# Fuzzy Bio-Interface: Indicating Logicality from Living Neuronal Network and Learning Control of Bio-Robot

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*Abstract*— Recently, many attractive brain-computer interface and brain-machine interface have been proposed. The outer computer and machine are controlled by brain action potentials detected through a device such as near-infrared spectroscopy (NIRS) and electroencephalograph (EEG), and some discriminant model determines a control process. In this paper, we introduce a fuzzy bio-interface between a culture dish of rat hippocampal neurons and the khepera robot. We propose a model to analyze logic of signals and connectivity of electrodes in a culture dish, and show the bio-robot hybrid we developed. We believe that the framework of fuzzy system is essential for BCI and BMI, thus name this technology "fuzzy bio-interface". We show the usefulness of a fuzzy bio-interface through some examples.

## I. Introduction

Recently, many attractive brain-computer interface and brain-machine interface have been proposed [1], [2]. The outer computer and machine are controlled by brain action potentials detected through a device such as near-infrared spectroscopy (NIRS) and electroencephalograph (EEG), and some discriminant model determines a control process. However, under the condition where spontaneous action-potentials and evoked-action potentials are contained in brain signal asynchronously, we need a model that serves as an interface between brain and machine for a better stable control in order to prevent runaway reaction of machine. This interface plays a very important role to secure the stability of outer computer and machine. The interface has two kinds of functions: (1) a decoding of the response action potentials to the control signal of outside machine and computer, and (2) an encoding of the sensor signal of the outside machine and computer to pattern of stimuli in brain and neuronal networks. Unfortunately, it is very difficult to identify such a function for the interface between machine and living brain and neuronal networks. Here we consider such an interface within the framework of fuzzy system. As a result, our study

is supportive of this framework as a strong tool of the bio-interface. During the Japanese fuzzy boom in 1990's, fuzzy logic has been proven effective to translate human experience and sensitivity into control signals of machines. Tsukamoto [3] has argued a concept of fuzzy interface such that fuzzy sets is regarded as a useful tool to intermediate between language and mathematics. We believe that the framework of fuzzy system is essential for BCI and BMI, thus name this technology "fuzzy bio-interface".

In this paper, we introduce a fuzzy bio-interface between a culture dish of rat hippocampal neurons and the khepera robot [4]–[8]. We propose a model to analyze logic of signals and connectivity of electrodes in a culture dish, and show the bio-robot hybrid we developed [9], [10]. First, we discuss how to indicate the logicality and connectivity from living neuronal network in vitro [11]. Rat hippocampal neurons are organized into complex networks in a culture dish with 64 planar microelectrodes. A multi-site recording system for extracellular action potentials is used in order to record their activities in living neuronal networks and to supply input from the outer world to the vitro living neural networks. The living neuronal networks are able to express several patterns independently, and such patterns represent fundamental mechanisms for intelligent information processing [12], [13]. We follow the works of Bettencourt et al. [14] such that they classify the connectivity of action potentials of three electrodes on multi-site recording system according to their entropies and have discussed the characteristic of each classification. However, they only discuss the static aspects of connectivity relations among the electrodes but not the dynamics of such connectivity concerning how the strength of electrode connection changes when a spike is fired. To address this issue, we develop a new algorithm using parametric fuzzy connectives, that consist of both $t-norm$ and $t-conorm$ operators [15], in order to analyze those three electrodes.

Next, to control a robot, several characteristics of the living neuronal networks are represented as fuzzy IF-THEN rules. There are many works of robots that are controlled by the responses from living neuronal networks [16]–[21]. Unfortunately, they have not yet achieved a certain task that experimenter desired. We show a robot system that controlled by a living neuronal network through the fuzzy bio-interface in order to achieve such a task [4]–[8]. This fuzzy bio-interface consists of two sets of fuzzy IF-THEN rules: (1) to translate sensor signals of robot into stimuli for the living neuronal network, and (2) to control (i.e. to

determine the action of) robot based on the responses from the living neuronal network. We estimated the learning of living neuronal networks with an example of straight running with neuro-robot hybrid [9]. We show the usefulness of a fuzzy bio-interface through some examples.

## II. NEURON CULTURE AND MULTI-ELECTRODE ARRAY

The conduct of all experimental procedures was governed by The Animal Welfare, Care and Use Committee in AIST. The hippocampus neurons were prepared from a Wister rat on embryonic day 17-18 (E17-18) and cultured by the previously described method [12]. Briefly, neurons were dissociated by treatment with $0.175\%$ trypsin (Gibco, U.S.A.) and cultured by plating 500,000 cells in a $7mm$ diameter-glass ring on poly-D-lysine coated MED probe (Alpha MED Sciences, Japan), which has 64 planar placed microelectrodes. The medium is based on D-MEM/F12, supplemented with $5\%$ horse serum (Gibco, U.S.A.) and $5\%$ fetal calf serum (Gibco, U.S.A.).

The field action potentials were recorded 10-100days after the start of the culture. The spontaneous action potentials (sAPs) were gathered with the MED64 system (Alpha MED Sciences, Japan) at a 10-20 kHz sampling rate. Evoked field action potentials (eAPs) at 62 sites in the cultured networks were recorded with the MED64 system at a 20 kHz sampling rate. All experiments were carried out at room temperature $(20 - 25°)$. The recorded spikes were detected by our developing program, sorted and classified by the amplitude versus decay time distributions using k-means cluster cutting method and converted to event trains.

## III. ACQUISITION OF LOGICALITY IN NEURONAL NETWORKS

The fuzzy connective operators consist of $t - norm$ and $t - conorm$ operators. The $t - norm$ $T$ is a projective function expressed by $T(x, y) : [0, 1] \times [0, 1] \rightarrow [0, 1]$, which satisfies four conditions, boundary conditions, monotonicity, commutativity and associativity. The $t - norm$ operator $T$ includes logical product, algebraic product, bounded product and drastic product. The $t - conorm$ operator $S$ is dual function of the $t - norm$ operator, which is expressed by $S(x, y) : [0, 1] \times [0, 1] \rightarrow [0, 1]$, and includes logical sum, algebraic sum, bounded sum and drastic sum.

On the other hand, many parametric $t - norm$ and $t - conorm$ operators have been proposed. By changing the values of parameter, the parametric fuzzy operator expresses any operator between the drastic $t - norm$ and the drastic $t - conorm$. For example, the parametric fuzzy operator proposed by Schweizer [15] is expressed as follows:

$$T(x, y) = 1 - ((1 - x)^{p_n} + (1 - y)^{p_n}$$
$$- (1 - x)^{p_n} (1 - y)^{p_n})^{1/p_n} \quad (1)$$
$$S(x, y) = (x^{p_c} + y^{p_c} - x^{p_c} y^{p_c})^{1/p_c} \quad (2)$$

where, $p_n$ and $p_c$ are parameters.

By changing values of the parameter $p_n$ and $p_c$, the Schweizer $t-norm$ and $t-conorm$ express logical operator

$(p_n = p_c = \infty)$, algebraic operator $(p_n = p_c = 1)$ and drastic operator $(p_n = p_c = 0)$.
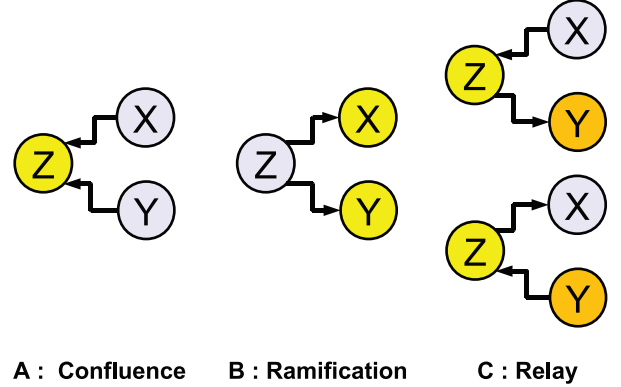
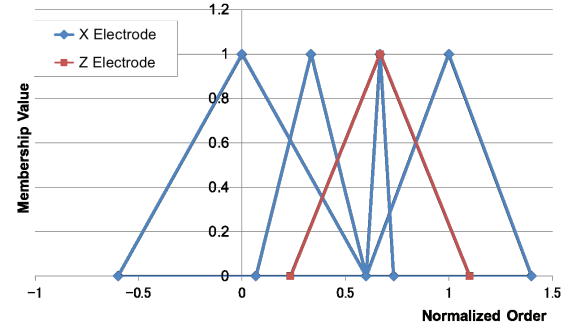

Fig. 2.   Connectivity of Electrodes



Fig. 3.   Fuzzy Sets of the First Example of Electrode Analysis

By the Schweizer $t - norm$ and $t - conorm$ operator, we formulate a new algorithm for analyzing logicality and connectivity of the living neuronal networks. Now, we selected an arbitrary set of three electrodes $x, y, z$, and analyzed the coherence pattern between the three electrodes. First, we distribute the data of pulse-time series in several time-bins, and define a time-delay deviation between time-bins of two electrodes. The proposed algorithm is shown in Figure 1. For the electrode $z$, we shape a fuzzy set of the pulse frequency, $F_i^z$, at the $i$-th time-bin by the following membership function which has the center $a_i^z$ and width $c_i^z$.

$$a_i^z = \frac{p_i^z - sp^z}{lp^z - sp^z} \quad (3)$$
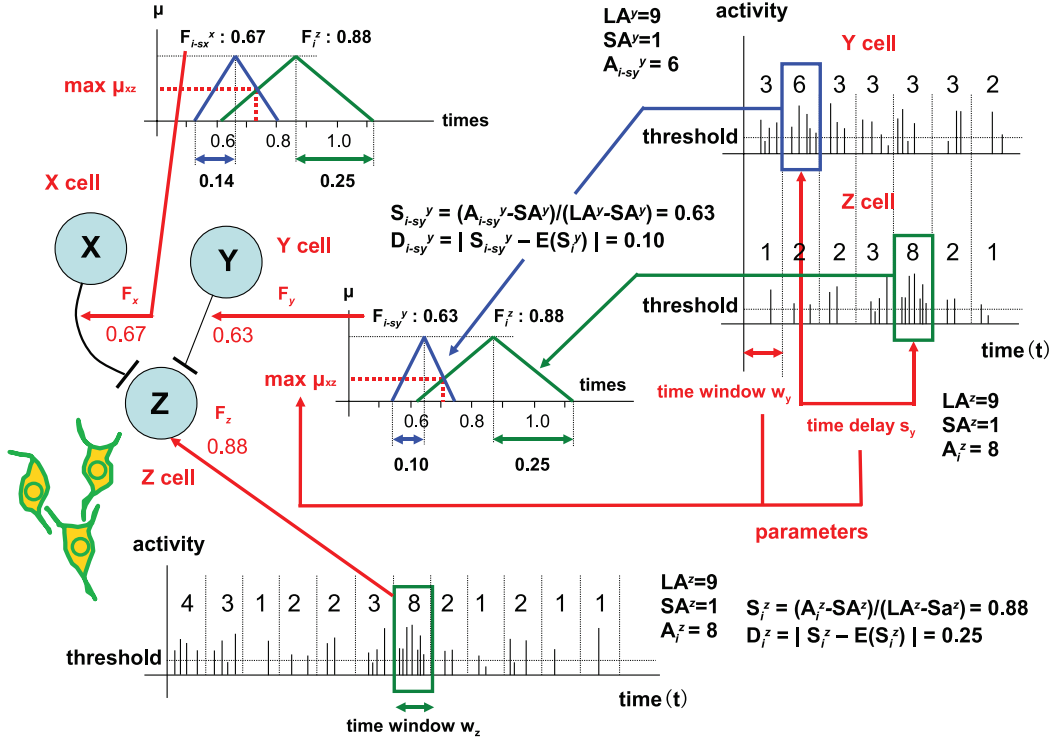$$c_i^z = |a_i^z - E(a_i^z)| \quad (4)$$

μ

$F_{i-s_x}^x : 0.67$   $F_i^z : 0.88$

max $\mu_{xz}$

times

0.6   0.8   1.0
0.14   0.25

**X cell**   **Y cell**

$LA^y=9$
$SA^y=1$
$A_{i-sy}^y = 6$

activity

**Y cell**

3  6  3  3  3  3  2

threshold

**Z cell**

$S_{i-sy}^y = (A_{i-sy}^y\text{-}SA^y)/(LA^y\text{-}SA^y) = 0.63$
$D_{i-sy}^y = | S_{i-sy}^y - E(S_i^y) | = 0.10$

**X**   **Y**

$F_x$ 0.67   $F_y$ 0.63

μ   $F_{i-sy}^y : 0.63$   $F_i^z : 0.88$

1  2  2  3  8  2  1

threshold

time (t)

max $\mu_{xz}$   times

time window $w_y$
time delay $s_y$

0.6   0.8   1.0
0.10   0.25

$LA^z=9$
$SA^z=1$
$A_i^z = 8$

**Z**   $F_z$ 0.88

**Z cell**

parameters

activity

4  3  1  2  2  3  8  2  1  2  1  1

threshold

$LA^z=9$
$SA^z=1$
$A_i^z = 8$

$S_i^z = (A_i^z\text{-}SA^z)/(LA^z\text{-}Sa^z) = 0.88$
$D_i^z = | S_i^z - E(S_i^z) | = 0.25$

time (t)

time window $w_z$

Fig. 1.   Algorithm for Analysis of Action Potentials in Cultured Neuronal Network

where, $p_i^z$ is the number of pulse at the $i$-th time-bin, $sp^z$ and $lp^z$ are the minimum and maximum number of $p_i^z$, respectively. $E(a_i^z)$ is the average value of $a_i^z$.

The membership function $F_{i-s_x}^x$ with the delay deviation $s_x$ of the electrode $x$ is shaped as same as the electrode $z$. Our purpose is to let the degree of coincidence, $\mu_{xz}^*$, between $F_i^z$ and $F_{i-s_x}^x$ maximize in the parametric conditions of the electrode $x$. To let the degree of coincidence maximize, the width of time-bin $w_x$ and the delay deviation $s_x$ are changed widely.

$$\mu_{xz} = \sup_t \mu_{F_i^z}(t) \wedge \mu_{F_{i-s_x}^x}(t) \qquad (5)$$

$$\mu_{xz}^* = \max_{w_x, s_x} \mu_{xz}. \qquad (6)$$

We calculate $\mu_{yz}^*$ between the electrode $y$ and the electrode $z$ as same as the electrode $x$ and the electrode $z$. When two couples of coincidence degrees, $\mu_{xz}^*$ and $\mu_{yz}^*$, were obtained, we can represent the connection of electrodes as a kind of connectivities as shown in Figures 2.

Next, we calculate the output of the Schweizer operator with two centers of membership functions, $a_{i-s_x}^x$ of the electrode $x$ and $a_{i-s_y}^y$ of the electrode $y$.

$$T(x,y) = 1 - ((1 - a_{i-s_x}^x)^{p_n} + (1 - a_{i-s_y}^y)^{p_n}$$
$$- (1 - a_{i-s_x}^x)^{p_n}(1 - a_{i-s_y}^y)^{p_n})^{1/p_n} \quad (7)$$

$$S(x,y) = ((a_{i-s_x}^x)^{p_c} + (a_{i-s_y}^y)^{p_c}$$
$$- (a_{i-s_x}^x)^{p_c}(a_{i-s_y}^y)^{p_c})^{1/p_c}. \quad (8)$$

We minimize the error deviation between the center $a_i^z$, and the Schweizer's output, $T(x,y)$ and $S(x,y)$, by changing the parameter $p_n$ of $t-norm$ and $p_c$ of $t-conorm$.

$$p^* = \{p_n, p_c | \min_{p_n, p_c}(|T(x,y) - a_i^z|, \; |S(x,y) - a_i^z|)\}. \quad (9)$$

To illustrate the proposed algorithm, we show a simple numerical example. The spike frequency of three examples of electrodes $x$ and $z$ are shown in Figures 3 to 5, and Table I. At each example, we search a time-bin of $x$ electrode which coincides most with the spike frequency of the sixth time-bin of electrode $z$. At the first example, the spike frequency "2" of the fourth time-bin of electrode $x$ coincided most with the spike frequency "2" of the sixth time-bin of electrode $z$ with the degree $\mu_{xz}^* = 1.0$ of fuzzy sets. Figures 3 shows the result of the first example. At the second and third examples, the spike frequency "3" of the ninth time-bin of electrode $x$ coincided most with the spike frequency "2" of the electrode $z$ with $\mu_{xz}^* = 1.0$, and the spike frequency "1" of the sixth time-bin of electrode $x$ coincided most with the the spike frequency "1" of the sixth time-bin of electrode $z$ with $\mu_{xz}^* = 0.44$, as shown in Figures 4 and Figures 5. We can understand these results intuitively.

Finally, we analyzed the logicality of neuronal networks. We detected action potential by the electrode of 64 channels in $20KHz$ for 120 seconds. For tangible data analysis, we picked a spike fired at the 60th electrode ($60el$), and selected three sets of electrodes for analysis. Figure 6 shows the
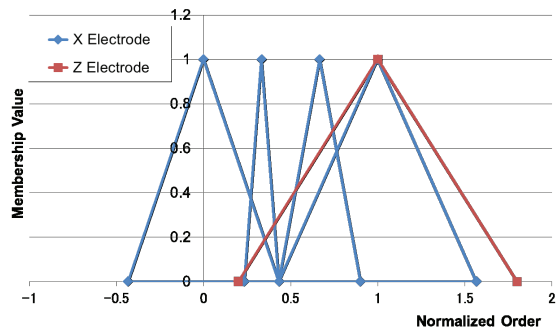
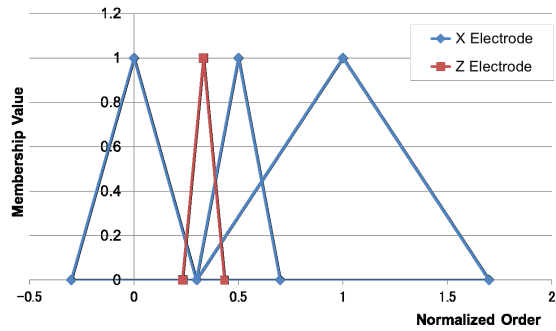Fig. 4.  Fuzzy Sets of the Second Example of Electrode Analysis



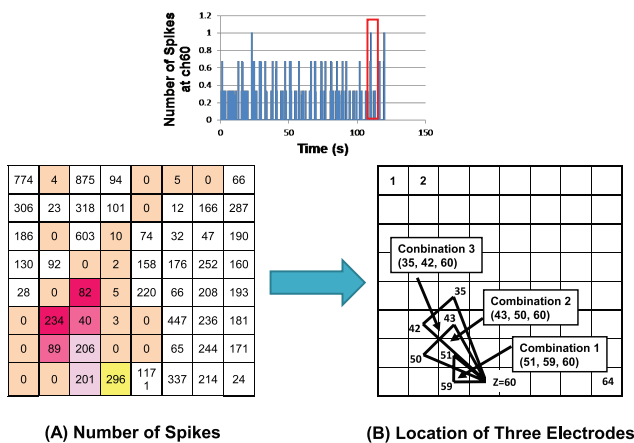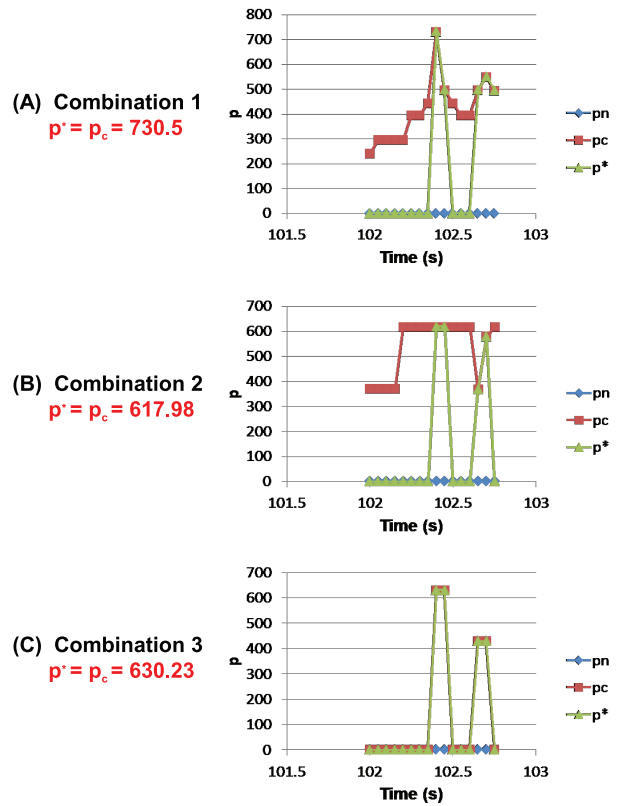Fig. 5.  Fuzzy Sets of the Third Example of Electrode Analysis



Fig. 6.  Experiments



Fig. 7.  Analysis of Action Potentials

location of three combinations. At the $60el$, we detected the increasing of pulse frequency at $102.4s$ after the pulse frequency drastically decreased to 6 times at $95s$. We focus this pulse fired at $102.4s$, and analyzed how this pulse influenced for the following three combinations.

1.   $(x, y, z) = (51el, \ 59el, \ 60el)$
2.   $(x, y, z) = (43el, \ 50el, \ 60el)$
3.   $(x, y, z) = (35el, \ 42el, \ 60el)$

The result is shown in Figure 7. At the first combination of the electrodes $(51el, 59el, 60el)$, the maximum degrees of coincidence were obtained as $\mu^*_{xz} = 0.85$, $\mu^*_{yz} = 0.75$ with $w_x = 11s$, $w_y = 10s$, and the parameter of Schweizer operator was converged to $p^* = p_c = 730.5$. At the second combination of the electrodes $(43el, 50el, 60el)$, the maximum degrees of coincidence were obtained as $\mu^*_{xz} = 1.0$, $\mu^*_{yz} = 1.0$ with $w_x = 11s$, $w_y = 10s$, and the parameter of Schweizer operator was converged to $p^* = p_c = 617.98$. At the third combination of the electrodes $(35el, 42el, 60el)$, the maximum degrees of coincidence were obtained as $\mu^*_{xz} = 0.76$, $\mu^*_{yz} = 0.91$ with $w_x = 11s$, $w_y = 10s$, and the parameter of Schweizer operator was converged to $p^* = p_c = 630.23$. From these results, we conclude that the pulse fired at $60el$ at $102.4s$ propagates to $(51el, 59el) \rightarrow (43el, 50el) \rightarrow (35el, 42el)$, and then, the parameters of Schweizer operator have been

TABLE I

EXAMPLES OF ELECTRODE ANALYSIS

| Time Bin | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|
| Example 1 | | | | | | | | | | |
| X | 2 | 3 | 2 | **2** | 0 | 1 | 3 | 1 | 3 | 1 |
| Z | 0 | 0 | 1 | 3 | 1 | **2** | 0 | 0 | 0 | 0 |
| CofX | 0.67 | 1.0 | 0.67 | **0.67** | 0.0 | 0.33 | 1.0 | 0.33 | 1.0 | 0.33 |
| WofX | 0.067 | 0.4 | 0.067 | **0.067** | 0.6 | 0.27 | 0.4 | 0.27 | 0.4 | 0.27 |
| CofZ | | | | | | **0.33** | | | | |
| WofZ | | | | | | **0.27** | | | | |
| $\mu$ | 1.0 | 0.6 | 1.0 | **1.0** | 0.36 | 0.52 | 0.6 | 0.52 | 0.6 | 0.52 |
| Example 2 | | | | | | | | | | |
| X | 2 | 3 | 2 | 0 | 0 | 1 | 0 | 1 | **3** | 1 |
| Z | 0 | 0 | 1 | 0 | 0 | **2** | 0 | 0 | 0 | 1 |
| CofX | 0.67 | 1.0 | 0.67 | 0.0 | 0.0 | 0.33 | 0.0 | 0.33 | **1.0** | 0.33 |
| WofX | 0.23 | 0.57 | 0.23 | 0.43 | 0.43 | 0.1 | 0.43 | 0.1 | **0.57** | 0.1 |
| CofZ | | | | | | **0.33** | | | | |
| WofZ | | | | | | **0.8** | | | | |
| $\mu$ | 0.68 | 1.0 | 0.68 | 0.19 | 0.19 | 0.26 | 0.19 | 0.26 | **1.0** | 0.26 |
| Example 3 | | | | | | | | | | |
| X | 0 | 1 | 0 | 2 | 0 | **1** | 0 | 2 | 0 | 0 |
| Z | 0 | 0 | 1 | 2 | 3 | **1** | 0 | 0 | 0 | 0 |
| CofX | 0.0 | 0.5 | 0.0 | 0.1 | 0.0 | **0.5** | 0.0 | 0.1 | 0.0 | 0.0 |
| WofX | 0.3 | 0.2 | 0.3 | 0.7 | 0.3 | **0.2** | 0.3 | 0.7 | 0.3 | 0.3 |
| CofZ | | | | | | **0.33** | | | | |
| WofZ | | | | | | **0.1** | | | | |
| $\mu$ | 0.17 | 0.44 | 0.17 | 0.17 | 0.17 | **0.44** | 0.17 | 0.17 | 0.17 | 0.17 |



Fig. 8. Living Neuronal Network and Robot
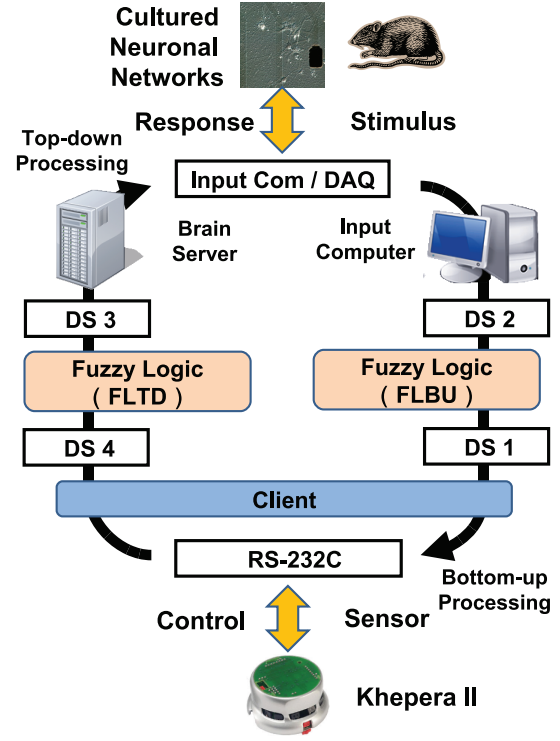


Fig. 9. Experimental Course

converged to infinity, $p^* = p_c = 730.5$ at $(51el, 59el)$, $p^* = p_c = 617.98$ at $(43el, 50el)$, and $p^* = p_c = 630.23$ at $(35el, 42el)$. These parameters mean logical sum. However, we should notice that the parameter of Schweizer operator at around $102.4s$ is $p^* = p_n = 0.0$, which means the drastic product. Given this result, we conclude that the logic of signals among the electrodes was shifted to the logical sum from the drastic product. Consequently, the logic of signals among electrodes drastically changes from the strong $AND$-relation to the weak $OR$-relation when a crowd of the pulses was fired.

## IV. FUZZY BIO-ROBOT SYSTEM

Fuzzy bio-robot includes two kinds of fuzzy logic units as fuzzy bio-interface, that is FLTD and FLBU. The FLTD, Fuzzy Logic unit in Top Down, is located in the top-down processing, and infers the rotation speed of robot actuator from the pattern of action potential in the multi-electrode array. The FLBU, Fuzzy Logic unit in Bottom Up, is located in the bottom-up processing, and infers two electrical stimulation points in the multi-electrode array from the output values of robot sensors. Figure 8 explains the architecture of living neuronal networks and robot.

We designed a closed loop, where the Khepera II receives the rotation speed of actuator from FLTD with eight inputs of patterns in [-20, 20] by multi-electrode array. Additionally, the multi-electrode unit receives stimulation points from FLBU with eight IR sensors of the robot. We designed 512 fuzzy rules with eight inputs and two output in FLBU and FLTD, respectively.

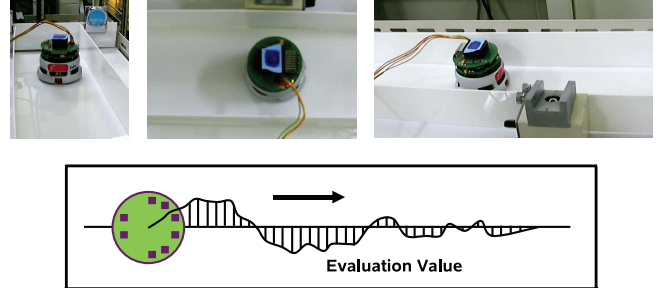Now, we should explain how to design fuzzy rules in the FLTD. First, we divide 64 electrodes in eight parts as inputs for FLTD, and we define two kinds of membership functions of "High" and "Low" at eight parts of electrodes. Two electrodes are arbitrarily selected as stimulus points, and we detect the potential response for the first stimulus from other 62 electrodes. The pulse pattern of potential responses is input to the antecedent part of fuzzy rules, and the membership value of each rule is calculated. Next, we detect the pulse pattern of potential responses for the second stimulus, and calculate the membership value of each rules. By two different membership values, we calculate the subtraction between them and assign the motor speed of robot actuators to rules whose differentials are large. We additionally adjust the value of motor speed better with the steepest descent method. If the neuronal networks have regularity of logical potential response well, the robot will

be controlled well.

To demonstrate the regularity of neuronal networks, we applied the fuzzy bio-robot system to the straight running. We estimate if the Khepera robot can run straight in a track without bumping into a wall. The running track is the length of 120mm and the width of 90mm. Figure 9 shows the running track.
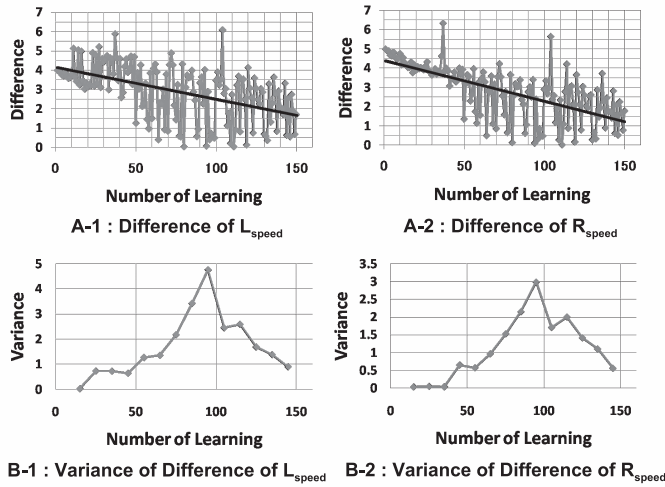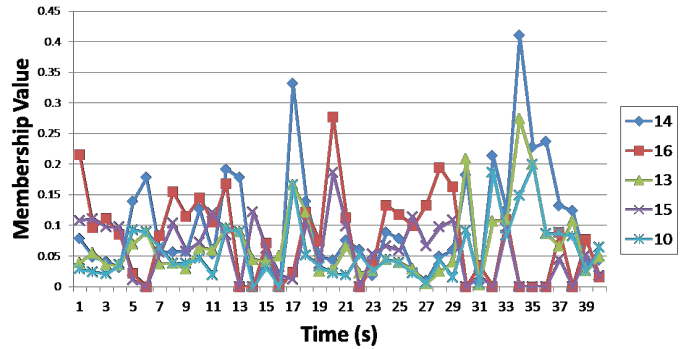


Fig. 10.    Learning of Fuzzy Rule



A : Membership Value for $L_{speed}$ of FLTD



B : Membership Value for $R_{speed}$ of FLTD

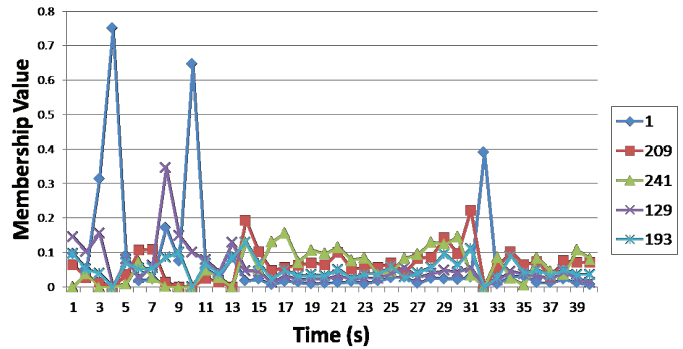Fig. 11.    Membership Values of FLTD until 40s

The deviation between the output of FLTD and the target output is shown in the part A of Figure 10. The variance of each 10 times of learning is shown in the part B. The deviation is gradually decreasing according to the number of learning. Actually, the deviation of the left actuator, $L_{speed}$, decreased by $40.3\%$ for 50 learning times and it becomes 1.673. The deviation of the right actuator, $R_{speed}$, decreased by $27.8\%$ and it becomes 1.224.

To discuss the fuzzy rules for avoiding collision with wall in more detail, we monitored fuzzy rules whose membership values are relatively higher until 40 seconds as shown in Figure 11. At the part A, the Khepera robot detects the wall in the left side, and turns the right with the 13th and 14th fuzzy rules, or the 15th and 16th fuzzy rules, simultaneously. The specificity of these fuzzy rules pattern appears regularly. In other words, the neuronal networks have regularity of logical potential response.

We observed the trajectory of the Khepera by camera which placed above of the track course. We image a base line drawing along the centerline of the track course from the start position of the Khepera. We detected the deviation between the base line and the trajectory of the Khepera, and defined absolute value of the deviation as the evaluation value. Figure 12 shows a trajectory of the Khepera. Figure 13 shows the change of the evaluation by trials of iteration. The evaluation value is decreased by trials of iteration, and the Khepera could run along the base line. We conclude that the decreasing of evaluation will come from mainly the learning of neuronal networks.
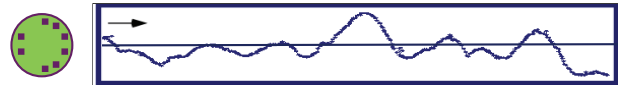


Fig. 12.    Trace of Khepera Robot

In addition, we calculated the ratio that the Khepera ran the whole distance by 20 trials in a course as shown in Table II. Among 20 trials, the Khepera completed the task 16 times, and it crashed on the wall and stopped there 4 times. In this result, we may conclude that the logic of signals among living neuronal networks represented as fuzzy IF-THEN rules for the fuzzy bio-interface is rather efficient and effective comparing to the other similar works. In such works, the success rate of $80\%$ is considered extremely high.
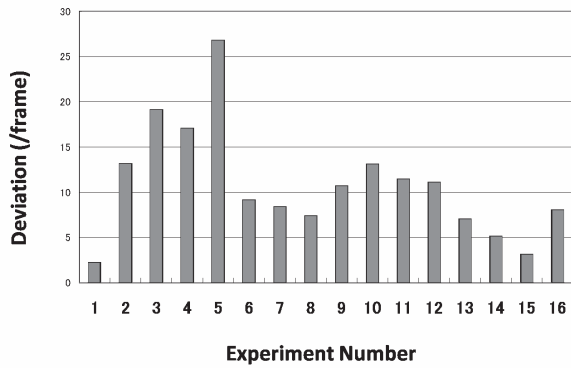
Fig. 13.   Trace Deviation of Khepera Robot

TABLE II

COMPLETED RUN RATE OF KHEPERA ROBOT

|  | Experiment Number | Rate |
|---|---|---|
| Completed Run | 1,3,4,5,6,7,8,9,10,14 | |
| | 15,16,17,18,19,20 | 80.0% |
| Incomplete Run | 2,11,12,13 | 20.0% |

## V. CONCLUSION

In this paper, we discussed how to indicate logicality of living neuronal network with data method of fuzzy connective operator, and applied fuzzy bio-interface to control fuzzy bio-robot. We should discuss the relationship between learning of living neuronal networks and adaptability of fuzzy logic more deeply in the near future.

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