point we decided to purse two different methods to compare these areas. The first one involved the comparison of the area differences between pre and post lesion. Therefore, we computed the differences by subtracting the pre-lesion PSTH area from that of the post-lesion one

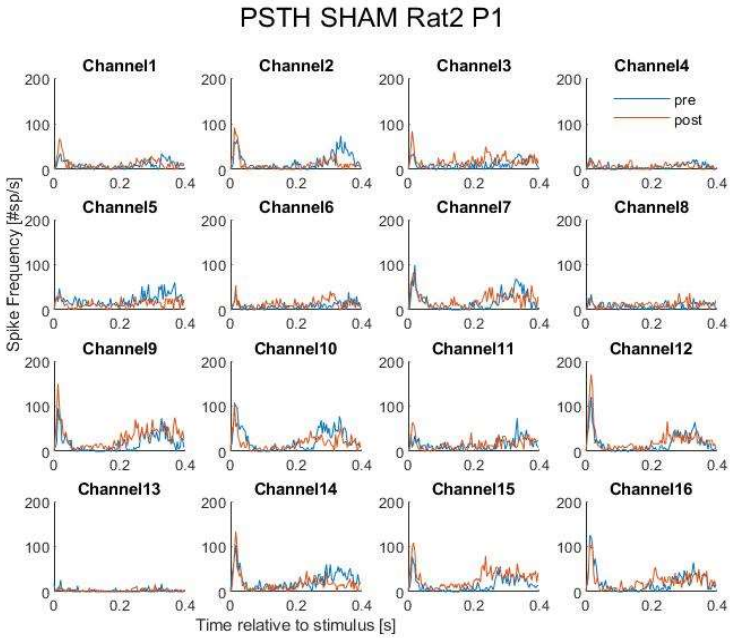

Fig. 2 - PSTH plots of all 16 channels of one of the control rats in P2

and then dividing each difference for the pre-lesion area, in order to obtain the percentage value. A negative value means a decrease in the area, conversely a positive value points to an increase. Thus, the two populations were compared for each brain region. The second method consisted in plotting the obtained areas in a scatter plot, whose axis are the area pre lesion and the area post lesion, as shown in figure 3. Then, we computed the regression line passing from the origin for the data points of each rat by means of the MATLAB *backslash* operator. The slopes of the injured rats against the slopes of the control ones were compared. In this way, a slope less than 1 means a diminution of population activity, whilst a slope bigger than 1 stands for an increase in activity. Also in this case, we distinguished between the two brain regions.

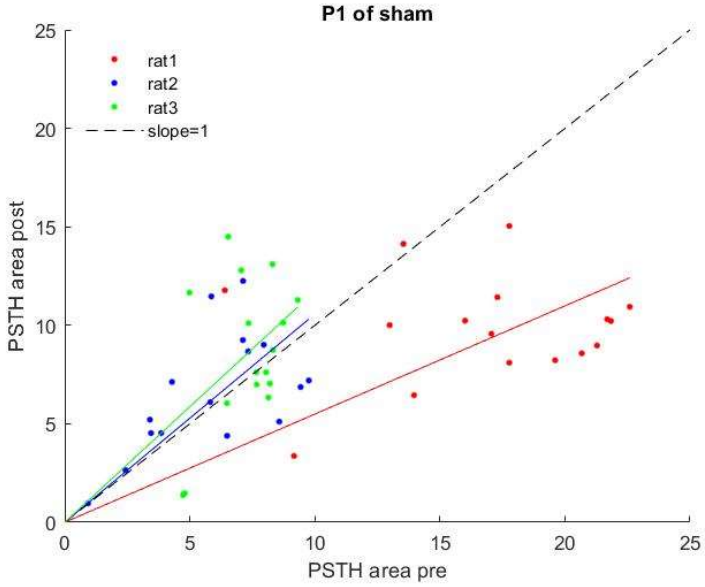

Fig. 3 - Scatter plot of the PSTH areas of the three control rats in P1. Each rat is coded with a different color and for each one the regression line is plotted.

In both methods, the data obtained were compared by means of a t-test, applied between control and injured rats. Thus, we considered two distributions containing the results of all samples in both populations. The t-test assumptions were verified by means of MATLAB *vartest2*.

RESULTS

We considered separately the two populations, and we built the histograms of PSTH area difference for both P1 and P2. By looking at the figure 4, in P1 both distributions are shifted towards negative

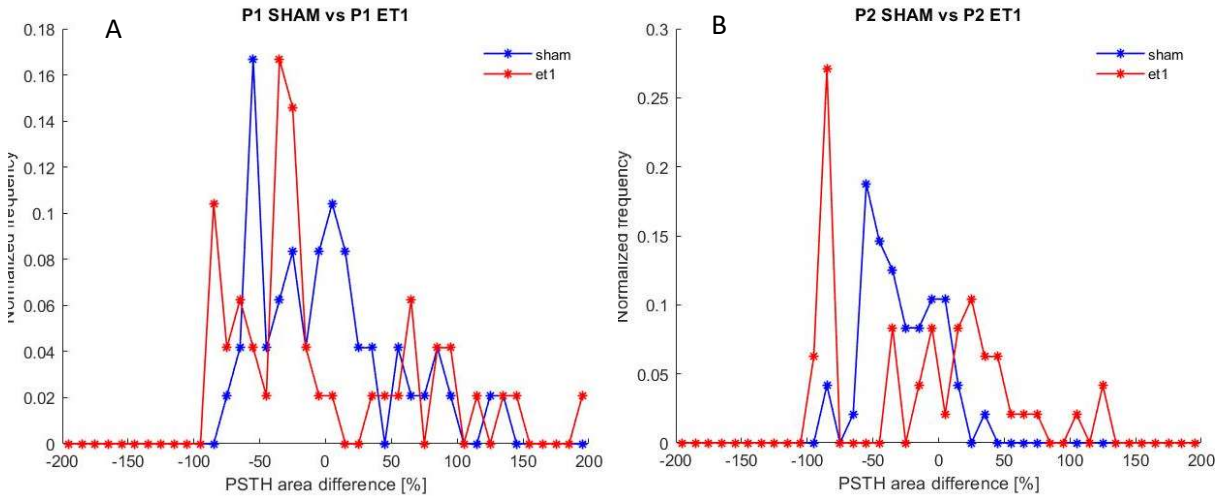

Fig. 4- (A) Distributions of PSTH area difference in P1 of both SHAM and ET1. (B) Distributions of PSTH area difference in P2 of both SHAM and ET1.

values, meaning a possible decrease in population activity. However, the distributions are too irregular to be described just by looking at the graphs. Therefore, in order to see if there is a significant statistical difference between control and injured rats, we performed a t-test. First, we tested the assumptions of the t-test on the data. Both normality and homogeneity of variance were tested with *vartest2*, but they were verified ($p<0.05$) only for P1. By performing the t-test on P1, we get $p>0.05$ resulting in the acceptance of the null hypothesis, meaning that in P1 the two distributions have equal mean values. Since no difference is found we cannot say that the lesion produced a reduction of activity in injured rats. As for the other method, we took into account the slopes of the regression lines passing from the origin to compare control and injured (figures 5). Theoretically, regression lines

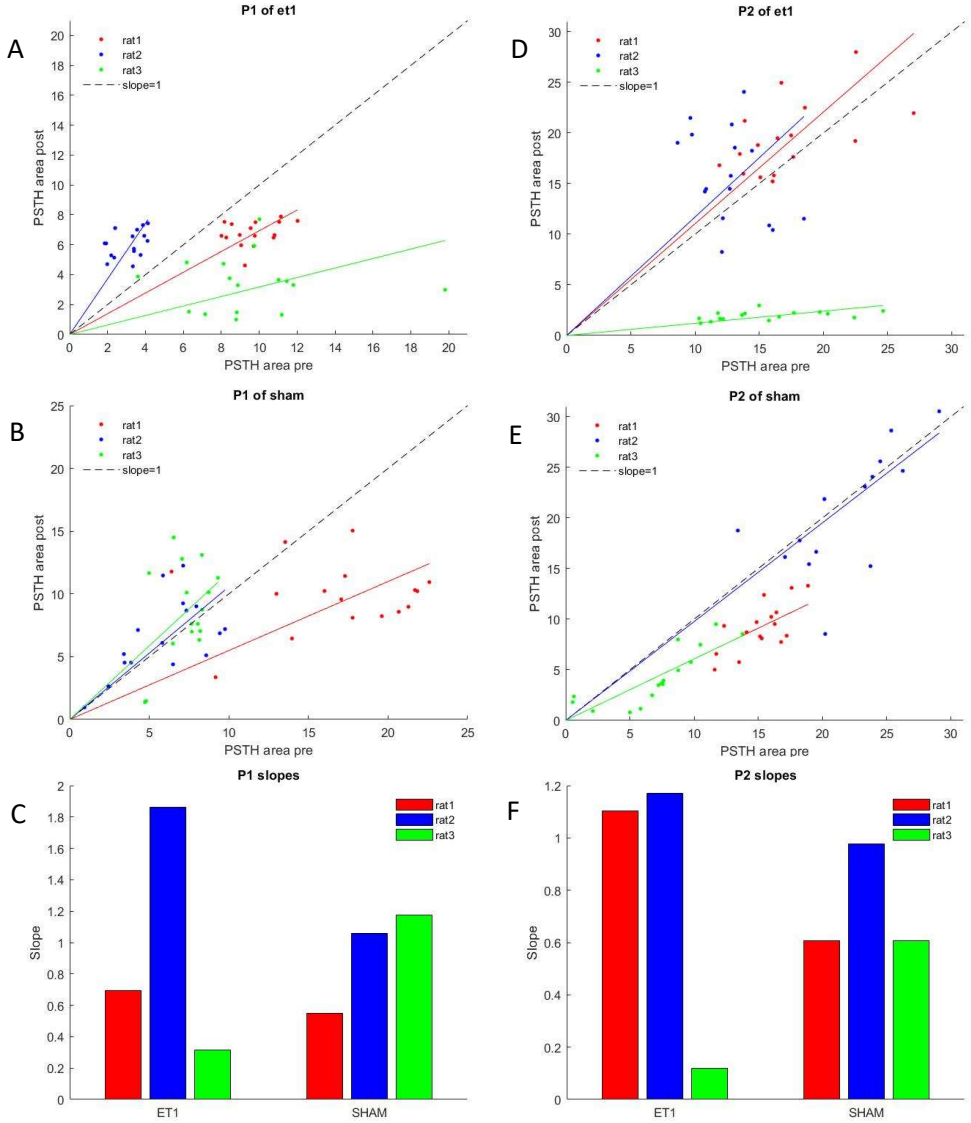

Fig. 5 – (A-B-D-E) Scatter plots of the area values in both populations divided based on the brain region. (C-F) Histograms of the slope values in P1 (C) and P2 (F).

in SHAM should correspond to the bisector, since the PSTH areas should not change after the lesion, while ET1 lines in P1 should have a slope lower than one, since ET1 activity in P1 should decrease after the lesion. However, the plots show that we cannot derive a unifying conclusion, indeed the data strongly depend on the rat considered. For instance, if we look at figure 5A, in can be noticed that rat 2 shows an increase in the PSTH areas in contrast to the other two rats. Therefore, we decided to perform the t-test on the slope distributions. However, either for P1 and for P2 the test results in

p>0.05, meaning no statistical difference is found between control and injured rats. For completeness' sake, the same procedure was repeated without distinction between the single rats (figure 6). What

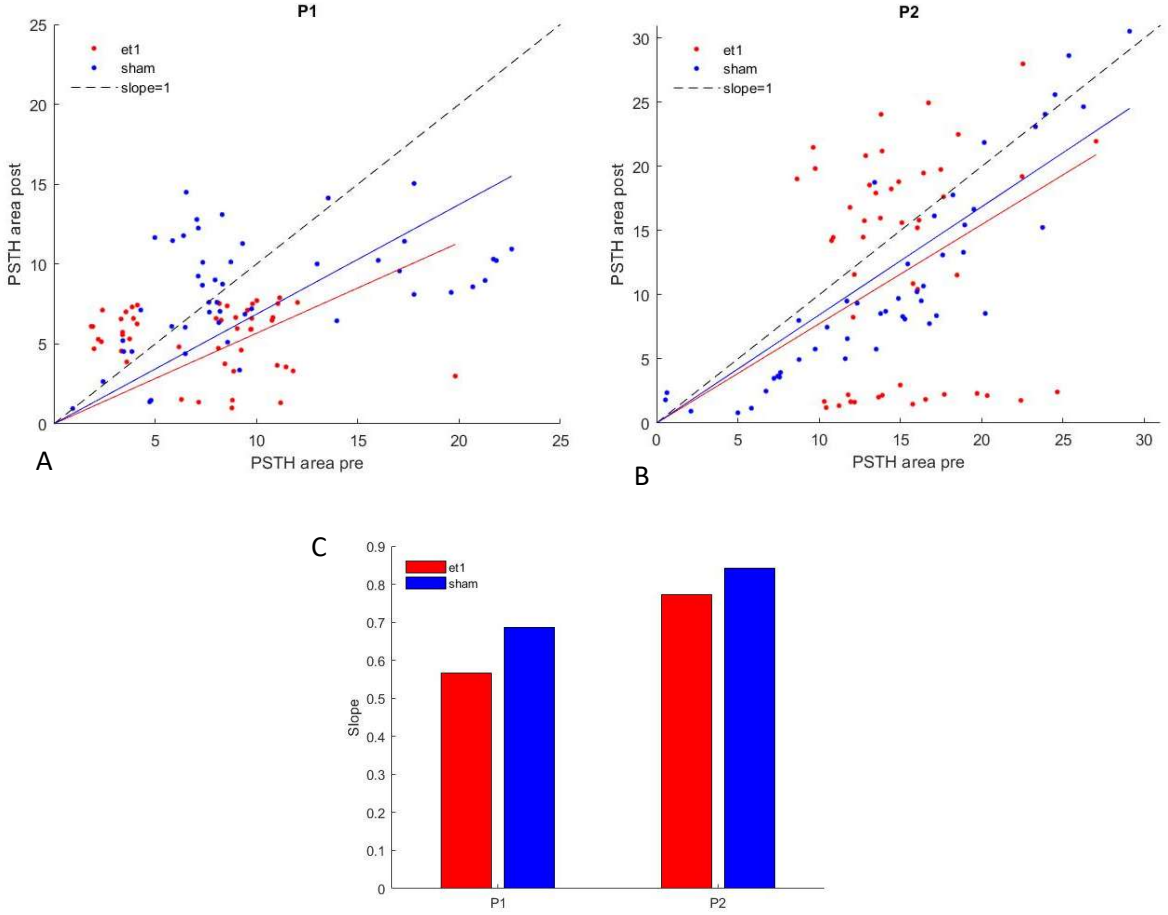

Fig. 6 – (A) Scatter plot of PSTH areas in P1. (B) Scatter plot of PSTH areas in P2. (C) Comparison of the slopes of ET1 and SHAM in both areas.

is found by looking at the graphs is that, overall, in both populations the amount of activity spontaneously decreases. This is true even for P2, where the population activity is not supposed to change, since it's the stimulated region.

FINAL DISCUSSION

As it has been discussed in the previous sections, the goal of this experiments was to analyze and interpretate which are the effects of the lesion, if it was able to disrupt the communication inside both areas, affecting on the number of spikes produced under a certain stimulation. By creating the PSTH and its surrounding analysis it has been noticed that every injured animal suffers a decrease of the value of those histograms comparing pre and post lesion mappings. However, from this experiment we cannot firmly assess that this is always valid since we did not find a significant difference with the control animals. For this particular reason, the research can be extended in the near future with more samples, trying to find a general assumption for the lesions occurring in that area of the brain and affecting the communication in both analyzed areas, P1 and P2.

BIBLIOGRAPHY

- Chiappalone, M., Massobrio, P. and Martinoia, S. (2008), Network plasticity in cortical assemblies. European Journal of Neuroscience, 28: 221-237. https://doi.org/10.1111/j.1460-9568.2008.06259.x