MINISTRY OF EDUCATION
TECHNICAL UNIVERSITY
OF CLUJ-NAPOCA, ROMANIA

Electronics, Telecommunications and Informational Technologies Engineering

# PhD THESIS
**- SUMMARY -**

# Dynamical mechanisms underlying gamma oscillations in functional and dysfunctional circuits

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**- Cluj-Napoca -**
**2022**

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# 1 Introduction

The greatest endeavor of human intelligence may be to comprehend the brain; there is undeniable value in knowing how our thoughts, actions, and emotions are processed. The brain, which weighs about 1.5 kilograms and has over 100 billion neurons connected to one another nearly 100 trillion times, is the most complicated organ in the human body.

Over the past 60 years, many mysteries about the functioning of the brain have been resolved. For instance, advancements in the fields of microscopy and staining methods revealed the building block of the brain - the neuron, whereas progress in the fields of electrophysiology taught us that neurons communicate with each other *via* electrical and chemical signalling. Moreover, we learned that the brain is composed of two types of neurons, excitatory and inhibitory which can engage in a synchronous "firing dance", giving rise to *brain waves*.

Modern imaging techniques have demonstrated that the brain's architecture has a topological organisation, typically divided into four subsections called *lobes* where each lobe is associated with a specific brain task. For instance, the frontal lobe is associated with planning and problem-solving tasks, the parietal lobe is the situ for language processing, whereas temporal and occipital lobes are associated with memory and visual interpretation, respectively.

One of the most interesting brain topics that spark fervent debates across the scientific community and is also the main topic of this thesis is brain oscillations. Brain oscillations are repetitive patterns of neural activity produced specifically by the central nervous system in both cortical and subcortical areas. Experimental evidence produced in the last two decades of research underline their role in brain communication and computation. In particular, a fast brain oscillation also known as gamma (30-80Hz) has received considerable attention due to its involvement in several cognitive processes such as attention, memory, and information processing. Furthermore, clinical studies have further evidenced the importance of such rhythm as several neurological disorders such as Alzheimer's disease and schizophrenia share a marked impairment of gamma oscillations.

Understanding brain oscillations, their role and how their alteration leads to brain disorders, involves investigating the mechanisms through which these oscillations may emerge. It is now acknowledged that such brain rhythms originate in cortical circuits through a mechanism of push-

pull interaction between the activity of two populations of neurons (i.e., excitatory and inhibitory). In particular, there are two type of cortical networks necessary for the gamma oscillations to emerge: the interneuronal gamma (ING) - a cortical circuit formed by solely interneurons which generates gamma activity under tonic excitation and the pyramidal-interneuron gamma (PING) - a cortical network formed by both excitatory pyramidal cells and inhibitory interneurons where fast excitation provided by pyramidal cells and delayed feedback inhibition provided by inhibitory interneuron alternate, giving rise to oscillatory activity specifically within the gamma band.

This thesis focused on three research objectives. Our first objective consisted of performing visual experiments using different stimuli in the animal model and study oscillatory dynamics within local circuits such as ING and PING. Secondly, we focused on developing and implementing novel time-frequency analysis useful in the quantification of fast oscillatory activity such as the gamma rhythm observed in the recorded neurophysiological data. Herein, we implemented a novel method called superlet-transform (SLT) and contributed in the development of its derivates such as the Fractional superlets (FSLT). Furthermore, we developed a novel method for spike detection using a machine learning approach and showed that our method has better performance compared to the classical threshold-based method. Finally, we focused on the etiology of brain disorders that share a marked impairment of gamma oscillations such as schizophrenia and Alzheimer's disease, performed behavioral experiments in the animal model in order to find human phenotypes of the disorders, and proposed novel treatments that could improve the symptomatology.

The work for this thesis required a multidisciplinary approach, integrating biological and medical fields with more technical ones like engineering, computer science, and signal processing. In the paragraphs that follow, we will provide a summary of this work.

# 2 Methods

There are several methods discussed in this thesis that can be conceptually split into two categories: experimental methods used to record the brain's electrical activity and data analysis methods used for decoding the recorded extracellular brain signals. The next sections, will summa-

rize each category, with emphasis on the second category as it comprises novel methods introduced by the thesis itself.

## 2.1 Recording the brain's activity

The first part of the thesis's method section discusses experimental techniques used for recording the brain's electrical activity. In particular, the centrepiece of this section consists in describing *in vivo* electrophysiological recordings, which have been widely performed by the author. The advantage of such technique is that allows brain scientists to investigate the dynamics of neural circuits and correlate its findings with animal behaviour. All experimental studies performed in this thesis were conducted on animal subjects, in particular mice of the species Mus Musculus, and followed strict ethical guidelines established by the local and the European Union legislation in the matter of animal welfare.

A typical *in vivo* electrophysiological experiment starts with getting access to the animal's brain by performing a stereotaxic surgery conducted in a sterile environment. The animal is first anaesthetized, and subsequently placed on a stereotaxic instrument which keeps the animal's head in a fixed position and allows for a correct identification of the targeted area that we want to record from. Access to the brain is achieved by performing a craniotomy - an invasive surgical procedure which consists in removing a small part of the scalp and subsequently the bone with the help of a scalpel and a dental drill, respectively.

Once the animal's brain is exposed, recording devices such as silicon probes are first, mounted on the stereotaxic's manipulator and secondly, slowly lowered into the brain. This procedure can take up to 1 hour, as minimal damage to the brain following the insertion must be ensured. Depending on the scientific question, different silicon probes can be used. For instance, if one is interested in an animal behaviour during an exploration task that must be recorded for multiple days, *chronic* silicon probes are preferred. As the experimental procedures performed in this thesis were the type of non-recovery (i.e., the animal does not recover after the experiment) silicon probes of the type *acute* were used. That is, the probe is inserted for a few hours, and then recovered, cleaned and stored for reuse. To minimize animal use, multiple recordings are collected from each animal, over a period of 6-8 hours.

Once inserted, the silicon probe records two types of the brain's extracellular signature: spikes - fluctuations of the membrane potentials of

the neighbouring neurons and local field potentials - activity of synchronized neurons nearby of the recording site. The recorded signal is initially of low amplitude, thus requiring further amplification. Once the signal is amplified, it is further digitized using an analog-to-digital converter (ADC) and ready to be pre-processed and analysed. Depending on the experimental demands, the signal can be pre-processed into two types: local field potentials - low frequency part of the recorded signal consisting in extracellular potentials with frequencies ranging from 0.5-300Hz and multi-unit activity - higher frequency part of the recorded signal consisting of neuronal spiking activity with frequencies above 500Hz.

## 2.2 Decoding extracellular signals

Advancements in engineering allows scientists to record large amounts of electrophysiological data through large electrode arrays that have hundreds of channels at a high sampling rate (>32kHz). However, as much as pre-processing the electrophysiological data is generally a simple procedure, analysing such data can be a more complex endeavour. In the next sections, we will summarize all the methods used and developed in this thesis to analyse both local field potentials and multi-unit activity.

### Local field potentials

Local field potentials are usually obtained by filtering the raw electrophysiological signal with a digital band-pass filter with a cut-off frequency between 0.5-300Hz. Once the signal is filtered and downsampled (i.e., for computational efficiency) time-frequency analysis can be be performed in order to characterize the dynamics of brain oscillations. Popular investigative methods used to extract time-frequency information from LFPs consist in performing a short-time Fourier transform (STFT) or a Continuous Wavelet Transform (CWT). However, both methods present some short comings regarding the precise estimation in time and frequency (i.e., they are not Pareto optimal). This limitation is caused by the Heisenberg-Gabor uncertainty principle which states that is impossible to have a simultaneously precise estimation of a signal in both, time and frequency. That is, the precise estimation of a signal in time requires trading off precision in frequency, and vice-versa. To overcome this limitation, an improved method of CWT called Superlets (SLT) has been developed.

### 2.2.1 Superlet transform

Briefly, the Superlet transform (SLT) combines geometrically the power values of a set of Morlet wavelets with different number of cycles. Parameters of the SLT include base cycles - *c*, the number of of cycles of the shortest Morlet wavelet and the order - *o*, the number of wavelets in the set. SLT outperfoms STFT and CWT, especially when estimating fast oscillation packets present in electrophysiological data. An example of its outstanding performance is shown in Figure 1. Detection of oscillation packets present in human electroencephalography (EEG) data are clearly more visible with SLT compared to STFT, CWT, and MMCE.

$$SLT_{x,c_1,o_f}(f,t) = \left[\prod_{i=1}^{o_f} P_x(c_1 i, f, t)\right]^{\frac{1}{o}} \tag{1}$$

where

- $x$ is the input signal $x(t)$,
- $c_1$ is the number of cycles of the shortest wavelet,
- $o$ is the order of the superlet at frequency $f$,
- $P_x(c, f, t)$ is the power ($2|A|^2$) of the response $R_x$, i.e. the convolution of the signal with the wavelet.

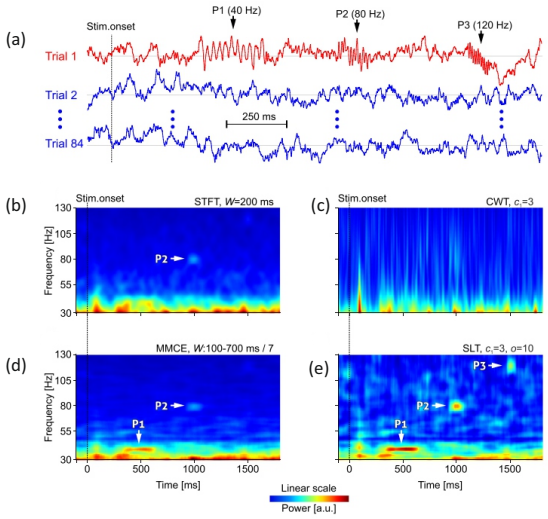


**Figure 1: Detection of fast oscillation packets with different time-frequency methods.** (a) Three oscillation packets with frequencies at 40,90, and 120Hz were inserted in a single trial in a human EEG recording. (b),STFT (c) CWT, (d) MMCE, (e) SLT. The fast oscillation packets are clearly more visible in the SLT scalogram. Taken from Moca *et al.* 2021

### 2.2.2 Fractional superlets

As mentioned in the previous section, the order of the superlet is the number of the wavelets in the set. Originally, this number was defined as an integer number which is adapted in discrete steps depending on the frequency of interest. That is, low frequencies require a low order while high frequencies, higher orders. This adaptiveness of the superlets is also known as Adaptive SLT (ASLT). When ASLT was deployed on electrophysiological data, the scalogram was affected by banding. To solve this issue we introduced a novel method called fractional superlet (FSLT) which allows for the order to assume fractional numbers. FSLT solves the banding probelm, providing a scalogram which is smoother compared to the one performed with the traditional ASLT.

$$FSLT_{x,c_1,o_f}(f,t) = \left[ P_x(c_1(o_i+1),f,t)^{\epsilon} \prod_{i=1}^{o_i} P_x(c_1 i,f,t) \right]^{\frac{1}{o_f}} \qquad (2)$$

where,

- $o_i$ is the integer part of the order

- $\epsilon$ is the fractional part.

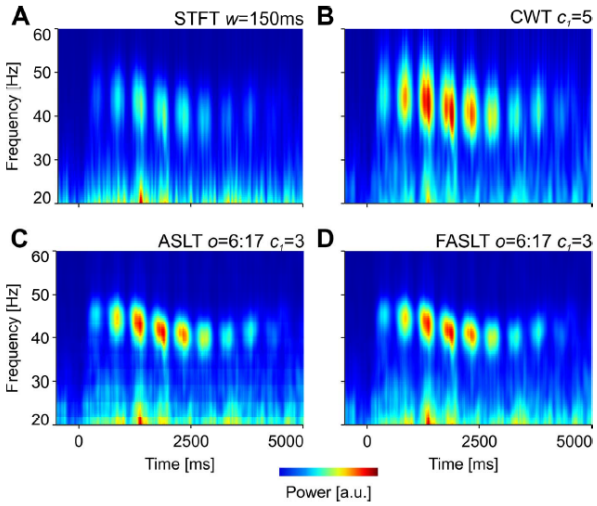


**Figure 2: Comparison of four time-frequency representations (TFR) on electrophysiology data.** The banding problem is clearly visible in ASLT (c). (d) FASLT offers the smoothest TFR. Taken from Bârzan *et al.* 2021

### 2.2.3 Content evaluation of TFR using machine learning

As previously mentioned, oscillatory processes in the brain can be investigated using a series of time-frequency methods such as STFT, CWT, SLT, and Choi-Williams distribution. However, assessing which method offers a better time-frequency representation is not an easy task. In this study, we introduce an empirical method based on machine learning which allows for the identification of the ”best” TFR based on how much information they ”carry” about the experimental conditions. The input data used in this study consisted in human EEG data recorded during a visual recognition task and LFPs recorded from the mouse visual cortex during a visual stimulation task. To ascertain which TFR performed better, maximum accuracy and learning curves were measured.

In addition, we wanted to investigate which spectral features of the TFRs were more relevant for the classification. This was done by using a feature perturbation method, called *joined feature permutation*. Briefly, the method consists of perturbing a set of features that are strongly correlated in the input layer of the artificial neural network (ANN) and doing a post-evaluation of the performance using two metrics: accuracy of

classification and mean-squared error. This method allows for the identification of both important features for the classification and also those that act as distractors in the data (i.e., noise).

## 2.3 Multi-unit activity

Neuronal spiking activity is usually obtained by filtering the raw electrophysiology signal with a band-pass filter with cut-off frequencies at 300Hz and 7kHz. Once the signal is properly filtered, it is visually inspected in search of spikes. Traditional spike detection is done by setting an amplitude threshold. That is, each time the signal amplitude crosses a certain threshold, the spike is extracted and used for further analysis, such as *spike sorting*.

### 2.3.1 MLSpikeDetector

As mentioned previously, spikes are detected using an amplitude threshold that can be set either manually or automatically and is generally based on the signal's standard deviation or interquartile interval. An automatic threshold is preferred when analysing neural recordings with tens of channels. As much as this method is straight and easy to implement, it has some drawbacks. For instance, hard thresholds tend to neglect spikes just below the threshold, resulting in an amplitude distribution of the spikes that is cropped unnaturally. To tackle this problem, we build a novel spike detection algorithm using a machine learning approach which outperforms the classical threshold based method. Our method can be summarized as follows. First, an automatic threshold based on the signal's standard deviation is set and spikes are detected. Secondly, a MLP classifier with two output nodes *(spike and non-spike)* is trained on the spikes detected in the first step. Thirdly, the trained classifier is slid along the recorded data, sample by sample, and the points for which the classifier's spike class output probability is high are recognized as spikes and saved in a probability signal. Finally, the resulting probability signal is thresholded and spikes are extracted.

### 2.3.2 Measuring neuronal interactions using curve fitting

Following the spike detection process, spikes can be regarded as time series of discrete events (i.e., spike trains) and can be investigated using

time-series analysis methods. In particular, one can extract a spike train for each putative neuron and make inference regarding neuronal interactions and synaptic connectivity within the recorded data. This is done by performing a cross-correlation analysis, where features such as peaks, troughs, and satellite peaks of the computed cross-correlation histogram (CCH) reflect neural connectivity. For instance, a CCH with a peak at zero delay is thought to represent a correlated neuronal firing. Quantification of CCH features is achieved using different methods. However, these methods present some limitations such as *a priori* assumption of the data. Furthemore, they work best with rich spike trains failing when this is not the case (e.g., neurons with low firing rates). Another fact that must be considered is that neuronal interactions can be of a sub-millisecond scale. Thus, performing a CCH with a bin size of millisecond order can be pointless and it may lead to misinterpretation of the data.

More reliable methods for the measurement of neuronal interactions have been proposed. For instance, fitting a Gabor function to a computed CCH provides reasonably good results. In this study, we fitted a Gabor function to a large dataset of computed CCHs of real experimental data using different optimisation algorithms such as Trust Region (TR) and Levenberg-Marquardt (LM) and assessed the goodness of fit for each fitting problem using $\chi^2$.

# 3 Neural oscillations in brain disorders

Neuronal oscillations appear to be dysfunctional in conditions of the brain such schizophrenia and Alzheimer's disease, according to experimental research using both animal and human models. A notable weakening of gamma oscillations appears to be a common feature of both brain disorders. In this thesis, these brain disorders are reviewed, and alternative treatment approaches that have been tested in our research institute are proposed. Each disorder will be briefly discussed in the following two parts, and section 4 will show the findings of our research.

## 3.1 Schizophrenia

Approximately 1% of people worldwide suffer from schizophrenia, a disorder of self and reality. Although the exact cause of the condition is unknown, genetic, environmental, and neurochemical variables are believed

to play a role. Symptoms of schizophrenia include positive (i.e., hallucinations, paranoid delusions), negative (i.e., social withdrawal, loss of motivation), and cognitive (i.e., impaired volition and memory).

Clinical research on schizophrenia patients has shown a dysfunctional neural circuit as a result of disruptions in neurotransmission, structural changes in neuronal projections, and alteration of axonal integrity. Numerous investigations have also discovered aberrant oscillatory activity and poor synchronization, especially in the gamma rhythm. Additionally, aberrant gamma oscillations appear to have clinical correlates (i.e., hallucinations, psychomotor poverty). A more detailed description of these findings is presented in the thesis.

There has been a consensus among researchers over the past 20 years that schizophrenia may also have its roots in N-methyl-D-aspartate (NMDAR) hypofunction. The fact that NMDA antagonists, such as ketamine, cause schizophrenia-like symptoms when administered, has increased the appeal of this viewpoint. NMDARs are glutamate receptors that are important for a number of neurobiological functions of the brain, including neuronal plasticity, learning, and memory. Activation of NMDAR requires glutamate as a principal neurotransmitter and an agonist, which consists of either L-glycine or D-serine.

The mechanism through which NMDAR hypofunction might cause schizophrenia is the following. Dysfunctional NMDARs may lead to a altered excitatory transmission within the GABAergic system, which in turn leads to disrupted gamma oscillations. As a reminder, the GABAergic system is formed by inhibitory interneurons, which are crucial in the emergence of gamma oscillations (see ING or PING mechanisms described in the introduction). Thus, if the inhibitory population does not have sufficient tonic excitation, rhytmicity, particularly within the gamma band, is lost.

In this study, we wanted to explore the NMDAR hypofunction hypothesis by performing *in vivo* and behavioral experiments in the schizophrenia mouse model and test if NMDAR function can be restored using modulatory agonists such as D-serine. D-serine is an important co-agonist of NMDAR, and recent evidence has shown that levels of D-serine are altered in schizophrenic patients. Result of both experimental and behavioral experiments are presented and discussed in section 4.

## 3.2 Multi-sensory gamma therapy

Gamma abnormalities have also been identified in Alzheimer's disease patients, as was discussed in the previous section. Additionally, research in the AD mouse model has shown that aberrant gamma activity is present even before the start of symptoms, indicating their potential as an early biomarker.

Memory loss, personality disturbances, and irritability are signs of AD. Although the exact origins of this condition are unknown, it is believed that a combination of genetic, dietary, and environmental factors contribute to the disorder. Patients affected by AD present an unusual build-up of proteins in various regions of the brain also known as beta-amyloid plaques (A$\beta$) which have been associated with neurological deterioration.

In order to cure AD symptoms, a new therapy approach specifically targets gamma oscillations. Particularly, a novel therapeutic strategy known as *gamma entrainment using sensory stimulation* (GENUS) has found that subjecting an AD mouse model to light flickering stimulation at 40 Hz for an hour each day for a week increased the phagocytic activity of *microglia* which in turn decreased levels of A$\beta$ and tau proteins — the pathological markers of AD. Additionally, mice displayed improved performance in spatial learning and memory tasks when the light flickering stimulation was coupled with a sound stimulation at 40Hz.

In this study, we re-engineered and used GENUS therapy and used cutting-edge time-frequency analysis, including the superlet transform (SLT), to measure gamma entrainment in the animal model. Additionally, we experimented with various light stimulation wavelengths to gauge their impact on cortical entrainment. Section 4 of our summary discusses our findings.

# 4 Results and discussion

The outcomes of this thesis span a variety of disciplines, including biology, medicine, and signal processing. These findings have been presented at conferences and published in several journals.

## 4.1 MLSpikeDetector

We first carried out some *in vivo* experiments and recorded from the mouse visual cortex during a visual stimulation task to test our approach for

spike detection utilizing machine learning. Our findings suggest that the spike detection achieved by our method is on par with the classical method- it detects >95% of spikes. Moreover, our method has a novelty rate of over 15%, which indicates that at least 15% of all spikes are overlooked with the traditional threshold-based method. As a result, as seen in Figure 3, the spike amplitude distribution is no longer chopped, but it decays naturally.

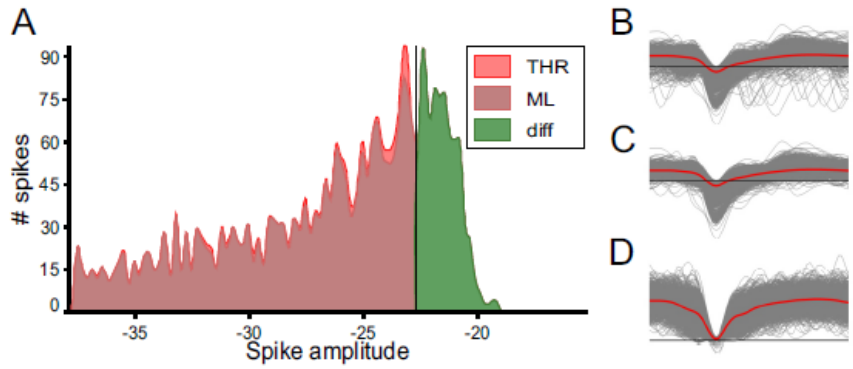


**Figure 3: Amplitude distribution of detected spikes.**(a) A comparison of MLSpikeDetector (dark red) with the conventional threshold-based technique (bright red); The green area displays the number of spikes found exclusively by MLSpikeDetector; (b) threshold detected spikes; (c) MLSpikeDetector spikes; and (d) the difference between (c) and (d). Taken from Bârzan & Ichim. 2020.

## 4.2 Measuring neuronal interactions with curve fitting

In this study, we were interested in the temporal dynamics between two populations of neurons - inhibitory and excitatory - at both millisecond and sub-millisecond resolution. Thus, a huge dataset of computed CCHs from real experimental data was fitted with a Gabor function using several optimisation algorithms, including Trust Region (TR) and Levenberg-Marquardt (LM), and the quality of fit for each fitting problem was evaluated using $\chi^2$. The following is a summary of our findings. Our fitting parameters included both real and imaginary parts when we employed the Levenberg-Marquardt (LM) algorithm to solve our fitting problem, which should not have happened because our Gabor function should be solved in the real domain. Thus, we came to the conclusion that the fitting problem

needed to handle some bound restrictions in order to be solved in a real domain. In our situation, the Trust Region (TR) optimisation algorithm was used to do this. When comparing the two algorithms, TR performed better and needed less iterations and function evaluations. Furthermore, showed increased robustness to initial parameters.

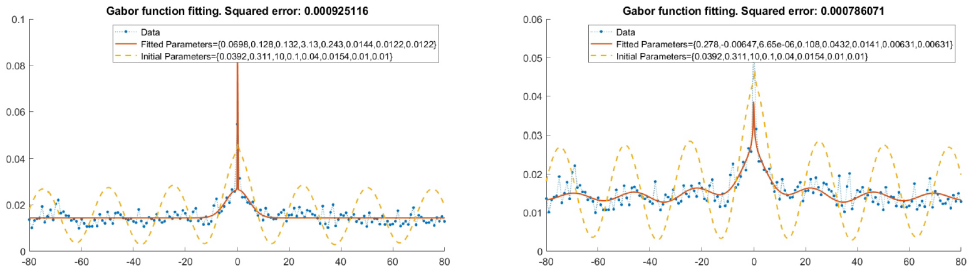


**Figure 4: An example of Gabor fitting on cross-correlation histogram.** (left) Curve fitting using LM. (right) Curve fitting using TR. Taken from Ichim *et al.* 2019

## 4.3 Content evaluation of TFR using machine learning

With this study, we introduce a machine learning-based empirical method that enables the identification of the ”best” TFR based on the amount of data they ”carry” regarding the experimental conditions. One of the input data sources used for the evaluation consisted in human EEG data collected while performing a visual recognition task. We measured maximum accuracy and learning curves to determine which TFR worked best. We found that the superlet transform (SLT) outperforms the other methods - it reaches saturation at 70% within 400 epochs; by comparison, ChoiW, its closest rival, only catches up to it around epoch 900. Result are shown in Figure 5.

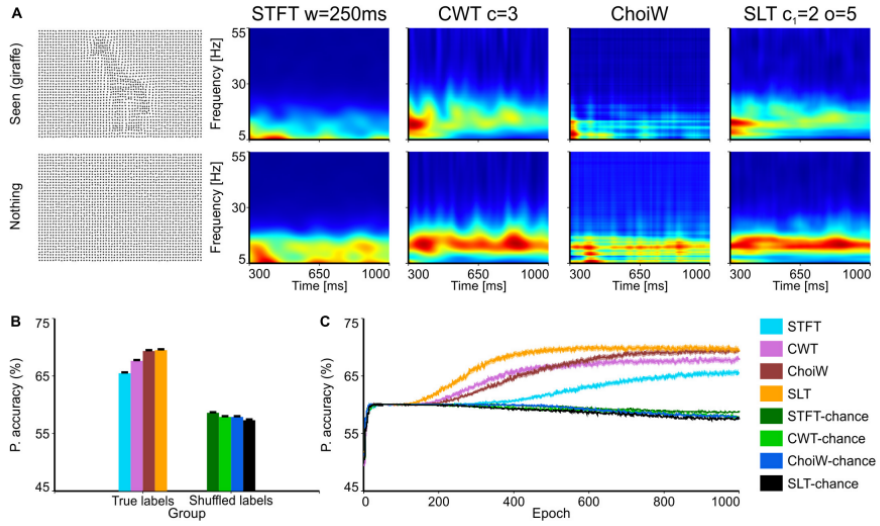


**Figure 5: Classification performance of four TFRs on human EEG data** (a) EEG experimental conditions - seen (top), nothing (bottom); (b) Classification accuracy of the four TFRs (left) and of the shuffled labels (control); (c) Learning curves of validation sets in (b). Taken from Bârzan *et al.* 2022.

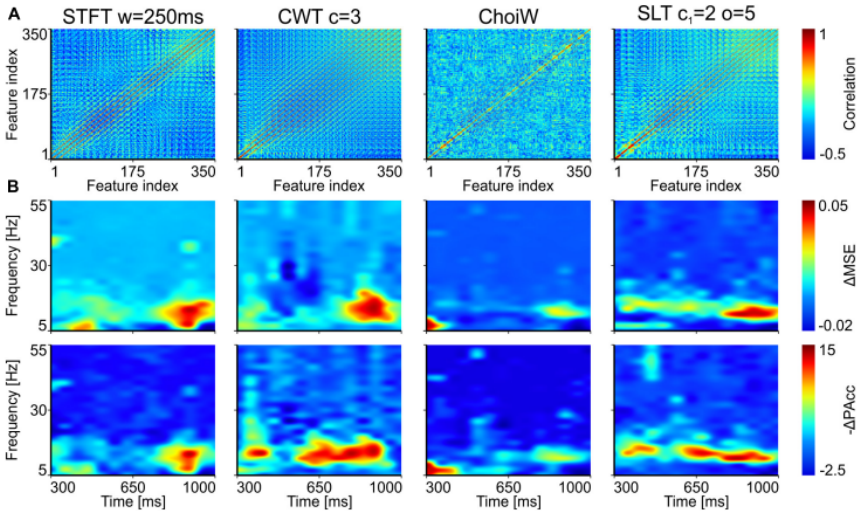


**Figure 6: Feature perturbation results in TFRs.** (a) Correlation matrix for the features extracted from each TFR method. (b) Feature importance metrics - bottom ($\delta$PAcc), difference in mean squared error ($\delta$MSE) (top). Taken from Bârzan *et al.* 2022.

This study also provides a brand-new technique called *joint feature permutation*, which makes it possible to determine which features of the

TFRs are most crucial for classification. Figure 6 displays the findings of this investigation.

## 4.4 Behavioural assessments of schizophrenia symptomatology in the murine model

Our work sought to identify behavioural abnormalities in mice that are indicative of the negative-cognitive symptoms of schizophrenia and to investigate whether modulatory NMDAR agonists, such as D-serine, may be used to restore NMDAR function. In order to treat the negative and cognitive symptoms of schizophrenia, for which traditional treatments have failed, restoring NMDAR signaling through the administration of D-serine may pave the way for innovative therapeutic agents. The groups of mice participating in this study were NR2DKO-2D-E knock out (i.e., homozygous mice that had mutation of the gene - NMDA hypofunction) and NR2DKO-2D-E wild type (i.e., homozygous mice that had no mutation - control group).

### Tail suspension test

Negative-like symptoms in schizophrenia patients appear as a lack of drive, social withdrawal, and anhedonia, as was discussed in the preceding section. Identifying the same symptoms in the murine model might be quite difficult. To do this, we used the behavioural despair model known as the Tail Suspension Test, which is one of the most popular paradigms for the evaluation of depression symptomatology in mice (TST). The following justifies the test's use. Mice who are subjected to unavoidable short-term stress (such as tail suspension) gradually lose hope of escaping and adopt an immobile stance (also known as TST immobility, which is comparable to a depressive symptom in humans - negative symptoms of schizophrenia. We found that knock out mice presented higher immobility scores when compared to the wild type ones suggesting a passive behaviour which is comparable to the lack of motivation observed in schizophrenia patients.

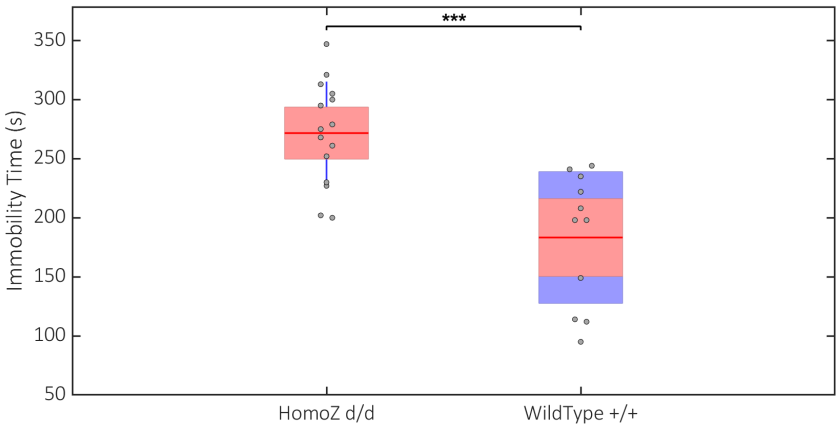


**Figure 7: Tail Suspension Test results.** Subjects in the knock out group presented continuous periods of immobility suggesting a phenotype.

## Novel object recognition test

Working memory deficits are one of the main cognitive symptoms of schizophrenia. We used the Novel Object Recognition Test to see if the experimental group (knock-out) had memory impairments (NORT) and if these memory impairments could be ameliorated by administration of D-serine.

NORT relies on the rodent's natural propensity for exploration. Generally, rodents frequently engage with unfamiliar objects more than they do with well-known ones. Therefore, NORT evaluates the animal's capacity to identify a new object in a comfortable setting. Three phases make up NORT: habituation, familiarization, and testing, which are carried out over the course of three days. Behavoral animal tracking during the three phases of NORT was achieved by implementing DeepLabCut which is an open source software based on transfer learning with deep neural networks.

We used NORT to evaluate memory deficits both before and after D-serine administration (1.2 mg/500 mL) in the water *ad libitum*. We used a crossover study as the basis of our investigation, in which mice in each group first received water and then D-serine. Our results indicate that D-serine administration does not produced any memory improvements ad seen in Figure 8. This result could be justified by the fact that D-serine administration was probably administered too late in the experiment (due to SARS-Cov-2 pandemic).

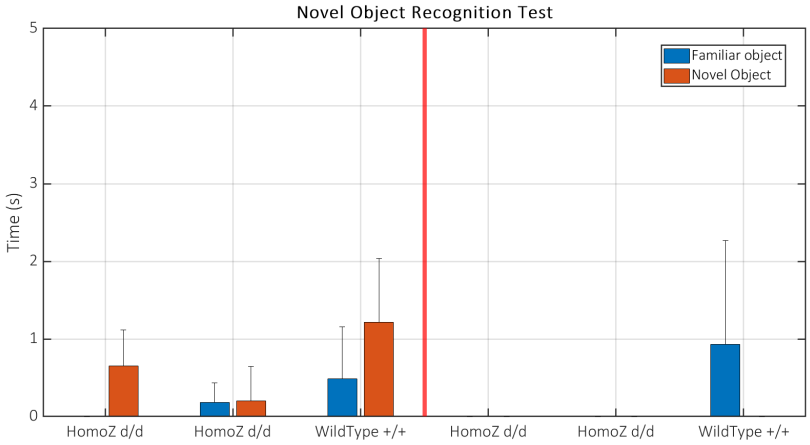


**Figure 8: Novel object recognition test results.** Time (in seconds) spent by knock out and control group with novel and familiar object. The line in red separates the experimental timeline in two phases: (left) - NORT before D-serine administration. (right) NORT after D-serine administration.

## 4.5 Extracellular modulation of NMDARs by D-serine

Different concentrations of D-serine were supplied intracranially during this part of the experiment, and *in vivo* electrophysiological experiments were conducted in the two experimental groups (knock out and wild type). A control condition, in which artificial cerebrospinal fluid (aCSF) was given as opposed to D-serine, was added to the experimental design to see whether D-serine had any impact at a circuit level. The intracranial administration of aCSF and D-serine was therefore alternated while datasets were being recorded.

In Figure 9, two animals from the two groups — wild type (i.e., non-mutant mice - top scalograms) and knock out (i.e., mutant mice - bottom scalograms) are used to compare the aCSF and D-serine conditions. Scalograms were produced using the superlet transform (SLT). The left and right scalograms, respectively, demonstrate the effects of aCSF versus Dserine treatment.

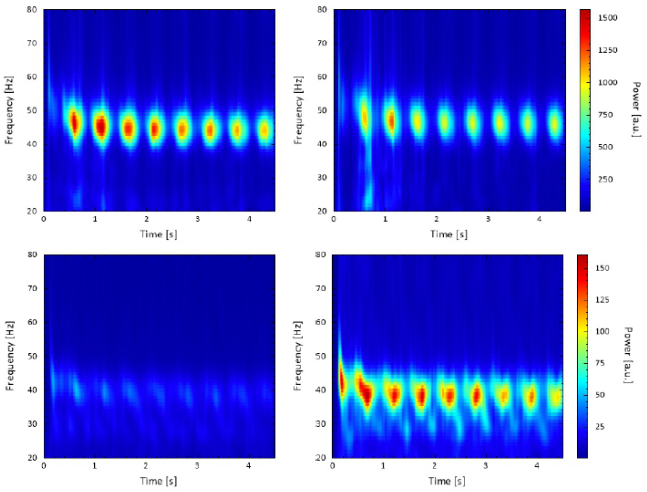


**Figure 9: Gamma power changes in aCSF versus D-serine condition.** Gamma power is clearly stronger when D-serine was administered in the knock out mouse.

Additionally, we sought to quantify D-serine neuromodulatory impact at a circuit level and its contribution to gamma oscillogenesis. We found out that intracranial administration of D-serine increased the gamma oscillatory power in both groups. However, the knock out group appears to be more affected by D-serine than the wild type group, where only marginal effects were seen. These findings suggest that D-serine restored NMDAR activity in the knock out group leading to a robust neural synchronization within the gamma band.

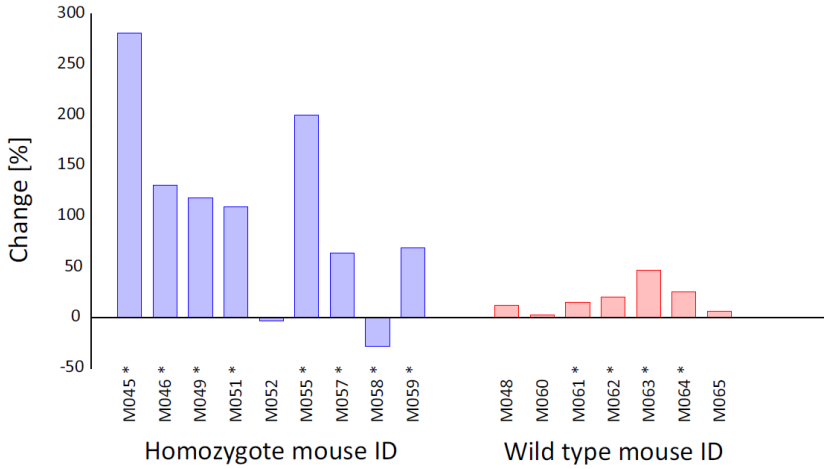


**Figure 10: Gamma power changes after intracranial D-serine administration, specified as percentual change in the gamma band power.**

## 4.6 Multi-sensory gamma therapy

We used GENUS treatment to achieve gamma entrainment and then looked into how different wavelengths of light stimulation might affect the latter. Our hypothesis is that different light wavelengths used in the flicker stimulation could either increase or decrease the cortical neuronal entrainment. Thus, we build a second stimulation panel and compared the effects of blue (460nm) versus white flicker in the primary visual cortex of three anaesthetized adult mice, designated M081, M082, and M083. The same animal provided many recordings, which were interspersed (blue-white-blue-white). A 50% duty cycle flicker stimulus was presented monocularly for 6 seconds at various frequencies (7, 10, 20, 30, 40, 50, and 60Hz). To obtain the same brightness (i.e., 280 lux) for both colours, we varied the driver voltage of each led panel. We found out that the magnitude of gamma entrainment at 40Hz was significantly higher in the blue condition across three animals. Our results are shown in Figure 10 and indicate that GENUS therapy could be improved by the use of the blue flicker instead of the traditional white one.

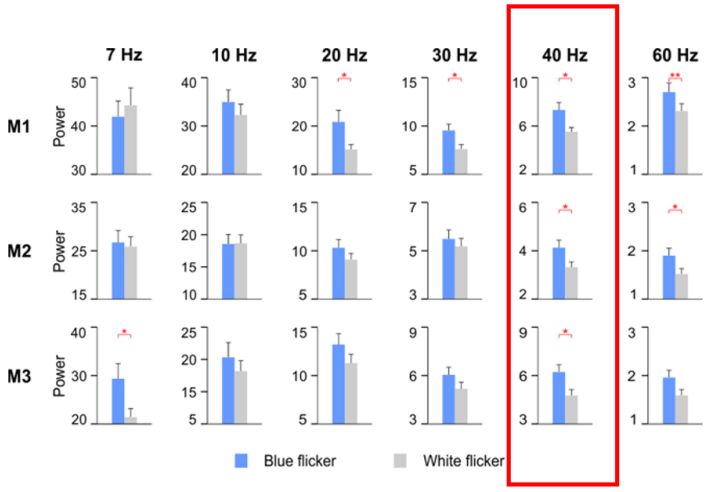


**Figure 11: Blue vs. white light responses during flicker stimulation in three animal subjects.** Taken from Ichim *et al.* 2022

# 5 Conclusion

This thesis introduces several methods useful in investigating brain dynamics and in particular fast cortical rhythms such as gamma oscillations. It does so with a collection of findings that have potential therapeutic implications in brain disorders including schizophrenia and Alzheimer's disease. Additionally, it introduces novel time-frequency techniques such as Superlet transform (SLT) and Fractional superlets (FSLT) that come in handy when investigating oscillatory activity in the brain, especially high-frequency oscillations such as gamma.

Our work reflects the highly multidisciplinary nature of neuroscience and shows how gamma oscillations may be exploited effectively in a variety of therapeutic and practical applications.