

Interactive software for the sonification of neuronal activity

Argan Verrier

Sorbonne Université
argan.verrier@etu.upmc.fr

Elim Hong

Neuroscience Paris Seine
Institut de Biologie Paris Seine
Sorbonne Université
elim.hong@inserm.fr

Vincent Goudard

Institut d'Alembert
LAM
Sorbonne Université
vincent.goudard@sorbonne-universite.fr

Hugues Genevois

Institut d'Alembert
LAM
Sorbonne Université
genevois@lam.jussieu.fr

ABSTRACT

We present a software for the sonification of neuronal activity acquired by calcium imaging of neurons in the brain of zebrafish larvae. The objective was to facilitate the observation of the temporal and spatial patterns of activity as well as the synchronous phenomena between different neurons. The parameter mapping approach was used. The method we present relies on manually drawn regions of interest (ROI), whereby, the average fluorescent values of the ROIs are computed to drive FM synthesis parameters. The ROIs represent neurons and each ROI is associated with a FM synthesizer producing sound signal. An interface was developed to allow one to tune the parameters of the sonification and manage the data sets. This paper presents the different mapping that were tested, their implementation in the software, and the interface that was developed.

1. INTRODUCTION

There are 86 billion neurons in the human brain. We are only starting to understand how these neurons are connected and how they function to drive movement, emotions and cognitive abilities. To better understand how the brain works, scientists use model organisms including rodents, fish, and flies. Many studies have shown the similarities between different animal species with humans. Recently, the zebrafish has become a popular model system to study brain function due to its transparency and small size as there are 'only' around 100,000 neurons in the larval brain. Brain activity can be monitored using genetically encoded calcium sensor, GCaMP, which can be expressed only in subset of neurons [1]. When neurons become active, it results in an increase in calcium levels. When calcium binds to GCaMP, it causes the protein to increase its fluorescence intensity. In other words, we can monitor the activity of

neurons by measuring the changes in its fluorescence levels using GCaMP. This technique is called calcium imaging and is especially powerful as it allows the monitoring of real-time activity in as many neurons as can be imaged under a microscope at a single cell level. However, the downside is that due to the large number of neurons, it is difficult to find the best approach to recognize the different types of activity in the brain. This work was initiated to find a novel method to more easily recognize and differentiate the diverse activity of hundreds of neurons.

This paper presents a software for the sonification of calcium imaging data. This project is a collaboration between the LAM (Lutheries-Acoustique-Musique) team of the Institut Jean Le Rond d'Alembert - Sorbonne université and the FINN (Formation and Interaction of Neural Networks) team in the department of Neuroscience Paris Seine of the Institut de Biologie Paris-Seine - Sorbonne Université.

The goal of this project was to study if sound information in addition to visual information would help clarify the different types of neuronal activity. The device was designed to facilitate the recognition of synchronous neuronal activity in the nervous system [2], as well as their temporal and spatial activity patterns. A special attention was given to the ease of use of the software and the aesthetic aspect of the sonification.

The parameter mapping was used along with an FM synthesis. Several mapping methods were tested to study their effect in processing the data. We firstly present the state-of-the-art of the sonification approach, mainly focusing on the imaging application. Next, we present the material and methods used in this study. We then introduce the software and the mapping method used, implementation of the sonification, and finally the results.

2. STATE OF THE ART

Sonification refers to the representation of data using a sound signal, in the same way that *visualization* is the representation of data using image. In this section, we present the main sonification techniques and their applications in the medical imaging field.

Both sonification and visualization are intended to explore data sets. In [3] Hermann states that sonification is well suited for large data sets hard to represent visually, as well as allowing perception of patterns and structures within the data. Sonification methods have a large field of applications, ranging from the sonification of the center of gravity of a skier [4] to the monitoring of electrical grids [5]. In [6] Hermann presents the main sonification techniques : *audification*, *earcon*, *auditory icon*, *parameter mapping* and *model based sonification (MBS)*. *Audification* consists in "the direct translation of the waveform of data into sound" [7] : the data, audible or not, is transformed to be heard by the user. *Auditory icon* and *earcon* are sound events related to data or interactions. MBS consists of creating a numeric model based on the data properties, which produces a sound signal when excited by the user [6].

Parameter mapping consists of driving the parameters of an audio synthesis with parameters of the data to sonify [6]. According to [8] it is the most widely used sonification technique and studies about the perception of different mappings have been carried out [9, 10]. One of the main issues encountered is the non linear perception of some sound characteristics [10], especially the pitch, which is the most commonly used [8], as well as the perception of the mapping, which may not be intuitive.

Medicine is an important field of application. Data such as neuronal activity or heart beat can be sonified to help the physician with a diagnostic. It is interesting to note that these methods are usually used to complement visual information.

In [11] the authors present a method to facilitate the diagnosis of Alzheimer's dementia. The parameter mapping is used along with PET/CT scan imaging. The activity of two affected lobes is compared to a reference, unaffected lobe for each patient. Each lobe is allocated an oscillator, the reference lobe with a base reference frequency. The frequencies of the two affected lobes are deviations from the base frequency, calculated based on the deviation of activity between each lobe with the reference lobe. The interaction between the different frequencies results in an audible beating pattern, which differed according to the severity of the disease progression.

In [12] the authors propose a method to help find anomalies in pictures from medical imaging using statistical descriptors, applied to mammography. Parameter mapping is used and the descriptors drive synthesis parameters. The user explores the picture with a moving region of interest on which the descriptors are computed.

The authors of [13] present two sonification methods using an arbitrary number of voxels for functional magnetic resonance imaging (fMRI). The first method uses a low number of voxels to drive pitched notes, and thus produces evolving chords. The second method uses a large number of voxels to control a large number of notes to obtain an evolving sound texture.

Finally, in [14] the authors present different approaches to help identify cells affected by cervical cancer. They use parameter mapping and present several mapping methods

to be used with visual information. It is interesting to note that they take into account the aesthetics of the produced sound, since the cytologists who use these methods would need to listen to the sound for hours.

3. MATERIALS AND METHODS

3.1 Calcium imaging data

The data set used for the sonification was from calcium imaging of spontaneous neuronal activity of habenular neurons in the brain of larval zebrafish carried out in the Hong lab. Spontaneous neuronal activity refers to endogenous neuronal activity without any external stimuli. The habenula is an evolutionarily conserved area of the vertebrate brain important for mediating aversive emotions [15].

The transgenic line *Tg(gng8:gal4ff;UAS:GCaMP7a)* [16], expresses the GCaMP calcium sensor only in habenular neurons. The fish were maintained at 28°C on a 14:10h light:dark cycle in a recirculating system. All experiments were carried out in agreement with the European Directive 210/63/EU and the French application decree "Décret 2013-113". The project has been approved by the ethical committee "Comité d'éthique Charles Darwin"(APAFIS15909-2018070912072530 v5). The fish facility has been approved by the 'Service for animal protection and health', (A-75-05-25).

6 and 7 days old larvae were used for experiments. They were anesthetized in Tricaine (0.01%), their brains were dissected using fine forceps in Ringer's solution (134mM NaCl, 2.1 KCl, 1.2mM MgCl₂, 10 mM HEPES, 10mM Glucose, 2.1 CaCl₂, pH = 7.6) and mounted in 1.2% low-melt agarose.

The calcium imaging was performed on a spinning disk microscope (Zeiss Axio Examiner.Z1) with a 40X water immersion objective (NA=0.95) at a rate of 5Hz. As described in [16] the average fluorescence intensity of habenular neuron soma were calculated using manually drawn region of interest (ROI) in FIJI [17]. The average fluorescence intensity over each ROI was then extracted and analyzed with custom-written Matlab scripts. The amplitude of calcium activity was computed using the following relation:

$$\frac{\Delta f(t)}{f} = \frac{f_i(t) - f_{mean}}{f_{mean}} \quad (1)$$

Where $f_i(t)$ is the mean intensity over the i^{th} ROI at a given time (the *signal* of the ROI), and f_{mean} is the mean value of the ROI over the entire recording. K-means clustering was carried out on Matlab with *kmeans* function, k=5 was selected based on correlation coefficient values and manual analysis of calcium activity using a heatmap.

The data set consisted of mean values computed over the ROIs for each frames. The number of ROIs ranged from 129 to 198. They are represented as a N_{ROI} -dimension discrete signal of time sampled at the rate of 5Hz. The length of the recordings was around 3000 frames.

Additional materials used: the video generated from the recording, the location of the ROIs on the videos and the

clusters extracted using the k-means clustering, which consisted of ROIs allocated to different clusters.

The sonification software was developed using Max/MSP, which allows quick prototyping and interaction with the user. The data was formatted using custom written Matlab scripts.

3.2 Goals and constraints

The sonification is intended to be used with the associated video : the auditory information is complementary to the visual information.

The two main goals for the sonification were the synchronization between neurons (neuron groups), and activation patterns (spatial and temporal). The aesthetic of the sound was another point of focus for this project. Another aspect of the sonification we focused on was the interaction with the user. Rather than passively listening to the resulting sound signal, the user is able to interact and explore the data using the sound. The user is able to select the data sets to sonify and also set the parameters of the sonification to be used with different sets of data. In summary, we strived to make the the software as easy to set-up and use without prior experience.

The parameter mapping approach was chosen as it was well documented, and studies on the perception of different mapping were previously carried out. MBS and audification approaches were excluded because of our high dimension data set.

We focused on transition of the values from a base value, corresponding to a non-active ROI, to a high value corresponding to an active ROI.

4. THE SOFTWARE

In this section, we describe the software and the sonification approach developed. The materials used are the averaged fluorescence intensity computed over each ROI with the coordinates of the ROIs, and the video recording of the calcium (neuronal) activity.

4.1 Overview

The software consists of an interface which allows the user to set the sonification parameters, display the calcium activity recording (movie) and provide the audio feedback corresponding to the sonified data.

The approach used for the sonification is based on manually drawn regions of interest (ROIs). Each ROI is represented by an FM synthesizer whereby the carrier is obtained by reading a wavetable at a pitched frequency f_c , the *note* of the ROI. This frequency is modulated by a signal obtained by reading another wavetable at the modulating frequency f_m . The parameters of the synthesis are driven by the properties of the signal of the corresponding ROI. More details about the mapping are given in Section 4.2.

The user interacts with the data by forming groups of ROI, which all have the same carrier frequency and a same wavetable used for the carrier signal. In the software, the groups of ROIs are represented by their colors. The audio signal is the summation of the signals produced by each

ROI. The overview of the sonification process is illustrated in Figure 1.

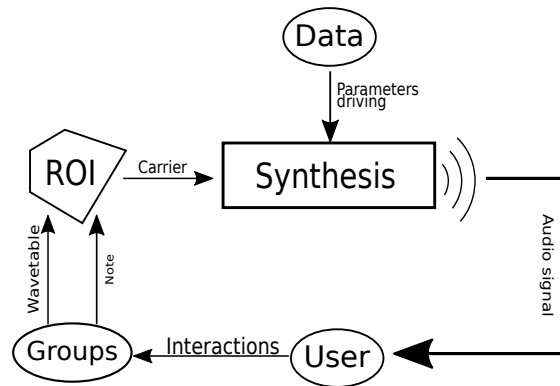


Figure 1. Schematic of the sonification approach. Each ROI is associated to an FM synthesizer where the parameters are driven by the corresponding signal. The user can interact by forming groups of ROIs and choosing the wavetables and notes.

For each group, the user chooses the wavetable of the carrier and the note. Before the sonification process, the signals are preprocessed and normalized. This process is described in Section 5.1.

The software itself is a Max/MSP patcher that contains a user interface (Figure 3), and a video display for the user to select the ROIs (Figure 2). Through the interface the user can set the sonification parameters, manage the data and ROI groups, and tune the video and audio feedback. The videos can be played at 5 frames per second (real time recording) to a default speed of 25 frames per second. This choice was arbitrary and can be modified by the user.

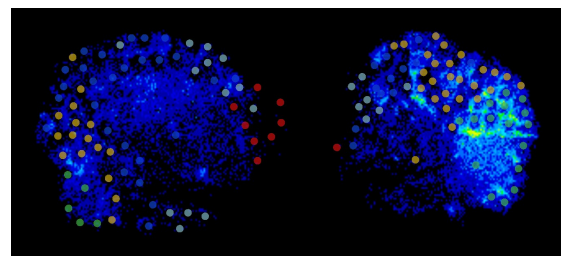


Figure 2. Video display of the calcium activity in the habenula, obtained as described in section 3.1. The ROIs are represented by colored dots, which the user can click to select for sonification.

4.2 Mapping

For a given ROI, the properties of the corresponding signal are used to drive the parameters of the FM synthesis. Four parameters are used: the amplitude A , the inverse of the harmonicity ratio h , the modulation index I and the panning (stereophonic or quadriphonic). The following variable t refers to discrete times sampled at audio rate

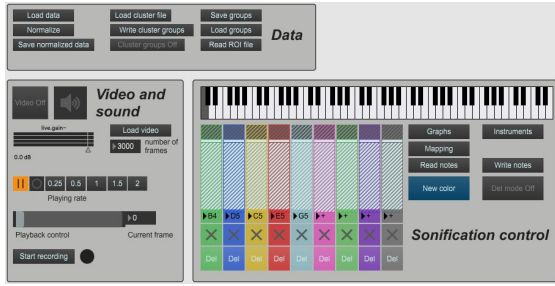


Figure 3. Main user interface. This panel allows the user to manage the data sets, set the audio and video feedback and select the ROI groups.

(32kHz or 44,1kHz). The driving parameters are computed for each frame of the video. The fluorescence value refers to the normalized and preprocessed signals as described in section 5.1.

We define h as the inverse of the harmonicity ratio:

$$h = \frac{f_m}{f_c} \quad (2)$$

The modulation index I is defined as

$$I = \frac{A_m}{f_m} \quad (3)$$

where A_m is the amplitude of the modulation signal.

The amplitude A is mapped to the fluorescence value, as it was assumed that the relationship between the intensity and the loudness of the sound would be intuitive. The inverse of the harmonicity ratio h is mapped to the rate of fluorescence change. This quantity is computed over two frames with the normalized and preprocessed fluorescence signals. The panning of the sound is mapped to the location of the ROI on the video. The effect of stereophony and quadriphony can be investigated. Lastly, the modulation index is mapped to an additional parameter that can be defined based on the ROI group. The mapping is summarised in Table 1.

Synthesis parameter	Driving parameter
Amplitude	Fluorescence value
h	Fluorescence rate of change
Modulation index	Group parameter
Panning	Location of the ROI

Table 1. Mapping the data to synthesis parameters. Four tested parameters are described in this table.

The group parameters used for this mapping are based on formulas that reflect the properties of the user-made ROI groups. The main purpose is to provide feedback to the users on the arbitrary groups they made. Four different parameters can be selected: average, standard deviation, activation ratio and activation ratio rate of change. The group parameters are computed for each frame of the recording. The parameters can be computed using the fluorescence values or its rate of change.

The average consists of the averaged chosen value over the whole values of a given group at a given frame. Thus, it can be expressed as

$$F_{gj} = \frac{\sum_g f_{ij}}{N_g} \quad (4)$$

where F_{gj} designates the value for the group g at the frame j , N_g is the number of ROIs in the group g , f_{ij} is the chosen value for the i^{th} ROI at the j^{th} frame and \sum_g is the summation over the group g . We note that when the rate of change is selected as the value to be processed, the parameter corresponds to the rate of change of the average fluorescence in the group computed over two frames. In the same way, the modulation index can be mapped to the standard deviation of the selected value, computed over the whole group. The activation ratio is the ratio of the active ROIs over the total number of ROIs in the group. The parameter is expressed as

$$F_{gj} = n_{gj}/N_g \quad (5)$$

where n_{gj} is the number of active ROIs in the group g at the j^{th} frame. N_g is the total number of ROIs in the group g . The rate of change of the activation ratio is computed over two frames.

5. IMPLEMENTATION

In this section we discuss the implementation of the sonification process into the Max/MSP patcher. The required material is the video of the calcium activity of habenular neurons. The ROIs are selected and average fluorescence intensity over each ROI calculated using FIJI. The files containing the average fluorescence intensity and the location of the ROIs are re-formatted using Matlab scripts¹. The user can either make their own ROI groups or use pre-identified clusters using the k-means clustering method.

Two more effects can be used: a detune feature, based on the rate of fluorescence change and a plucking-like sound event triggered by the fluorescence value.

5.1 Preprocessing and normalization

The current version of the software enables the user to pre-process the video and apply various noise-reduction and normalization processes, which are the first two steps of the sonification process. We are interested in the perception of the activation of the ROI, which is the transition from a low value (the base activity within the ROI), to a high value. The normalization process ensures that the signals are between 0 and 1, to allow one to use the software with different data sets without the need to re-scale the parameters.

Two methods were used to compute the processed values. The first method uses the average value and standard deviation of the signals computed over the whole recording. The processed signal corresponding to the i^{th} ROI $f_{pi}(t)$ is determined as

$$f_{pi}(t) = \frac{|f_i(t) - f_{imean}|}{f_{imean}} \quad (6)$$

¹ Credit for the ROI reading function goes to Dylan Muir.

Or

$$f_{pi}(t) = \frac{|f_i(t) - f_{imean}|}{\sigma_i} \quad (7)$$

where f_{imean} is the mean value of the signal over the whole recording and σ_i is the standard deviation of the same signal computed over the whole recording.

The other method is a running average of N frames. The number of frames to average can be determined by the user. Hence, the n^{th} sample of the signal of the i^{th} ROI f_{pi_n} is expressed as

$$f_{pi_n} = \frac{1}{N} \sum_{j=n}^{n+N} f_{i_j} \quad (8)$$

where f_{i_j} is the j^{th} sample of the i^{th} ROI.

Next, the data set is normalized. Three methods were used: normalization by the maximum value per frame, the maximum value over the whole data set, or the maximum value per signal. With the first method, the signal is normalized frame-by-frame by the maximum value of the data set in the current frame. Thus, the normalized value f_{ni_j} of the processed signal f_{pi} at the j^{th} frame can be written as

$$f_{ni_j} = f_{pi_j} / F_j \quad (9)$$

where F_j is the maximum value of the data set in the j^{th} frame.

The second method consists of normalizing every signal by the maximum value over the whole data.

Finally, the last method consists of normalizing the signals independently by their own maximum value over the entire recording. The choice of normalization can be selected by the user, and can be changed without reloading the data.

The normalized and processed signals are used for the sonification process. The rates of change are computed with these signals, and normalized using the third method.

5.2 Mapping the amplitude

The values used to drive the amplitude are the processed and normalized signals of the data set. However, the relationship between the amplitude of a sound signal and the loudness perceived by the auditor is not linear and may depend on the frequency [18]. Thus, describing the relationship between the data parameter and the driven synthesis parameter is a crucial part of the sonification design.

The property intended to be perceived with this mapping method is the temporal organization of the activated ROIs. Three steps were implemented. Firstly, a threshold was placed to exclude the values corresponding to a non-active state. The resulting signal was then sharpened (by analogy to the sharpening effect used in image processing) with its rate of change. Lastly, the signal was scaled to drive the amplitude. To further enhance the perception, a compressor is added after the synthesis.

For the sake of simplicity, the notation $f_{ni}(t)$ remains constant during the whole process.

The values that correspond to the non-active ROIs were set to 0. We considered the ROI active when its fluorescence value is above 1 standard deviation. Thus, the ROI

is active when the corresponding normalized signal $f_{ni}(t)$ is above the threshold value $k\sigma_i$ where σ_i is the standard deviation of $f_{ni}(t)$ and k is an arbitrary coefficient set by the user.

The discrimination is implemented by interpolating the signal's value with a curve defined by two points ($k\sigma_i$, 0) and (1, 1). A slope is added to smooth the transition. The output value is determined as

$$f_{out} = g(f_{ni}(t)) f_{ni}(t) \quad (10)$$

where $g(f_{ni})$ is the interpolated value.

The discriminated signal is then used to drive the amplitude. This is carried out in real time whereas the normalization and the preprocessing are done prior to the sonification. This allows the user to have a more dynamic control over the threshold. The main purpose of the discrimination process was to exclude the static sounds corresponding to the base fluorescence values to clarify the sonification.

The sharpening effect is intended to enhance the perception of the activation by adding its rate of change to the signal to intensify the event. The rate of change is scaled with an interpolating curve to amplify the effect and reject the lowest rate of change values. The output signal is determined as

$$f_{out} = f_{ni}(t) \left[1 + a \frac{df_{ni}(t)}{dt} \right] \quad (11)$$

where a is an arbitrary coefficient set by the user that controls the value added to the signal. We note that since the fluorescence decay is considered to be less significant, only the positive values of the rate of change were used.

The last transformation made to the fluorescence signal is the scaling. This step defines the relationship between the driving parameter and the driven parameter, the amplitude. The scaling is done by interpolating the signal with another curve. The purpose is to amplify the variation of the signal between a base state and an active state (Figure 4). The curve is made of three zones and is intended to

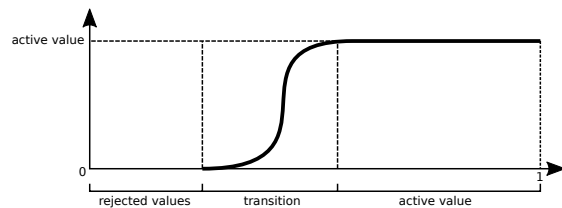


Figure 4. Interpolating curve used to scale the signal before driving the amplitude. Three zones are shown : the rejected values, the transition and the active value, which is constant.

work as a compressor, as it is meant to enhance the attack. The first zone consists of the rejected values. This gives a finer control over which values will be sonified. The next zone is the transition, whereby the slope controls the activation speed. A steeper curve will sharpen the transition as the opposite will smooth it. The last zone sets the value attributed to the active state.

5.3 Mapping the modulation index

In each frame, the fluorescence values and rate of change are calculated for each ROI. The selected values are routed to the appropriate group. The parameters associated for each group are then computed. Before driving the synthesis the parameter goes through two interpolating curves. The first scales the parameter between 0 and 1. The second curve scales the modulation index to drive the synthesis. Having two scaling curves instead of one allows the user to independently scale the data parameter and the modulation index. As each parameter are configured independently, the user is able to design the relationship between the parameter and the synthesis while keeping a same range of variation in the resulting sound. After scaling, the modulation index is multiplied by an arbitrary coefficient value for amplification. Convincing results were achieved with a value of 20 which allows the variations to be easily audible without adding too much brightness as it would make the information harder to distinguish and less aesthetically pleasing.

6. INTERFACE

In this section, we describe the interface, which enables the user to perform the steps to control the sonification : tuning the parameters, loading data sets, allocating ROIs to groups, managing the groups and choosing the wavetables. The user has the possibility to shape the wavetable, choose wavetables from audio files or even draw his own wavetables. We also kept in mind musical applications that could be achieved by tweaking the parameters and the wavetables, and the ease of use of the software.

Additional features were also implemented : saving and loading sets of groups and notes, buffers, ability to switch between clusters and groups, and monitoring the activity in specific ROIs.

The interface is built around three panels : the main panel (Figure 3), the mapping panel and the instrument panel (Figure 5). The main panel is the main interface where the user can access other panels as well as managing the data sets. The mapping panel contains the controls over the sonification parameter. The instrument panel allows the user to select the buffers and the waveshaping functions. The interface is completed by the video display where the user can select the ROIs.

The mapping panel is organized as a flowchart. Only the most important parameters are displayed : the scaling curves and the parameters of the sharpening and discrimination. The curve used for the discrimination and the parameters of the compressors are not accessible, as they were considered to be less sensitive to the data to sonify. Lastly, four presets were designed and implemented.

The instrument panel allows the user to set the buffers used as wavetables and waveshaping functions. While this choice is not crucial to the sonification, we wanted the user to be able to experiment for musical applications. We chose only to leave the most crucial information : the shape and the allocation of the groups. This feature is experimental and would need further work. The default waveforms

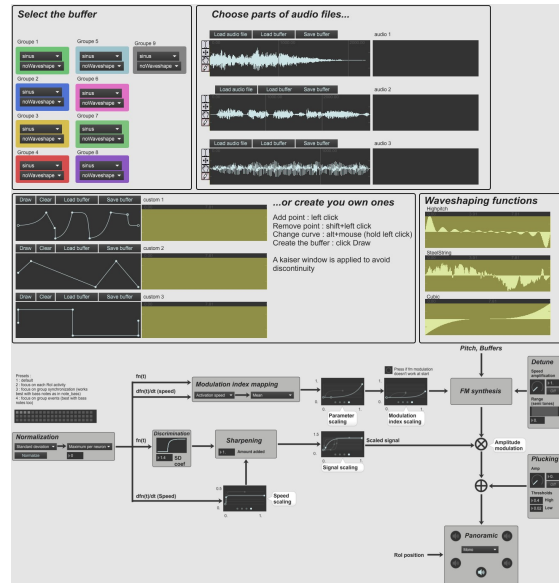


Figure 5. Instrument panel (top) and mapping panel (bottom). The mapping panel contains the settings to modify the mapping. It is organized as a flowchart : The top line represents the mapping to the modulation index, the middle line represents the mapping to the amplitude. The instrument panel allows the user to set the wavetables used by the different groups of ROIs, and apply waveshaping functions.

and waveshaping functions were sufficient and different waveforms were mainly used to highlight a certain group. However, the goal was to allow experimentation and musical applications, to have more control over the buffer and their parameters, and for the advanced user to use external software and load the results in the program.

7. RESULTS AND DISCUSSION

In this section we present and discuss the results of the software and the mapping presented above. In particular, we present how they were used to design four presets : 1) general purpose (default), 2) focus on the activity within individual ROIs, 3) focus on monitoring synchronicity within the ROIs grouped in cluster made groups, and 4) focus on monitoring consecutive group activation. Note that the evaluations presented in this section were established by the authors. Hence, these results are preliminary and requires further investigation.

7.1 Normalization and preprocessing

The choice of the normalization and preprocessing method allows the user to obtain various results.

The running average was not ideal as it tended to smooth most of the events. Average and standard deviation normalization produced similar sounding results. Moreover, they tended to reduce static notes being played, which was aesthetically more convincing. As we were mainly inter-

ested in variations in the signal, these two methods were used for the four presets.

The normalization by the maximum per frame tended to smooth the variation from frame to frame. This was sub-optimal as we are mostly interested in the variation.

Lastly, the normalization by the maximum value over the whole data tended to emphasize the variations of intensity from one signal to another. Since we were more interested in the activation patterns and not the relative amplitude of different signals, this method was considered to be of less interest.

7.2 Mapping the modulation index

Mapping the modulation index allows the user to access additional information about the user-made or pre-clustered (based on k-means clustering method) ROI groups.

The average over the whole group processed with the fluorescence value or its rate of change provides information about the mean fluorescence in the group, or the rate of change of the mean fluorescence. These mappings were found to enable the user to monitor group activation : the sound gets brighter when the group is highly activated, or when the mean value increases. However, using the rate of change of the fluorescence was perceived to be less comprehensible when using user-made groups compared with pre-clustered groups. In the case of user-made groups the less homogeneous activity would produce an almost constant brightness harder to interpret. This configuration was found to be less sensitive to the data, and required less modification from one set to another. We chose the average fluorescence for the default preset.

On the other hand, the activation ratio and its rate of change provided information about whether or not a large portion of the group is active, when fluorescence is chosen as the value, or becoming active, when fluorescence rate of change is chosen. We mainly used the fluorescence value, as the rate of change processed with both processing tended to produce an almost constant brightness harder to interpret.

The activation ratio was used to provide information about the synchronicity of the groups. If the ROIs are synchronised and become activated at the same time, a very bright sound is produced, with great variations. On the contrary, if only a few ROIs are activated randomly, a more constant brightness is produced. Therefore, this configuration was used in preset 3. The activation ratio rate of change used with the fluorescence intensity produces small bursts of bright sound that would occur when the group gets quickly activated. These quick changes were used to perceive the moments when several ROIs in the same group gets active. Moreover, the difference in the sound between synchronous and non-synchronous groups were emphasized by the fact that a synchronous group would produce smaller bursts. This configuration was used in preset 4.

The Standard deviation was considered to be less useful than the others. The result is harder to interpret as the parameter would get lower if all the ROIs are active at the same time, but would increase if only a small number of ROIs are active.

Lastly, the use of the fluorescence rate of change of individual ROIs were used to monitor ROI activity without focusing on group events. This configuration was used in preset 2, along with the plucking-like sounds to emphasize the activation.

From an aesthetic point of view, the settings of these mappings allows us to obtain different sounds such as a constant, fluctuating brightness or small bursts. Attention was given to obtain aesthetically pleasing results. Indeed, the drawback of the mapping is that the result is sensitive to the data and can result in too much or too little brightness. Hence, it is important that the user has control over the scaling process.

7.3 Mapping the amplitude

Mapping the amplitude was highly dependant on the scaling of the fluorescence. Increasing or decreasing the range of rejected value were used to adapt to different data sets, by rejecting static sounds considered to be irrelevant or unpleasant. The slope of the scaling curve can be used to emphasize the transition. Therefore, for presets 1 and 2 as we wanted the information to be mainly conveyed by the amplitude, a rapid transition was used. On the other hand, for presets 3 and 4, we wanted to use the brightness to convey information about the groups. Hence, a smoother curve was used.

The main drawback of this mapping was that as the values used as fluorescence intensity were the mean value of the ROI, the spatial distribution (size of the ROI) had not been taken in account. Hence, some activation were not well represented and could not be perceived depending on the size of the ROI.

7.4 Selected sound frequency

Initially, we distributed the ROI groups on the notes of the C major scale (rather than random frequencies) in order to keep an overall consonance.

The first requirement was to obtain a uniform relationship between perceived loudness and cell activity, regardless of the tone selected for the cell. We thus used isosonic curves [18] to map the activity of the ROI to the amplitude of the audio synthesis.

Another interesting point was the impact of the interval between the selected notes. Different intervals were tested. First of all when using small intervals, up to perfect 5th, consecutive activation were more easily perceived as melody, whereas larger intervals were more likely to be perceived as independent events. This phenomenon of streaming by pitch proximity [19] was used to monitor temporal patterns of activation between the groups.

Moreover, the use of low notes would produce two auditory streams : one with the notes themselves, and one with the high harmonics added by the frequency modulation. This effect of streaming by timbre [19] was used in presets 3 and 4.

The effects of the notes used on the perception should be further studied. In particular, future studies to analyze the relationship between the different notes and mapping used could be of great interest.

8. CONCLUSION

We developed a sonification software using parameter mapping to explore neuronal activity using calcium imaging data. An interactive interface was developed. The goal was to observe synchronicity phenomena between the neurons, as well as temporal and spatial activation patterns.

The mapping to the amplitude enables the recognition of temporal patterns of activation. The quality of the mapping to the modulation index largely depend on the choices of the user. We designed four presets to detect different parameters : general purpose, observation of ROIs activity, monitoring synchronicity within groups of ROIs, and monitoring temporal patterns of activation.

The selection of notes also had an impact on the perception of the sonification. Indeed, the relationship between mapping and notes allows one to obtain various effects.

Finally, the sonification of the visual media appears to enhance and clarify the visualization of neuronal activity. These conclusions should be completed with further studies. In conclusion, we have made an interactive software, which can be used as a tool to further comprehend the diverse and complicated modes of neuronal activity.

9. REFERENCES

- [1] C. Feierstein, R. Portugues, and M. Orger, “Seeing the whole picture: A comprehensive imaging approach to functional mapping of circuits in behaving zebrafish,” *Neuroscience*, vol. 296, pp. 26–38, Jun. 2015.
- [2] A. G. Blankenship and M. B. Feller, “Mechanisms underlying spontaneous patterned activity in developing neural circuits,” *Nature Reviews Neuroscience*, vol. 11, no. 1, pp. 18–29, Jan. 2010.
- [3] T. Hermann, “Taxonomy and definitions for sonification and auditory display,” in *Proceedings of the 14th International Conference on Auditory Display*, Paris, France, 2008, p. 8.
- [4] S. Hasegawa, S. Ishijima, F. Kato, H. Mitake, and M. Sato, “Realtime sonification of the center of gravity for skiing,” in *Proceedings of the 3rd Augmented Human International Conference on - AH '12*. Megève, France: ACM Press, 2012, pp. 1–4.
- [5] P. Cowden and L. Dosiek, “Auditory Displays of Electric Power Grids,” in *Proceedings of the 24th International Conference on Auditory Display - ICAD 2018*. Houghton, Michigan: The International Community for Auditory Display, Jun. 2018, pp. 113–120.
- [6] T. Hermann, A. Hunt, and J. G. Neuhoff, *The Sonification Handbook*, 2011.
- [7] G. Kramer, B. Walker, T. Bonebright, P. Cook, J. H. Flowers, N. Miner, and J. Neuhoff, “Sonification Report: Status of the Field and Research Agenda,” National Science Foundation, Tech. Rep., 1997.
- [8] G. Dubus and R. Bresin, “A Systematic Review of Mapping Strategies for the Sonification of Physical Quantities,” *PLoS ONE*, vol. 8, no. 12, p. e82491, Dec. 2013.
- [9] J. Ferguson and S. A. Brewster, “Evaluation of psychoacoustic sound parameters for sonification,” in *Proceedings of the 19th ACM International Conference on Multimodal Interaction - ICMI 2017*. Glasgow, UK: ACM Press, 2017, pp. 120–127.
- [10] B. N. Walker, “Magnitude estimation of conceptual data dimensions for use in sonification,” *Journal of Experimental Psychology: Applied*, vol. 8, no. 4, pp. 211–221, 2002.
- [11] L. Gionfrida and A. Roginska, “A Novel Sonification Approach to Support the Diagnosis of Alzheimer’s Dementia,” *Frontiers in Neurology*, vol. 8, Dec. 2017.
- [12] R. F. Cadiz and P. Irrarazaval, “Sonification of Medical Images Based on Statistical Descriptors,” in *ICMC 2015*, 2015.
- [13] I. Gomez and R. Ramirez, “A data sonification approach do cognitive state identification,” in *The 17th International Conference on Auditory Display (ICAD-2011)*, Budapest, Hongrie, 2011, p. 5.
- [14] A. D. N. Edwards, A. Hunt, G. Hines, V. Jackson, A. Podvoiskis, and J. Stammers, “Sonification strategies fox examination of biological cells,” in *The 16th International Conference on Auditory Display (ICAD-2010)*, Washington, D.C, USA, 2010, p. 8.
- [15] H. Viswanath, A. Q. Carter, P. R. Baldwin, D. L. Molfese, and R. Salas, “The medial habenula: still neglected,” *Frontiers in Human Neuroscience*, vol. 7, 2014.
- [16] E. R. Duboué, E. Hong, K. C. Eldred, and M. E. Halpern, “Left Habenular Activity Attenuates Fear Responses in Larval Zebrafish,” *Current Biology*, vol. 27, no. 14, pp. 2154–2162.e3, Jul. 2017.
- [17] J. Schindelin, I. Arganda-Carreras, E. Frise, V. Kaynig, M. Longair, T. Pietzsch, S. Preibisch, C. Rueden, S. Saalfeld, B. Schmid, J.-Y. Tinevez, D. J. White, V. Hartenstein, K. Eliceiri, P. Tomancak, and A. Cardona, “Fiji: an open-source platform for biological-image analysis,” *Nature Methods*, vol. 9, no. 7, pp. 676–682, Jul. 2012.
- [18] Y. Suzuki, V. Mellert, U. Richter, P.-T. Bundesanstalt, H. Møller, L. Nielsen, R. Hellman, K. Ashihara, K. Ozawa, and H. Takeshima, “Precise and Full-range Determination of Two-dimensional Equal Loudness Contours,” p. 10, 2003.
- [19] D. Deutsch, “Grouping Mechanism in Music,” in *The Psychology of Music*, 3rd ed. Elsevier, 2013, pp. 190–255.