A MUSICAL INSTRUMENT USING *IN VITRO* NEURAL NETWORKS

Eduardo R. Miranda[1], *Slawomir J Nasuto*[2], *Anna R. Troisi*[1], *Julia Downes*[2], *Antonino Chiaramonte*[1], *Matthew Spencer*[2], *Mark Hammond*[3], *Dimitris Xydas*[2], *Ben Whalley*[3], *Victor Becerra*[2], *Kevin Warwick*[2]

[1]Interdisciplinary Centre for Computer Music Research (ICCMR), University of Plymouth, UK
[2]Cybernetic Intelligence Research Group (CIRG), University of Reading, UK
[3]School of Pharmacy, University of Reading, UK

ABSTRACT

This paper presents a musical instrument, which uses *in vitro* neuronal networks to synthesise sounds. Cultures of dissociated neurons are grown on a dish with an embedded rectangular array of electrodes (MEA). Isolated neurons reconnect with one another via an extensive network of synaptically connected projections to form a dense monolayer of neurons. Cultures are provided with stimulation, which influences their activity, and can modify the culture's state. The core of the sound synthesis engine of our musical instrument is a monophonic additive synthesizer using sinusoidal oscillators. We devised a method to generate frequencies, phase and amplitude values for the oscillators from the electrical activity of the neurons.

1. INTRODUCTION

The field of Computer Music has evolved in tandem with the field of Computer Science. For example, computers have been programmed to generate music as early as the beginning of the 1950's [1]. The *Illiac Suite for String Quartet*, composed in the USA in late 1950's by Lejaren Hiller (composer) and Leonard Isaacson (mathematician), is often cited as the first piece of music using materials generated by a computer; e.g., the fourth movement was generated using a Markov chain. Nowadays, the computer is ubiquitous in many aspects of music, ranging from software for musical composition and production, to systems for distribution of music over the Internet. Therefore, it is very likely that future developments in Computer Science will continue to have an impact in music.

We are interested in exploring ways in which unconventional modes of computation may provide new directions for future developments in Computer Music. Research into unconventional computing is aimed at computational paradigms other than the standard von Neumann architecture, which have prevailed in computing since the 1940s [2]. In short, unconventional computation takes the computation (or part of it) from the silicon chip into the "real world", thereby harnessing the immense parallelism and non-algorithmic openness of physical systems.

New computational paradigms based on and/or inspired by the principles of information processing in physical, chemical and biological systems are promising new venues for the development of new types of computers, which may eventually supersede classical paradigms. For instance, it has been reported that reaction-diffusion chemical computers have been capable of performing a number of complex computational tasks, including the design of logical circuits [3].

There has been a growing interest in research into the development of hybrid wetware-silicon devices for non-linear computations using cultured brain cells. The ambition is to harness the intricate dynamics of *in vitro* neuronal networks to perform computational tasks [4]. This paper presents a musical instrument, which uses *in vitro* neuronal networks to synthesise sounds.

Related attempts to utilise biological networks as non-standard computational devices and exploring the interface between art and computer science include a technique to sonify data from *in vitro* neural networks off-line, [5] and an interesting artistic application of *in vitro* neural networks, to a graphical art installation [6] in which a device with cultured neurons was connected to a robotic arm, which drew images.

2. OVERVIEW OF THE INSTRUMENT

The overall architecture of our instrument is shown in Figure 1. Its synthesis engine is an additive synthesiser, implemented in Max/MSP, comprising ϑ sinusoidal oscillators; in the example presented in this paper $\vartheta = 16$. The instrument is played through a MIDI controller. It currently responds only to MIDI note messages and it is monophonic.

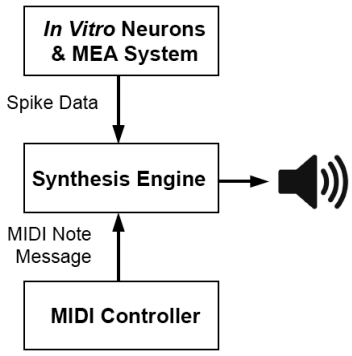

Figure 1. The overall architecture of the instrument.

The timbre of the instrument is defined by the electrical membrane potential fluctuations of the *in vitro* neurons. The membrane potential either fluctuates around the baseline when the neuron is at rest, or otherwise when

activated by its input it generates rapid stereotypical waveform, called an action potential, or a spike. The neurons constantly feed the synthesis engine with $n = \vartheta$ channels of neural activity; i.e., spike data. Each channel provides information to drive ϑ oscillators. One of the oscillators is set to produce a "fundamental frequency" which is calculated as a function of the respective note played on the MIDI controller. In short, the MIDI note provides a seed for the calculation of all partials using information provided by the *in vitro* neurons.

Even though the neurons are constantly feeding the oscillators with information, a sound is synthesised only when the system receives a MIDI note on message.

3. IN VITRO NEURONS, STIMULATION AND DATA CAPTURE

Cultures of dissociated cortical neurons (taken from rat cortices) are grown on a dish with an embedded rectangular array of electrodes (MEA)[1]. Once seeded, initially isolated neurons reconnect with one another via extensive network of synaptically connected projections to form a dense monolayer of neurons, [9]. Approximately 2,500-10,000 neurons live on the ~1mm^2 recording area of the MEA. The cells feed on nutrients supplied within the cell-culture medium that surrounds them, and they can live for several months.

Cultures may be provided with a range of stimulations, either pharmacological or electrical. Stimulation influences the cultures activity, and can modify the culture's state [10][11], or change the properties of the underlying connections between neurons [12]. Such preparations are used to investigate neuronal plasticity [12], learning and memory [9], and brain disease [13], they also enable the neurone-level effects of pharmacological agents to be investigated [10].

Figure 2 depicts the MEA-based system. Electrical pulses provide stimulation (top left) via selected electrodes to the surrounding neurons (top middle). The MEA (top right) is connected to stimulation and recording hardware. Bottom right is the electrical activity recorded from each channel of the MEA (a difference between fluctuations of the potentials recorded between two electrodes, one of which is a reference electrode). This activity corresponds to variations of field potentials of the clusters of neurons located within the vicinity of each electrode. The signals from each electrode are amplified (1100x gain) and recorded (25 kHz sampling rate), a threshold is applied to detect action potentials in the neurons' activity. A spike detected on one of the channels is shown in more detail (bottom left). The spikes detected on each channel, along with their timestamps are passed to the synthesis engine.

Data used in the present study were captured from cultures provided with three frequencies of electrical stimulation (0.33 Hz, 2 Hz, and 20 Hz). The culture's immediate response to these stimuli influenced the dynamics of the system.

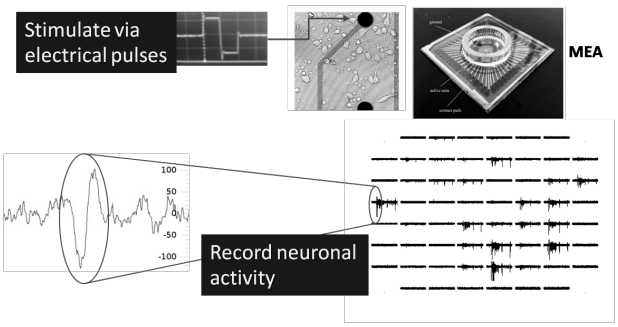

Figure 2. The MEA-based system.

4. SYNTHESIS ENGINE

The core of the sound synthesis engine of our musical instrument is a monophonic additive synthesizer using sinusoidal oscillators [7]. We devised a method to generate frequencies, phase and amplitude values for the oscillators from the electrical activity of the neurons.

We currently use 16 oscillators and although our MEA device provides up to 59 channels of neural data, we use only a sub-set of 16 channels. At this stage of our research, more electrodes would not capture activity that would convey salient perceptual difference in the resulting sound.

Sixteen channels of raw MEA data are pre-processed in order to produce streams of negative spike profile voltage values expressed in terms of μV. We decided to filter the incoming data in order to use only negative spikes that fall between -59μV and -12μV. Then, for the sake of convenience, we take the absolute values in order to work with positive finite numbers and subtract 12 in order to shift the values down to the range between 0 and 47.

The frequency value of the partials is calculated as a function of the MIDI note value played on the MIDI controller. For instance, MIDI note number 60, which is the note C4, yields the frequency 261.63Hz. The actual frequencies of the partials are obtained by multiplying the frequency of the MIDI note by an index i_n, which is obtained for each MEA channel C_n where γ_n is the

[1] We should mention that these neurons are not dissociated specifically for this project. Rather, they were subject of other experiments aimed at studying memory and data storage mechanisms in the brain. Ultimately, these are aimed at a better understanding of development and disorders that affect the brain such as Alzheimer's and Parkinson's disease.

associate value for this channel, n = {1, …, 16}, as follows:

$$i_n = \left(\frac{\gamma_n \times 14}{47} \right) + 2 \quad (1)$$

The phases ϕ_m for partials number 1 to 16 are calculated as follows, where $0 \le \phi_m \le 1$, $m = n = \{1, \ldots, 16\}$:

$$\phi_m = \frac{\gamma_n}{47} \quad (2)$$

Finally, the loudness of the fundamental frequency is determined by the MIDI velocity value, which in terms of MIDI values varies from 0 to 127, where 0 corresponds to silence and 127 corresponds to the maximum loudness.

With respect to calculating the individual amplitudes of the partials, we have assumed that the waveform yielded by our instrument would mimic the behavior of saw or square waveforms, whereby the amplitude of a partial is proportionally inverse to its position in the spectrum; the further its distance from the fundamental frequency, the lower its amplitude.

The amplitudes are calculated as follows. The amplitude values for the oscillators vary from 0 to 1, proportionally to spike values γ. The amplitude of the first partial is always equal to the MIDI velocity value mapped between 0 to 1. The amplitudes of the remaining 15 partials are obtained by multiplying the amplitude of the first partial by an index k_m. Then, the lowest amplitude of the other partials is 1/16 = 0.0625 of the fundamental amplitude, because we have 16 oscillators, and the highest value of k_m is 1 – 0.0625 = 0.9375. Therefore:

$$k_m = \left(\frac{\gamma_n \times 0.875}{47} \right) + 0.0625 \quad (3)$$

where $m = n = \{1, \ldots, 16\}$. The value 0.875 is the difference between 0.9375 and 0.0625.

In order to cater for the attack and release of the sounds, the instrument is furnished with a customisable breakpoint function envelope. In addition to the ability to specify proportional durations for the attack and decay of the sound, it is also possible to shape the amplitude of the sustain portion of the sound.

Also, we have implemented a version of the instrument where the spectrum of the resulting sound is further enriched by means of Frequency Modulation, or FM [7]. We use an additional sinusoidal oscillator to act as the modulator of the fundamental oscillator, which acts also as the carrier of the FM synthesis.

5. FUTURE WORK

Controlling behavior and understanding how information is coded and processed by the cultured neurons are two Holy Grails of research into *in vitro* neuronal networks.

In addition to building a musical instrument for artistic use, our research also addresses more basic scientific research problems, such as understanding the behavior of *in vitro* neuronal networks and their dynamics in order of to perform computational tasks. To this end, we are exploring the possibility of steering the networks with sound stimuli. In order to perform future experiments, we plan to mike the output of the synthesizer and perform a cochleogram analysis. The results of the analysis will be converted into stimuli data for the neurons on the MEA (Figure 3).

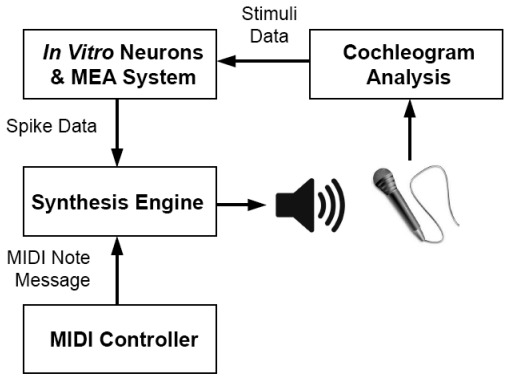

Figure 3. Training the system through auditory feedback.

6. CONCLUDING REMARKS

We have not been concerned with studying the computational properties of the cultured neurons. Rather, we have been primarily interested in studying how their behavior can be rendered into sounds. There are still a number of fundamental hard challenges to be addressed before one can study the plasticity of *in vitro* networks effectively. Inducing long-term changes in neuronal activity in response to stimulation is a challenging research area [14]. Moreover, the neurons require precisely controlled laboratory conditions, and maintaining cultures in a closed-loop setup for extended periods is non-trivial. Much research is needed to establish the most suitable experimental protocol. The experimental scenario described in section 5 cannot be developed satisfactorily until such techniques are mastered.

An important property of a musical instrument is its ability to produce different types of sounds with a certain degree of predictable control [8]. However, these changes should not hamper the identity of the instrument. As in the case of standard electronic musical instruments, different "notes" can be produced through the MIDI keyboard. One important property of our

instrument is that the spectrum of the sounds is dynamic; that is, it changes constantly. However these changes are subtle in the sense that the general identity of the instrument is preserved.

With improved means to control the neurons we hope to be able tune the instrument to produce different timbres. And once we have the means to harness the behavior of the network to perform computational tasks, then we hope to explore the possibility of building *intelligent living instruments* capable of high-level control of sounds interactively.

This work was in part supported by EPSRC grants EP/D080134/1 and EP/F033036/1, and by the Faculty of Arts with Plymouth University.

7. REFERENCES

[1] Doornbusch, P. (2005). *The Music of the CSIRAC: Australia's First Computer Music*. Victoria, Australia: Common Ground.

[2] Calude, C., Casti, J. L. and Dinneen, M. J. (Eds.) (1998). *Unconventional models of Computation: Proceedings of the 1st International Conference – UMC'98*. New York: Springer.

[3] Steinbock, O., Toth, A and Showalter, K. (1996). "Chemical wave logic gates". *Journal of Physical Chemistry* 100:18970-18975.

[4] DeMarse, T., Wagenaar, D. A., Blau, A. W. and Potter, S. M. (2001). "The Neurally Controlled Animat: Biological Brains Acting with Simulated Bodies". *Autonomous Robots* 11(3):305-310.

[5] Miranda, E. R., Bull, L., Gueguen, F., Uroukov, I. S. (2009). "Computer Music Meets Unconventional Computing: Towards Sound Synthesis with In Vitro Neuronal Networks", *Computer Music Journal* 33(1):9-18.

[6] Potter, A. M., DeMarse, T. B., Bakkum, D. J., Booth, M. C., Brumfield, J. R., Chao, Z., Madhavan, R., Passaro, P. A., Rambani, K., Shkolnik, A. C., Towal R. B. and Wagenaar, D. A. (2004). "Hybrots: Hybrids of Living Neurons and Robots for Studying Neural Computation", *Proceedings of Brain Inspired Cognitive Systems*. Stirling, UK.

[7] Miranda, E. R. (2002). *Computer Sound Design: Synthesis Techniques and Programming*. Oxford, UK: Focal Press.

[8] Miranda, E. R. and Wanderley, M. M. (2006). *New Digital Musical Instruments: Control and Interaction beyond de Keyboard*. Middleton, WI: A-R Editions.

[9] Marom, S. and G. Shahaf (2002). "Development, Learning and Memory in Large Random Networks of Cortical Neurons: Lessons Beyond Anatomy", *Quarterly reviews of biophysics* 35(1): 63-87.

[10] Tateno, T., Y. Jimbo, et al. (2005). "Spatio-Temporal Cholinergic Modulation in Cultured Networks of Rat Cortical Neurons: Spontaneous Activity", *Neuroscience* 134:425-437.

[11] Vajda, I., J. Van Pelt, et al. (2008). "Low-Frequency Stimulation Induces Stable Transitions in Stereotypical Activity in Cortical Networks", *Biophysical Journal* 94:5028-5039.

[12] Jimbo, Y., T. Tateno, et al. (1999). "Simultaneous Induction of Pathway-Specific Potentiation and Depression in Networks of Cortical Neurons", *Biophys. J.* 76:670-678.

[13] Chao, H., Y.,, Z. Liu, W.,, et al. (2003). "Effect of Recurrent Epileptiform Discharges Induced by Magnesium-Free Treatment on Developing Cortical Neurons in Vitro", *Brain Research* 142(1):1-6.

[14] Wagenaar, D. A., Pine, J., Potter, S.,M. (2006). "Searching for Plasticity in dissociated cortical cultures on multi-electrode arrays", *Journal of Negative Results in BioMedicine* 5(16).