ISSN: 2755-0192

Journal of Psychiatry Research Reviews & Reports

SCIENTIFIC Research and Community

Review Article

Open Access

The Multiple-Unit Firing Activity of Hippocampal CA1 Neurons Reflects Recent Prior Experience

Haotian Li[1], Takashi Kuremoto[1]*, Junko Ishikawa[2], Shingo Mabu[3] and Dai Mitsushima[2*]

[1]Nippon Institute of Technology, Japan

[2]Graduate School of Medicine, Yamaguchi University, Japan

[3]Graduate School of Sciences and Technology for innovation, Yamaguchi University, Japan

Abstract

The hippocampus plays an important role in the formation of episodic memory. To identify patterns of hippocampal firing activity specific to episodic memory, we performed Multiple-Unit Firing Activity (MUA) recognition using deep learning methods. Briefly, adult male rats habituated to their home cage experienced one of four experimental episodic stimuli (restraint stress, contact with a female rat, contact with a male rat, or contact with a novel object) for 10 minutes. The patterns of recorded brain spike signals (300–10 kHz) in hippocampal CA1 were classified using machine learning methods such as Convolutional Neural Networks (CNNs), Support Vector Machines (SVMs), deep learning model VGG16, and combination models composed of VGG16 with SVM or VGG19 with SVM. As a result, the model of VGG19 with SVM detected MUA with ripple-like wave firings corresponding to specific episodes, achieving a validation accuracy of 96.79% which was the highest recognition rate in all of deep learning models. The results suggest that MUA of CA1 containing ripple firings corresponds to specific episodic memories. By capturing ripple firings, MUA analysis can assess and diagnose memory function, which may help detect various cognitive disorders.

***Corresponding author's**
Takashi Kuremoto, Nippon Institute of Technology, Japan.

Dai Mitsushima, Graduate School of Medicine, Yamaguchi University, Japan

Received: April 01, 2025; **Accepted:** April 04, 2025; **Published:** April 15, 2025

Keywords: Hippocampus, Deep Learning, Episodic Memory, Ripple-Like Wave Firing, Multiple-Unit Firing Activity (MUA)

Introduction

The hippocampus is part of the limbic system and plays a critical role in the processing of spatiotemporal information, and the formation of short-term and long-term memory [1,2]. Recently, the spontaneous firing activity of hippocampal CA1 neurons and synaptic plasticity have been actively investigated [3,4]. Buzsáki showed that Sharp Wave Ripples (SPW-Rs), which can be observed in the mammalian brain, occur during the "offline" state of the brain under the influence of various neurotransmitters and neuromodulators [3]. Joo and Frank pointed out that these SWRs are associated with memory consolidation processes, decision making, planning, recall, and imagination [5]. Ishikawa et al. recorded Multiple-Unit Activity (MUA) in the Hippocampal CA1 region using deep electrodes in adult male rats habituated to their home cages [6]. They successfully captured thousands of ripple firings of MUA, which are spontaneous, short-term synchronized firing activities. Additionally, the rats were exposed to one of four experiences for 10 minutes: restraint stress (**restraint**), contact with a female rat (**female**), contact with a male rat (**male**), or observation of a novel object (**object**). Statistical pattern recognition was then performed using features such as amplitude, duration, arc length, and peak number of ripple firings (300–10k Hz) specific to each experience [6]. Ishihara et al. investigated the classification and characteristics of specific ripple firings related to episodic memory using a Convolutional Neural Network (CNN) and the Grad-CAM feature analysis method [7,8-11]. In the authors' previous research, various deep learning models, including CNN, CNN + SVM, VGG16, VGG16 + SVM, and ResNet50, were applied to the same dataset to examine the classification accuracy of each ripple firing pattern [12,13]. Among these methods, the proposed VGG16 + SVM approach achieved the highest classification accuracy [14-16]. Specifically, the average classification accuracies for the five types of MUA signal data, including signals with no ripples (**nothing**), were 86.63%, 87.40%, 92.80%, and 95.60% for each deep learning model, respectively [16]. In contrast, the MUA data used in previous studies were limited to the right brain of a single rat (C14R).

In this study, to elucidate MUA patterns related to episodic memory in the left and right cerebral hemispheres of different rats (rat C14 and rat C15), we applied not only the previous machine learning models—VGG16, VGG16 + SVM, ResNet50, and ResNet50 + SVM—but also VGG19 and VGG19 + SVM. The results of the ripple pattern recognition experiment showed that the average classification accuracy of VGG19 + SVM was the highest. The classification accuracies of the VGG19 + SVM model investigated in this study were 95.56% (C14 left brain), 96.79% (C14 right brain), 96.30% (C15 left brain), and 95.56% (C15 right brain), respectively. Among them, the classification accuracy for C14 right brain signals using VGG19 + SVM was the highest at 96.79%. In addition, there was no significant difference in the average classification rates of specific ripple firings between the left and right brains of C14 and C15 for 5-type MUA signals, however, it was found that the pattern classification rates of the left and right hippocampi differed depending on the experiences. Specifically, for the **female** and **restraint** conditions, firing in the right hippocampus (C14R, C15R) was relatively prominent for both rats. In contrast, for the **male** condition, firing in the left hippocampus (C14L, C15L) was relatively prominent. For the **object** condition, there was a significant difference in activity between the two rats. While the right hippocampal data of rat C15 (C15R) showed prominent firing patterns, rat C14 showed relatively low detection rates of specific ripple firings.

In Vivo MUA Recording

Ishikawa et al. measured the spike activity of hippocampal CA1 neuron groups in freely moving adult male SD rats habituated to their home cage using deep electrodes [6]. Figure 1 shows the rat during the measurement experiment (left) and the movable electrode inserted into the hippocampal CA1 region (right). The sampling rate of the in vivo MUA signal recording was 25 kHz, and signals in the range of 300–10 kHz were used for analysis after band filtering. The male rats to be recorded were habituated to their home cage. The schedule of MUA signal measurement is shown in Figure 2. First, hippocampal MUA was recorded for 15 minutes in the home cage. The rats were then exposed to one of the following experiences for 10 minutes: restraint stress (**restraint**), contact with a male rat (**male**), contact with a female rat (**female**), or contact with a novel object (**object**). Table 1 summarizes the total of five patterns, including the four experience patterns and MUA recordings without ripple firings. In addition, examples of 1-second MUA recordings (each consisting of a time series of 25,000 data length) are shown in Figure 3 (a- e).

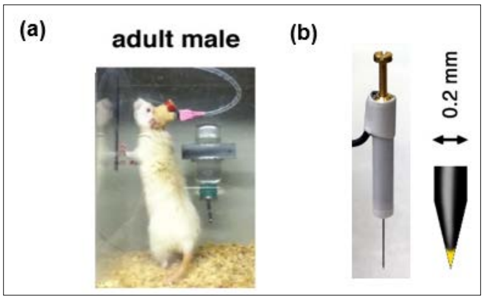


Figure 1: In vivo recording of MUA of a Male Rat [6] (a) Electrode-Implanted Freely Moving Male Rat (b) A Movable Electrode for Recording

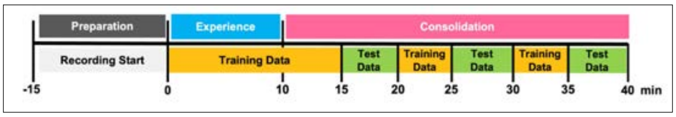
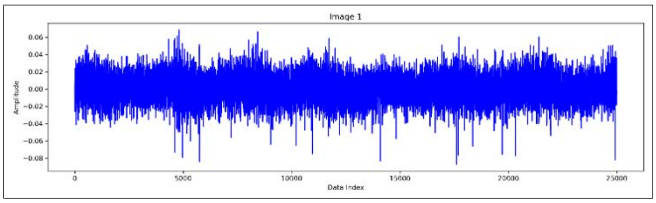
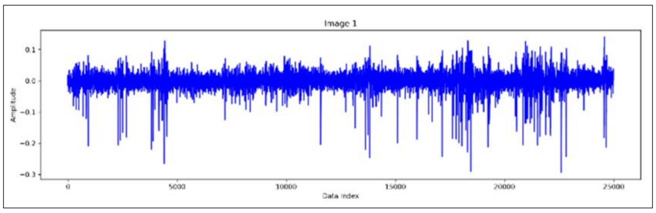


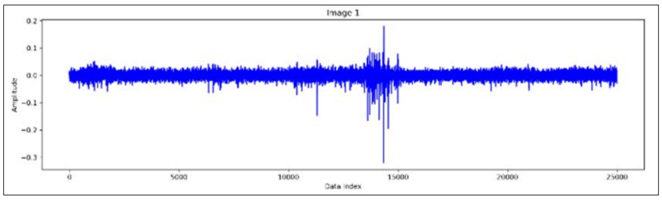
Figure 2: Schedule of MUA Recording and the Data used in the Event Classification Experiment [6].



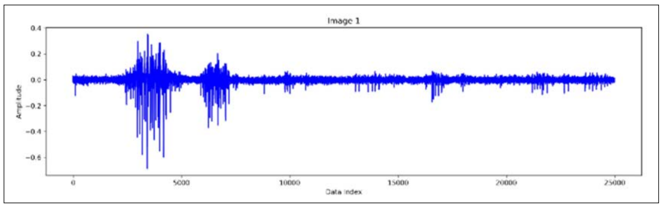
(a)Restraint stress (**restraint**)



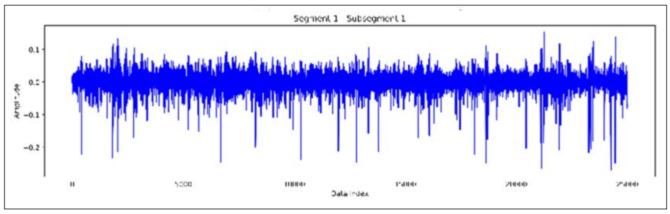
(b) Contact with a female rat (**female**)



(c) Contact with a Male Rat (**Male**)



(d) Contact with a Novel Object (**Object**)



(e) No Event (**Nothing**)

Table 1: Experience Events and the Number of Samples.

Event	Contents	Number
restraint	Restraint stress	400
object	Contact with a novel object	400
female	Contact with a female	400
male	Contact with a male	400
nothing	No event	400

Pattern Recognition of MUA Signals by Deep Learning Methods

To identify specific MUA time-series signal patterns resulting from different experiences, this study proposes the processing procedure shown in Figure 4. First, a Short-Time Fourier Transform (STFT) is applied to the original MUA signals, and the transformation results are saved as 256×256 image data. Next, the STFT images of the five classes of MUA are classified using VGG16, VGG16 + SVM, VGG19, VGG19 + SVM, ResNet50, and ResNet50 + SVM, respectively [11,12,15]. The reason for using multiple classifiers is that the classification accuracy varies depending on the target problem, and selecting the most suitable model for the target data of this study is necessary.

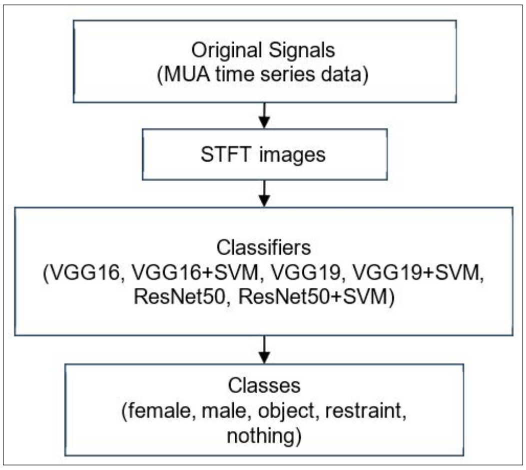


Figure 4: A Process of MUA signal recognition proposed in this Study.

STFT Image

When inputting time-series data into a classifier, using Short-Time Fourier Transform (STFT) as a preprocessing step and employing the time-series–frequency–power spectrum as the input image can achieve higher classification accuracy. Figure 5 shows examples of STFT images of MUA signals. For each original signal (1 second) consisting of 25,000 data points, the STFT window size was set to 1,024, and the window shift overlap (slide) was set to 512.

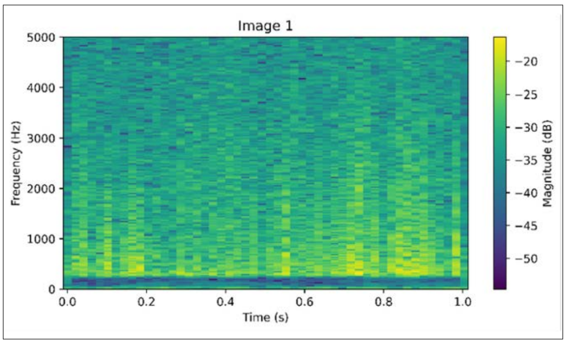


Figure 5: A Sample of STFT Image of an MUA Signal.

Deep Learning Models

VGG16 is a well-known deep convolutional neural network proposed by Simonyan and Zisserman in 2014 [11]. VGG16 consists of 13 convolutional layers and 3 fully connected layers, making it a representative deep learning model. In previous studies by the authors, the parameters of VGG16 trained on ImageNet were utilized through transfer learning, and the VGG16 + SVM model, which replaces the three fully connected layers with a Support Vector Machine (SVM), achieved the highest classification accuracy [13-16].

VGG19 is an extension of VGG16, with three additional convolutional layers, making it consist of 16 convolutional layers and 3 fully connected layers [11]. While VGG19 demonstrates superior performance in more complex image tasks, it requires higher computational costs. He et al. proposed Residual Network (ResNet) to build much deeper networks composed by convolutional neural networks without a loss of performance.

In the case of ResNet50,50 layers including convolutional, pooling, and fully connected layers are used similar to VGG16 or VGG19.

In this study, we investigate the performance of these models for the classfication/recognition of MUA patterns corresponding to different events experienced by rats at first, then we seek to elucidate the differences between the MUA patterns of left and right cerebral hemispheres triggered by specific events.

Experiment and Results

Fine-tuning of Deep Learning Models

MUA data shown in Table 1 was used to train deep learning models, i.e., VGG16, VGG16 with SVM, VGG19, VGG19 with SVM, ResNet50, and ResNet50 with SVM. The original MUA signals, which were time series data as shown in Figure 2, were transformed to be STFT images (see Figure 5) as the input images to deep learning models. 400 STFT images (224 × 224 × 3) for each event were used as training data and test data which ratio was 80% and 20%, respectively (Table 2), and 5-fold cross validation was executed. The number of training iterations for each model was determined based on the convergence of output errors. Specifically, VGG16 and VGG19 were trained for 80 iterations each, while ResNet50 was trained for 300 iterations. Figures 7 and 8 show the training process of the VGG19 with SVM model in cases of left and right brain MUA data were used.

Table 2: Image and Data Sizes Used in the Experiments.

Name	Value
sampling rate	25 kHz
input data (dimensionality)	224 × 224 × 3
number of training data	320 / event, 5 events (classes)
number of test data	80 / event, 5 events (classes)

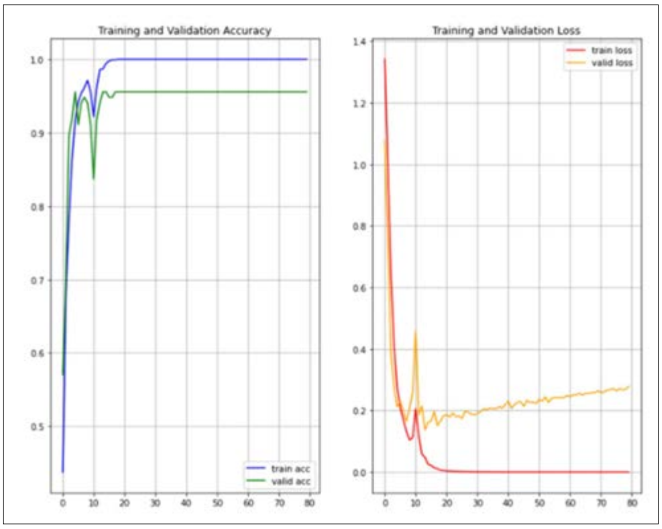


Figure 6: Learning Graphs of VGG19 with SVM (Data of the Left Brain in Rat C14) (Left: The Change of Recognition Rates by Training Times; Right: The Change of Loss by Training Times).

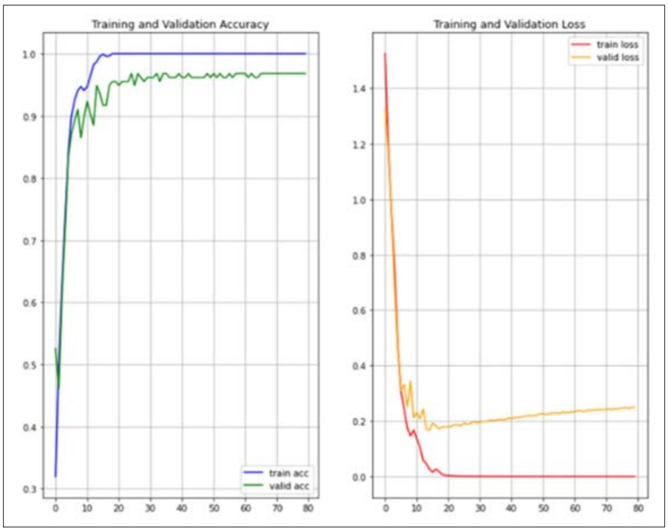


Figure 7: Learning graphs of VGG19 with SVM (Data of The Right Brain in Rat C14) (Left: The Change of Recognition Rates by Training Times; Right: The Change of Loss by Training Times).

MUA Pattern Recognition Results
The average classification rates of each model using the left and right brain data of C14 and C15 rats are shown in Figures 8 and 9, respectively. Among the deep learning models, VGG19 with SVM introduced in this study showed the highest classification accuracy, i.e., 96.79% and 95.56% for the MUA signals of right and left hippocampus of C14 rat, and 96.30% and 95.56% for C15. From the experimental results, it was found that the average classification accuracies of all models were approximately the same for MUA signals from the CA1 region of the hippocampus in both the left and right hemispheres. Comparisons between the left and right brains of C14 and C15 rats are shown in Figures 10 and 11, indicating that no individual differences were observed. However, when we examined the classification rates of MUA signals from the right and left hippocampus to specific events, it was revealed that the right brain of C14 and C15 rats had higher responses to the female and restraint events. To male and object events, the left brain of C14 rats responded higher, but C15 showed the opposite results. Model ResNet50, ResNet50 with SVM showed its priority to other models that the MUA signal related to no event (**nothing**). These predictions are derived by the event-specific classification rates shown in Figure 12.

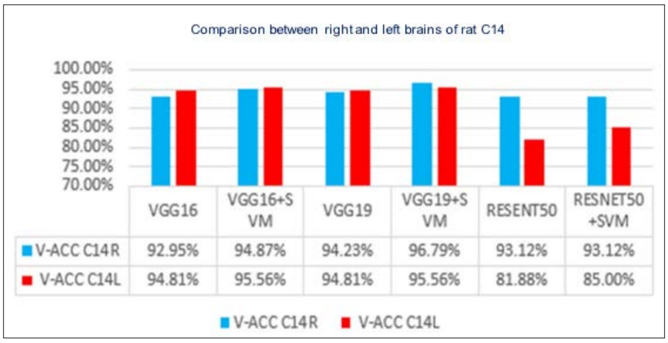


Figure 8: Recognition Rates of Different Deep Learning Models on left/right brain MUA patterns in the C14 Rat.

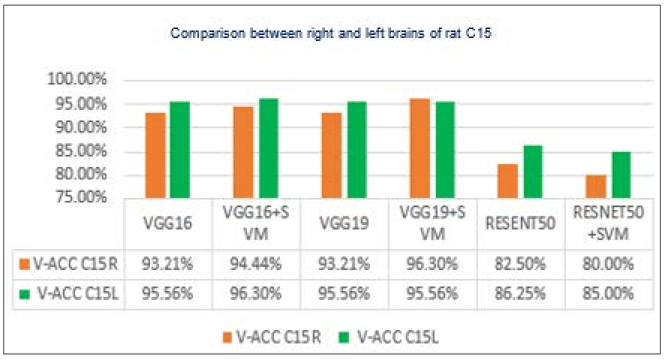


Figure 9: Recognition Rates of Different Deep Learning Models on left/right brain MUA patterns in the C15 Rat.

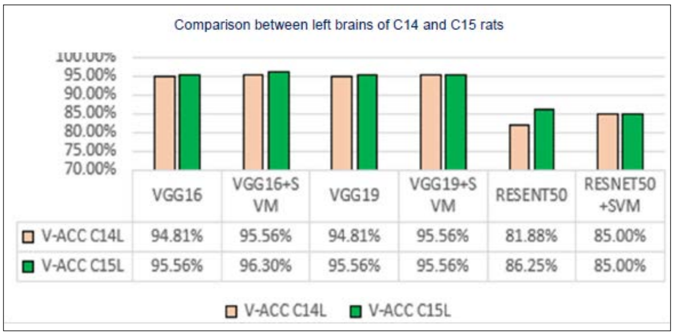


Figure 10: Recognition Rates of Different Deep Learning Models on left brain MUA patterns in C14 and C15 Rats.

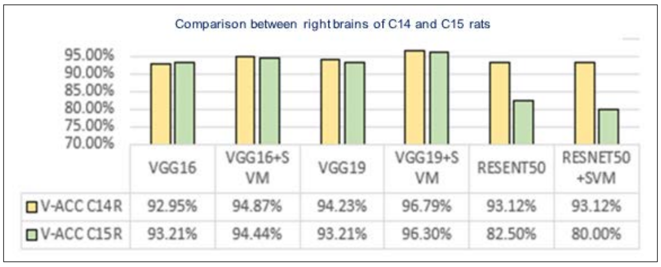


Figure 11: Recognition Rates of Different Deep Learning Models on right brain MUA patterns in C14 and C15 Rats.

Figure 12: Comparisons of Recognition Rates of MUA in Different Experiences Between Left-Right Brain by Different Deep Learning Methods.

Conclusions

In this study, we investigated how the MUA in the hippocampal CA1 area of two adult male rats responded to four types of experience: restraint stress (**restraint**), contact with a female rat (**female**), contact with a male rat (**male**), or observation of a novel object (**object**). Using deep learning methods, it was found that the firing activity of the hippocampus (CA1), located in the left and right hemispheres of the brain, showed different patterns depending on the events experienced. Although the average classification rates of the data related to these events did not show significant differences between the left and right hemispheres of the brain, and between individual rats, for the "**female**" and "restraint" experiences, however, the firing patterns in the right hippocampus were relatively more pronounced in both male rats. In contrast, for the "**male**" experience, the left hippocampus of both rats showed relatively more pronounced firing patterns. For the "**object**" experience, there was a significant difference in activity between the two rats: the right hippocampus of the C15 rat (C15R) showed a distinct firing pattern, while data measured from the same area of the C14 rat showed a relatively lower detection rate of specific ripple firings. In humans, the left hemisphere of the brain is considered to be primarily responsible for logical thinking, language, and analytical tasks, while the right hemisphere is involved in creativity, spatial awareness and emotional processing. Although the functional hemispheres in animal models are unknown, it is interesting to further explore and analyze our hypothesis by obtaining more MUA data from hippocampal CA1.

Conflict of Interest

The authors declare no conflicts of interest.

Acknowledgements

The study was supported JSPS KAKENHI Grant No. 19H03402, No.20K07724, No.22K12152, No.22H03709.

References

1. Andersen P, Morris R, Amaral D, Bliss T, O'Keefe J (2006) The hippocampus book. Oxford University Press.
2. Squire LR, Zola-Morgan S (1991) The medial temporal lobe memory system. Science 253: 1380-1386.
3. Mitsushima D, Sano A, Takahashi T (2013) A cholinergic trigger drives learning-induced plasticity at hippocampal synapses. Nature Communications 4: 2760.
4. Buzsáki G (2015) Hippocampal sharp wave-ripple: A cognitive biomarker for episodic memory and planning. Hippocampus 25: 1073-1188.
5. Joo HR, Frank LM (2018) The hippocampal sharp wave-ripple in memory retrieval for immediate use and consolidation. Nature Reviews Neuroscience 19: 744-757.
6. Ishikawa J, Tomokage T, Mitsushima D (2019) A possible coding for experience: ripple-like events and synaptic diversity. BioRxiv https://doi.org/10.1101/2019.12.30.891259.
7. Ishihara Y, Fujimoto K, Murai H, Ishikawa J, Mitsushima D (2023) Classification of ripple waves into experienced episodes using CNN. Proc. of 2023 RISP International Workshop on Nonlinear Circuits. Communications and Signal Processing (NCSP'23) pp: 90-93.
8. Ishihara Y, Tomohara Y, Fujimoto K, Murai H, Ishikawa J, Mitsushima D (2022) Extraction of episode-specific ripple firings patterns using Grad-CAM. Proc. of the 8th International Conference on Electronics and Software Science pp: 21-22.
9. LeCun Y, Bengio Y, Hinton G (2015) Deep learning. Nature 521: 436-444.
10. Sevaraiu RR (2019) Grad-CAM: Explanations from deep networks via gradient-based localization. arXiv: 1610.02391v4.
11. Simonyan K, Zisserman A (2014) Very deep convolutional networks for large-scale image recognition. preprint arXiv: 1409.1556.
12. He, K, Zhang X, Ren S, Sun J (2015) Deep residual learning for image recognition. In Proceedings of the IEEE conference on computer vision and pattern recognition pp: 770-778.
13. Kuremoto T, Sasaki T, Ishikwa J, Mabu S, Mitsushima D (2021) Hippocampus memory recognition using a deep learning method. Internal Medicine Review 7: 1-21.
14. Kuremoto T, Ishikawa J, Sasaki T, Mabu S, Mitsushima D (2023) Matching the ripple-wave to the episodic memory – a case study of rat, Stress Brain and Behavior 1: 19-30.
15. Kuremoto T, Ishikwa J, Mabu S, Mitsushima D (2024) Recognition of brain wave related to episode memory by deep learning methods. Artificial Intelligence Intech Open pp: 1-2.1
16. Russakovsky O, Deng J, Su H (2015) "ImageNet large scale visual recognition challenge", International Journal of Computer Vision 115: 211-252.