Proceedings of the 17th
IEEE International Conference on Nanotechnology
Pittsburgh, USA, July 25-28, 2017

Set Point Regulation of Astrocyte Intracellular Ca^{2+} Signalling

Michael Taynnan Barros[1] and Subhrakanti Dey[2]

***Abstract*— Neurodegenerative diseases are the current centre of attention in medicine due to their increased physiological and psychological burden on the ageing society and in the other hand the lack of efficient treatment to them. In parallel, nanotechnology opens possibilities to study neurodegeneration in the molecular level and uncover cellular properties at the nanoscale that possibly allow disease control using novel system biology methods. The communication between neurons and astrocytes explains how a failure in their communication impact neuronal activity, and how the intracellular Ca^{2+} signalling of astrocytes can interfere in the synaptic quality. This paper presents a theoretical investigation of a feed-forward and feedback control technique to regulate the quantity of IP_3 that determines the concentration of Ca^{2+} emitted from intracellular signalling. The analysis of the control model showed that the quantity of Ca^{2+} signalling can be stabilised at a desired level. A potential application is to facilitate the Ca^{2+} concentration around this desired level to maintain cellular homoeostasis for longer periods of time, which can lead to a technology for preventing neurodegenerative diseases. The proposed approach can result in novel solutions for both nanobiology and nanomedicine development, where synthetic biology can be used to program the control functionality into the cells. Other ways of implementing such technology are also explored, including nanoparticles, implantable devices and molecular communications.**

I. Introduction

Neurodegenerative diseases cause progressive, incapacitating cognitive, behavioural, and motor dysfunction [1]. In Europe, 35% of all disease burden is due to brain disorders [2], and depressive disorders are the single biggest source of disability in the high-income countries, and the third worldwide [3]. They are related to the quality of synapses in neuronal communications. The poor concentration of glutamate inside the synaptic channel will lead to the wrong propagation of the synapses, causing symptoms of most neurodegenerative diseases like lack of memory, insomnia and depression. Current treatment techniques of neurodegenerative diseases are limited to drugs that are not effective as they only help to eliminate symptoms, but not treat the disease and indeed are far away from achieving their cure.

Recently, nanotechnology has gained attention for the novel techniques that have emerged for treating diseases as well as neurodegeneration. Nanoparticles, control theory, synthetic biology and molecular communications are examples of such techniques that result in a targeted, optimised, mobile and reliable drug or gene therapies [4], [5]. This collection of techniques enables advanced disease treatment by accessing the molecular level of cells or tissues, which requires an understanding of certain cellular properties as well as their regulation.

The evident control of Ca^{2+} levels in astrocytes enables the indirect uptake of the glutamate release for improvement of the synaptic transmission [6], [7]. The first encountered challenge is to provide a theoretical approach for realising the possibility of controlling the astrocytes' Ca^{2+} concentration and, therefore, create a mathematical framework that will form the basis for potential alternative approaches to preventing neurodegenerative diseases.

For this purpose, the cytosolic Ca^{2+} is the focus of this study. Moreover, internal Ca^{2+} signalling is characterised by oscillations invoked by a particular range of IP_3. The elimination of such oscillatory behaviour will give a stable level of the desired Ca^{2+} concentration. The IP_3 is then a decisive factor for Ca^{2+} regulation, in which its increase is controlled by a constant factor, β. A mathematical model captures this dependent behaviour that enables control of the intracellular Ca^{2+} signalling by regulating IP_3 levels. This mathematical model is based on the **feed-forward and feedback** control mechanism to perform indirect astrocytes' cytosolic Ca^{2+} concentration regulation. Since proteins are more easily stimulated, IP_3 is then used as a starting point where its control will lead to the accurate stimulation of Ca^{2+} ions [8], [9]. On the other hand, the control function needs to be simple enough to be implemented in nanoscale settings, and therefore the model is developed upon needs to account only for the most important system characteristics.

The contributions of this paper are:

- **Control of IP_3-induced Ca^{2+} oscillations.** The mathematical model shows how a desired stable level of Ca^{2+} is achieved by removing the oscillation of the Ca^{2+}. The model also shows robustness over different Ca^{2+} signal patterns.
- **Review of implementation techniques of the control function.** Even though this paper remains in the theoretical domain, a study of possible implementation techniques that enables the control to be achieved both *in-vitro* and *in-vivo* are explored and include nanoparticles, synthetic biology, implantable

[1] Michael Taynnan Barros is with the TSSG, Waterford Institute of Technology, Ireland mbarros@tssg.org

[2]Subhrakanti Dey is with the Signal and Systems group, Department of Engineering Sciences, Uppsala University, Sweden subhra.dey@signal.uu.se

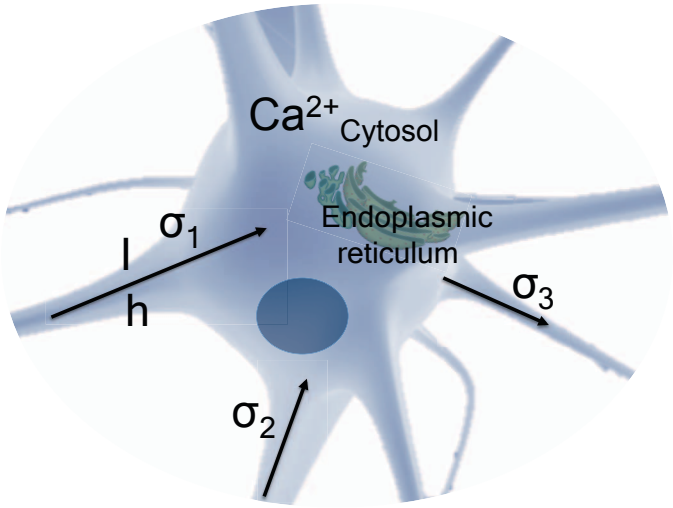

Fig. 1: Intracellular Ca^{2+} signalling model for astrocytes. The flux/efflux rates control the concentration of Ca^{2+} signalling. The function σ_1 models the Ca^{2+}-induced Ca^{2+} release (*CICR*). The function σ_2 denotes the leakage of Ca^{2+} ions to the cytosol from the *sarco(endo)plasmic reticum* (*SERCA*), and the function σ_3 denotes the efflux of Ca^{2+} from the *sarco(endo)plasmic reticulum* to the *endoplasmic reticulum*.

devices and molecular communications.

The rest of this paper is organized as follows. Mathematical modelling of astrocytes intracellular Ca^{2+} signalling is presented in §II, followed by the problem statement, §III, and the feed-forward feedback control in §IV. The results and analysis are presented in §V. The methodologies for potential implementation *in-vitro* and in-vivo are finally discussed in §VI followed by concluding remarks in §VII.

II. Astrocyte Intracellular Ca^{2+} Signalling

The intracellular Ca^{2+} signalling model in astrocytes consists of state equations for the Ca^{2+} concentration in the cytosol (C) (Eq. 1), kinetics of IP_3 receptors (h) (Eq. 2) as well as the IP_3 concentration (I) (Eq. 3). This model is proposed in [10], and a visual illustration of the model is presented in Fig. 1. The main state equations are defined as follows:

$$\frac{dC}{dt} = \sigma_1 + \sigma_2 - \sigma_3, \tag{1}$$

$$\frac{dh}{dt} = \frac{H - h}{\tau}, \tag{2}$$

$$\frac{dI}{dt} = \frac{1}{\alpha}(i_0 - I) + \beta\mathcal{H}(E_0 - 35) \tag{3}$$

where α is the degradation time constant of IP_3 concentration, i_0 is the IP_3 concentration in equilibrium, β is the production rate of IP_3 ions, E_0 is the presynaptic potential and $\mathcal{H}(.)$ is the Heaviside function. *Ca^{2+}-induced Ca^{2+} release* (*CICR*) is the trigger process of Ca^{2+} ions from the *sarco(endo)plasmic reticulum* by existing Ca^{2+} ions within the cytosol. The quantities H, τ are defined in (8), (9, (10). The function σ_1 models the *CICR* and is defined as:

$$\sigma_1 = vm^3h^3[c_0 - (1 + C_1)C] \tag{4}$$

where v is the maximal *CICR* rate, c_0 is the total cell-free Ca^{2+} concentration depending on the cytosol volume, and C_1 is the ratio between the cytosol and endoplasmic reticulum volume.

The IP_3 and Ca^{2+} ion binding process responsible for providing stable IP_3 kinetics are represented as:

$$m = \left(\frac{I}{I + d}\right)\left(\frac{C}{C + d_3}\right) \tag{5}$$

where d is the IP_3 dissociation constant and d_3 is the Ca^{2+} activation-dissociation constant.

The function σ_2 denotes the leakage of Ca^{2+} ions to the cytosol from the *sarco(endo)plasmic reticum* and is represented as:

$$\sigma_2 = v_1[c_0 - (1 + C_1)C] \tag{6}$$

where v_1 is the maximal rate of Ca^{2+} ions leakage from the endoplasmic reticulum.

The efflux of Ca^{2+} from the *sarco(endo)plasmic reticulum* to the *endoplasmic reticulum* (*SERCA*) is represented as:

$$\sigma_3 = \frac{v_2C^2}{k^2 + C^2} \tag{7}$$

where v_2 is the maximal rate of SERCA uptake, and k is Ca^{2+} binding affinity.

The following equations are important for modelling h:

$$H = \frac{Q}{Q + C} \tag{8}$$

$$\tau = \frac{1}{a(Q + C)} \tag{9}$$

$$Q = \frac{I + d}{I + d_2}d_1 \tag{10}$$

where d_1 is the Ca^{2+} inactivation dissociation constant, d_2 is the IP_3 dissociation constant and a is the IP_3 receptors binding rate for Ca^{2+} inhibition.

III. Problem Statement

Regulating Ca^{2+} levels in astrocytes can indirectly control the glutamate release and potentially improve the synaptic transmission in neuronal communication. The main challenge is to provide an analysis of the astrocytes' Ca^{2+} concentration and, therefore, create a theoretical framework that can be developed towards future approaches to preventing neurodegenerative diseases.

For this purpose, the problem of controlling levels of Ca^{2+} in the cytosol is investigated in this paper. More specifically, internal Ca^{2+} signalling is characterised by

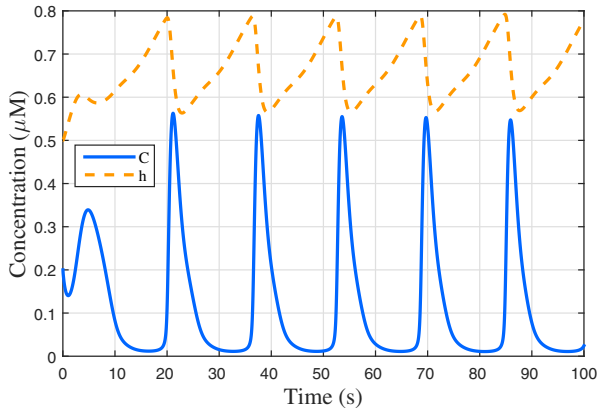

Fig. 2: Ca^{2+} oscillation with respect to time. In this illustration the $IP_3 = 0.5$ μM. The Ca^{2+} concentration (C - blue line) oscillates alongside with the kinetics of IP_3 receptors (h - dashed yellow line).

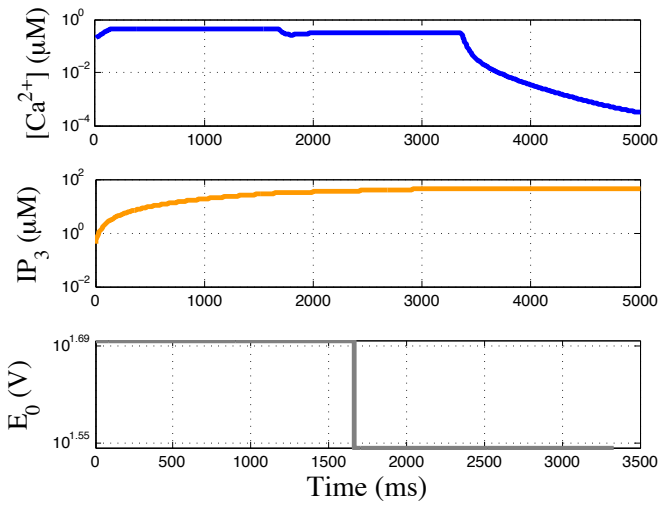

Fig. 3: Effect of both IP_3 and the E_0 on the Ca^{2+} concentration. The Ca^{2+} concentration is highly dependent on the increasing factor of IP_3 and, therefore, so is the stability.

oscillations invoked by a certain range of IP_3. Fig 2 shows the Ca^{2+} oscillation at $IP_3 = 0.5$ μM and when production rate of IP_3 ions (β - eqn. 3) varies from 0.1-1.5 μM/s. The elimination of such oscillatory behaviour (both of the Ca^{2+} oscillation (blue line) and the kinetics of the IP_3 oscillation (dashed yellow line)) will give a stable level of the desired Ca^{2+} concentration.

Fig 3 shows how the IP_3 can affect the intracellular Ca^{2+} signalling. An increase of IP_3 in the system is desired for regular Ca^{2+} concentration levels. Since β is responsible for the IP_3 increase, it is highly important for regulation of Ca^{2+} concentration levels. As soon as IP_3 becomes constant, the Ca^{2+} concentration will drop. The electrical component of the astrocytes also plays an important role (E_0 from Eq. 3). However, since the synapses happen periodically, we are interested in the design of control within one period when the synapses are activated.

The IP_3 is then a decisive factor for Ca^{2+} regulation, in which its increase is controlled by β. Such behaviour is going to be further explored with a mathematical model that enables the intracellular Ca^{2+} signalling control by regulating IP_3 levels.

IV. Feed-forward and Feedback Control of Intracellular Ca^{2+} Signalling in Astrocytes

Before proceeding further, we assume for the rest of the paper that $\mathcal{H}(E_0 - 35) = 1$. Note that for other values of the Heaviside function (such as $\mathcal{H}(E_0 - 35) = \frac{1}{2}$), a similar analysis can be applied. By denoting the column vector state $\mathbf{x} = [C\ h\ I]'$, we can rewrite (1), (2), (3) as the following nonlienar controlled state space system

$$\frac{d\mathbf{x}}{dt} = f(\mathbf{x}) + \mathbf{B}u \tag{11}$$

where $f(\mathbf{x}) = [f_1(\mathbf{x})\ f_2(\mathbf{x})\ f_3(\mathbf{x})]'$, and $f_1(.) = \sigma_1 + \sigma_2 + \sigma_3$, $f_2(.) = \frac{H-h}{\tau}$, and $f_3(.) = \frac{1}{\alpha}(i_0 - I)$, and $\mathbf{B} = [0\ 0\ 1]'$. The control variable u represents the state feedback and feedforward based IP_3 regulation parameter given by (see also [9]):

$$\beta = \beta_f - K_f(C - C_f), \tag{12}$$

where β_f is the feedforward control representing the desired IP_3 level I_f, C_f is the associated desired Ca^{2+} concentration level and K_f is the linear feedback gain. Note that although not visible in the above equation, there is an associated value of h as well, which is denoted by h_f. Denote the entire associated state vector as $\mathbf{x_f} = [C_f\ h_f\ I_f]'$. Then it follows that

$$\frac{d\mathbf{x_f}}{dt} = f(\mathbf{x_f}) + \mathbf{B}u_f \tag{13}$$

where of course, $u_f = \beta_f$. Here, the measured output is the Ca^{2+} concentration level, so that at the desired level, the output is given by $\bar{h}(\mathbf{x_f}) = C_f$.

Remark 1: Note that *without loss of generality*, one can assume that in the uncontrolled case, i.e., when $\beta = 0$, there is an equilibrium point at the origin for the nonlinear dynamical system $\frac{d\mathbf{x}}{dt} = f(\mathbf{x})$ [9], implying $f(\mathbf{0}) = 0$. This fact will be used in the stability analysis of the system when the control law (12) is applied in a subsequent subsection.

A. β_f and C_f Relationship

To obtain the required regulation factor β_f for a desired C_f, this paper presents a mathematical relationship between β_f and C_f as follows. Suppose there is an equilibrium point (in the controlled case when $\beta > 0$) at $(h^o\ c^o\ I_0)$, then $\frac{dh}{dt} = 0$ at $h = h^o$ and $\frac{dC}{dt} = 0$ at $C = c^o$, and $\frac{dI}{dt} = 0$ for $I = I_0$. Rewriting Eq. 1 and Eq. 2 we can compute I_o, from for Eq. 3, (since $\mathcal{H}(E_0 - 35) = 1$). As $t \to \infty$, I becomes a constant I_o, which is represented as $I_o = (i_0 + \alpha\beta)$. This relationship enables the computation of the control function (12).

TABLE I: Simulation parameters for astrocytes.

Variable	Value
v	$6s^{-1}$
v_1	0.11 s^{-1}
c_0	$2.0\mu M$
C_1	0.185
v_2	0.9 Ms^{-1}
k	$0.1\mu M$
d	$0.13\mu M$
d_1	1.049 s^{-1}
d_2	$0.9434\mu M/s$
d_3	$0.08234\mu M$
a	0.2 s^{-1}
α	$1/0.00014\mu M$
i_0	$0.160\mu M$
β	0.1-1.5μM
E_0	35

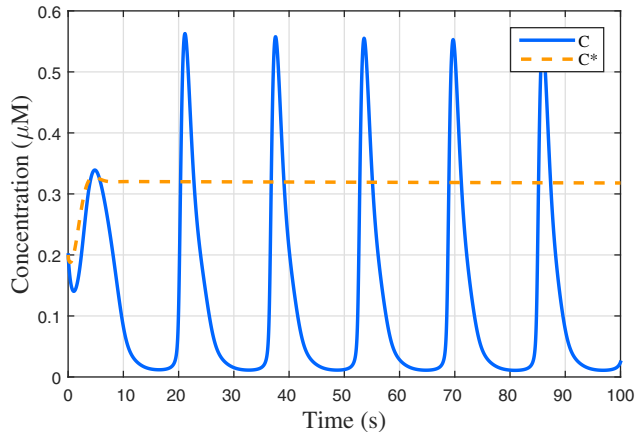

Fig. 4: Elimination of Ca^{2+} oscillation using the proposed feedback and feed-forward control technique. Regular Ca^{2+} oscillations C (solid lines) is compared to a controlled Ca^{2+} level C^* (dashed line) for $IP_3 = 0.5$ (μM).

V. Analysis

We now present an analysis of the proposed regulation of Ca^{2+} concentration levels for astrocytes. This is divided into four parts for a proper understanding of the system and also quantification of the application impact if this control technique is utilised. First, we start by showing how the control system will eliminate intracellular Ca^{2+} oscillations in astrocytes, solving the problem defined in Section III. This is followed by the disturbance analysis, where Gaussian noise is applied to the intracellular Ca^{2+} signalling for adding a controlled abnormal behaviour to the system and observing system effectiveness while looking at the feed-forward and feedback techniques separately. Finally, analyses of the envisioned applications of disease prevention are shown, which consist of three scenarios (i) elimination of oscillatory behaviour (ii) robustness over frequency modulation (iii) robustness over post-synaptic influence.

A total elimination of the Ca^{2+} oscillation is obtained using the proposed mechanism, and this is illustrated in Fig 4. The Eq. 12, which represents the state feedback and feed-forward control, can efficiently adjust β accordingly and maintain Ca^{2+} concentration levels throughout the period shown, by solving Eqs. 1, 2 and 3. For the control technique, we replace the β in Eq. 3 by Eq. 12, in order to integrate the feed-forward and feedback control element to the system. We chose a value of $C_f = 0.32$ μM, which is a mean value of the system and compute the desired β_f with an appropriate calibration of the K_f. This positive result demonstrates the effectiveness and potential of utilising the control technique to stabilise the excessive Ca^{2+} concentration that may lead to neurodegenerative diseases.

Two other scenarios were investigated with the objective of analysing different Ca^{2+} signal patterns. First, frequency modulation can be achieved by *saddle-node homoclinic bifurcation* [11], and implemented also in the model by changing the IP_3 concentration to higher levels, Fig 5. Second, astrocytes also receive membrane voltage signals from post-synaptic neurons that change the Ca^{2+} behaviour [12], Fig 6. These signals are also simulated abstractly mimicking the same behaviour in this scenario. *The control mechanism shows the high level of robustness for presenting the same performance even with different signal patterns of Ca^{2+}.* This shows the importance IP_3 levels in the Ca^{2+} intracellular signalling, and how controlling such protein is impacting in also controlling the Ca^{2+} behaviour.

VI. Methodologies for implementation *in-vitro* and *in-vivo*

The theoretical approach used in this paper is not directly linked to a particular implementation technique in systems biology. In this section, we present a discussion on different possibilities for implementing the proposed control system in both *in-vitro* and *in-vivo* settings.

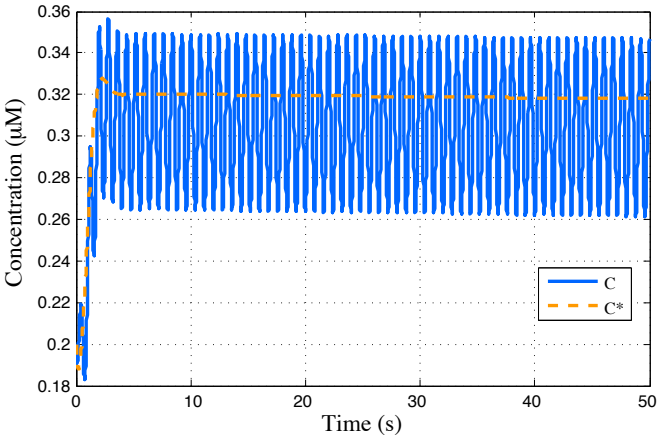

Fig. 5: Elimination of Ca^{2+} oscillation using the proposed feedback and feed-forward control technique now with frequency modulation by IP_3 increase. Regular Ca^{2+} oscillations C (solid lines) is compared to a controlled Ca^{2+} level C^* (dashed line) for $IP_3 = 1.0$ (μM).

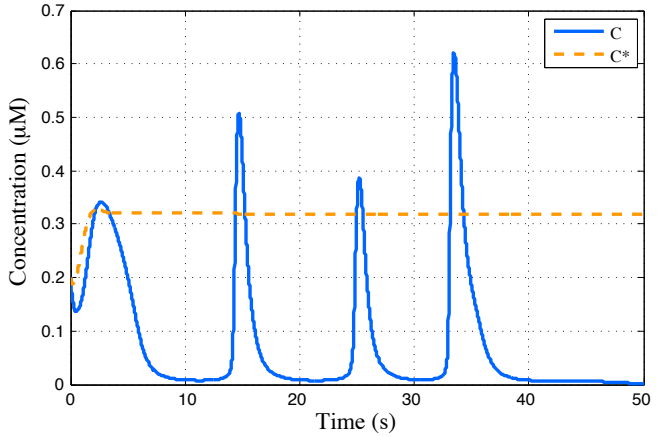

Fig. 6: Elimination of Ca^{2+} oscillation using the proposed feedback and feed-forward control technique now with influence from a post-synaptic neuron. Regular Ca^{2+} oscillations C (solid lines) is compared to a controlled Ca^{2+} level C^* (dashed line) for $IP_3 = 0.5$ (μM).

A. Nanoparticles

Nanoparticles have been already used in systems biology for predicting toxicity at the cellular level by measuring the modulation changes in intracellular Ca^{2+} signals. The mere presence of toxicity does not inhibit their applicability to biology due to existing techniques of decreasing toxicity and also the numerous potential benefits of their usage. Nanoparticles are known to bypass existing biological protection systems of living organisms to perform desired tasks including, attachment to cancerous cells, bypassing the blood-brain barrier, etc. Control of Ca^{2+} can be also be achieved with nanoparticles, which include: Carbon Black (CB), Titanium Dioxide ($TiO2$) or Zinc Oxide (ZnO) [13]. These nanoparticles are known to stimulate the IP_3 channels and influence the amplification of Ca^{2+} signals through the endoplasmic reticulum. However, this technique is also known for stressing cellular organelles and therefore causing cell death over time. Researchers are concentrating their efforts in determining which stress signalling pathways are induced downstream of nanoparticle exposure.

B. Synthetic Biology

Synthetic biology is paving the way for advancements in systems biology by allowing programmability of living organisms and extending their functionalities based on a particular application [14]. Synthetic biology can also program cellular signalling pathways with a particular gene expression that can change how molecules interact inside a cell. This can be considered a building block for advanced systems, in which the signalling pathways can include basic logic operation with an integration of logic circuits, that takes into account molecular levels of the cell to perform computation tasks. Controlling Ca^{2+} signalling pathways is another alternative solution towards implementing the presented control function. The gene expression can be changed to make the IP_3 concentration change adaptively based on the Ca^{2+} concentration through independent excitatory and inhibitory pathways. In this way, both very high and very low level of Ca^{2+} can be re-arranged to normal homoeostasis levels. The primary challenge is to perform this task distributively and with precision in particular cells. For this, targeted gene therapy has recently gained more attention due to the recent results, that also include regeneration of cellular functions in the brain, see for example [15].

C. Implantable Devices

Advancements in nanotechnology are enabling nanoscale devices with biocompatible materials to be implanted in numerous locations in *in-vivo* organisms with the challenge of regulating cellular activity. These devices use photostimulation to activate distinct pathways in synthetic cells, namely neuron dust [16]. The main advantage of such technology is the ability of accurately maintaining time-sensitive channels at stable levels, and in this case, the control of post-synaptic neurons by neuron dust devices would contribute to the regulation of Ca^{2+} signalling in astrocytes. The major drawback is the necessity of enabling cells to be photosensitive, which is a current research topic for neuron rehabilitation techniques combining photostimulation with targeted gene therapy solutions.

D. Molecular Communications and Nanonetworks

Molecular communications is a recent research area responsible for designing communication systems to actuate *in-vivo* but at the same time trying to not affect the existing biological processes [4], [17]. These tasks can be very challenging due to the usage of few biological channels to send information, and therefore, limiting the molecular communications system performance based on this high correlation with biological systems regulation functions. Addressing this drawback is a current research theme in the community, and researchers have suggested the use of silence communication techniques [18]. But this property can also be used to influence cellular signalling pathways in various degrees. A nanonetwork can be employed for communicating cells through molecular communications channels, with the receiving end being a targeted cell. This mechanism has been used to predict cellular properties in [19], [20], where the process is inverted to regulate the underlying Ca^{2+} molecular communication system. In this particular case, the receiver cell will have specific stimuli to start the regulation of intracellular signalling. The major challenge is to accurately send information over tissues due to the high influence of noise in biological communication channels [21].

VII. Conclusion

New approaches for possibly treating neurodegenerative diseases are appealing to address the low efficiency

of current methods. With the advancements in nanotechnology, now one can think of also controlling Ca^{2+} signalling in astrocytes for regulating gliotransmitters' concentration and indirectly control synaptic quality. A theoretical control theory approach is presented in this paper, where cellular state information such as Ca^{2+} concentration and IP_3 concentration are used in a control function that measures the proper amount needed for ultimately regulating Ca^{2+} at the desired level. These results lead to an exploratory discussion surrounding the future possibilities in implementing such approach, including nanoparticles, synthetic biology, implantable devices and molecular communications. Further research work needs to be carried out in both stability and disturbance analysis of this system, and also regarding how to deal with noise in intracellular environments.

Acknowledgement

The work described in this paper is facilitated by the Irish Research Council, under the government of Ireland post-doc fellowship (grant GOIPD/2016/650).

References

[1] W. W. Seeley, R. K. Crawford, J. Zhou, B. L. Miller, and M. D. Greicius, "Neurodegenerative diseases target large-scale human brain networks," *Neuron*, vol. 62, no. 1, pp. 42–52, 2009.

[2] J. Olesen and M. Leonardi, "The burden of brain diseases in europe," *European Journal of Neurology*, vol. 10, no. 5, pp. 471–477, 2003.

[3] C. J. Murray, T. Vos, R. Lozano, M. Naghavi, A. D. Flaxman, C. Michaud, M. Ezzati, K. Shibuya, J. A. Salomon, S. Abdalla *et al.*, "Disability-adjusted life years (dalys) for 291 diseases and injuries in 21 regions, 1990–2010: a systematic analysis for the global burden of disease study 2010," *The lancet*, vol. 380, no. 9859, pp. 2197–2223, 2013.

[4] M. T. Barros, "Ca 2+-signaling-based molecular communication systems: Design and future research directions," *Nano Communication Networks*, 2017.

[5] S. Sahoo, S. Parveen, and J. Panda, "The present and future of nanotechnology in human health care," *Nanomedicine: Nanotechnology, Biology and Medicine*, vol. 3, no. 1, pp. 20–31, 2007.

[6] A. Araque and M. Navarrete, "Glial cells in neuronal network function," *Philosophical Transactions of the Royal Society B: Biological Sciences*, vol. 365, no. 1551, pp. 2375–2381, 2010.

[7] G. Perea, M. Navarrete, and A. Araque, "Tripartite synapses: astrocytes process and control synaptic information," *Trends in Neurosciences*, vol. 32, no. 8, pp. 421 – 431, 2009.

[8] R. Antonelli, J. Harmand, J.-P. Steyer, and A. Astolfi, "Set-point regulation of an anaerobic digestion process with bounded output feedback," *IEEE Transactions on Control Systems Technology*, vol. 11, no. 4, pp. 495–504, July 2003.

[9] P. Raul, S. Manyam, P. Pagilla, and S. Darbha, "Output regulation of nonlinear systems with application to roll-to-roll manufacturing systems," *IEEE/ASME Transactions on Mechatronics*, vol. 20, no. 3, June 2015.

[10] M. Pitta, M. Goldberg, V. Volman, H. Berry, and E. Ben-Jacob, "Glutamate regulation of calcium and ip3 oscillating and pulsating dynamics in astrocytes," *Journal of Biological Physics*, vol. 35, pp. 383–411, 2009.

[11] M. De Pittà, M. Goldberg, V. Volman, H. Berry, and E. Ben-Jacob, "Glutamate regulation of calcium and ip3 oscillating and pulsating dynamics in astrocytes," *Journal of biological physics*, vol. 35, no. 4, pp. 383–411, 2009.

[12] S. Nadkarni and P. Jung, "Modeling synaptic transmission of the tripartite synapse," *Physical biology*, vol. 4, no. 1, p. 1, 2007.

[13] S. Hussain, L. C. J. Thomassen, I. Ferecatu, M. Borot, K. Andreau, J. A. Martens, J. Fleury, A. Baeza-Squiban, F. Marano, and S. Boland, "Carbon black and titanium dioxide nanoparticles elicit distinct apoptotic pathways in bronchial epithelial cells," *Particle and Fibre Toxicology*, vol. 7, no. 10, pp. 1–17, 2010.

[14] J. Hansen and Y. Benenson, "Synthetic biology of cell signaling," *Natural Computing*, vol. 15, no. 1, pp. 5–13, 2016.

[15] P. R. di Val Cervo, R. A. Romanov, G. Spigolon, D. Masini, E. Martín-Montañez, E. M. Toledo, G. La Manno, M. Feyder, C. Pifl, Y.-H. Ng *et al.*, "Induction of functional dopamine neurons from human astrocytes in vitro and mouse astrocytes in a parkinson's disease model," *Nature Biotechnology*, 2017.

[16] D. Seo, J. M. Carmena, J. M. Rabaey, E. Alon, and M. M. Maharbiz, "Neural dust: An ultrasonic, low power solution for chronic brain-machine interfaces," *arXiv preprint arXiv:1307.2196*, 2013.

[17] M. T. Barros, S. Balasubramaniam, and B. Jennings, "Comparative end-to-end analysis of ca2+ signaling-based molecular communication in biological tissues," *IEEE Transactions on Communications*, vol. PP, no. 99, pp. 1–1, 2015.

[18] M. T. Barros, S. Balasubramaniam, B. Jennings, and Y. Koucheryavy, "Transmission protocols for calcium signaling based molecular communications in deformable cellular tissues," *IEEE Transactions on Nanotechnology*, vol. 13, no. 4, pp. 779–788, 2014.

[19] M. T. Barros, S. Balasubramaniam, and B. Jennings, "Using information metrics and molecular communication to detect cellular tissue deformation," *IEEE Transactions on Nanobioscience*, vol. 13, no. 3, pp. 278–288, 2014.

[20] ——, "Adaptive transmission protocol for molecular communications in cellular tissues," in *The IEEE Conference on Communications*, 2014.

[21] M. T. Barros, S. Balasubramaniam, and B. Jenningsú, "Error control for calcium signaling based molecular communications," in *47th Annual Asilomar Conference on Signals, Systems, and Computers*, 2013.