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# Neuronal cultures playing Pong: First steps toward advanced screening and biological computing

Lena Smirnova[1] and Thomas Hartung[1,*]
[1]Johns Hopkins Bloomberg School of Public Health, Environmental Health and Engineering, Center for Alternatives to Animal Testing, Baltimore, MD, USA
*Correspondence: thartun1@jhu.edu
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**In this issue of *Neuron*, Kagan et al.[1] implement learning-in-a-dish as an important step toward organoid intelligence. These systems may complement the study of molecular and cellular mechanisms of cognition and allow innovations in pharmacological and toxicological studies of neurodevelopmental or neurodegenerative disorders as well as advances in biological computing.**

The advent of human stem cell technologies and bioengineering has enabled the realization of aspects of organ architecture and functionality, a.k.a. microphysiological systems (MPS).[2] The field of MPS is increasingly maturing with this year's creation of a World Summit series (https://mpsworldsummit.com/) and an International MPS Society (https://impss.org/). In the case of the brain, the ultimate functionality is cognition. With an elegant implementation, Kagan et al.[1] demonstrate aspects of learning by training stem-cell-derived human neuronal cultures to learn to play the simple computer game PONG. The scientists used the established feedback loop approach. Penalizing the culture with noise (uninterpretable input) whenever the controlled paddle misses the ball proved sufficient to improve game performance. Extending beyond this, well-controlled follow-up experiments demonstrated that the type of feedback applied seems related to the apparent learning effects. Likewise, numerous electrophysiological measurements, including nuanced metrics such as functional plasticity and information entropy, were found to accord with these findings. Beyond these thought-provoking findings, the paper suggests several directions for future research. Notably, no longer-term memory was achieved—the culture started any training session from start. As a next step, an increased complexity of the cellular model could be achieved, for example, by including supporting glia (astrocytes, oligodendrocytes, and microglia) and potentially enabling cascades of molecular and cellular events, leading to long-term potentiation and depression and, as a result, long-term memory.

The work expands on earlier studies, e.g., Shahaf and Maron[3,4] reported that cultures of rat primary cortical neurons demonstrated learning as the desired predefined response to low-frequency focal stimuli; they showed that after a learning curve, the desired response in the form of distinct electrophysiological patterns immediately followed the stimulus. The Potter group[5,6] used rat primary cultures and trained them to control a small moving device. Kagan et al.[1] replicated these results with both mouse primary and human iPSC-derived neuronal cultures, employing high-density microelectrode arrays and demonstrating training effects in a computer game environment. The new study by Kagan and collaborators illustrates how advances in stem cell culture and sensor technology, as well as increased computational power, bring the approach to the next level. However, their use of traditional 2D neuronal cultures does not yet leverage the opportunities of 3D cultures and the homeostatic culture conditions that can be achieved by organs-on-chip systems.

Kagan and co-authors referred to the system as "sentient." The claim of sentience of the model is bold even within the specific definition provided in this work, but so is the endeavor of synthetic biological intelligence. The terms of cognition or intelligence-in-a-dish refer to very basic elements of these high-level functions, i.e., the ability to perform simple computer functions, not human-level cognition and intelligence. Learning-in-a-dish is an ability to process an input and provide a measurable output, as a learned adequate response to the stimuli enabled by the presence of the necessary molecular machinery and physiological features such as learning circuits of long-term memory. The use of human cells necessarily prompts concerns about moving toward consciousness, as has been voiced in the context of brain organoid development.[7,8] While the absence of long-term memory here alleviates such concerns, the ongoing progress in brain organoids with the prerequisites for memory brings such developments into reach. Notably, coining the term organoid intelligence (OI), a community is forming to leverage this potential. We had the privilege to host an OI Community-forming workshop with 50 eminent scientists earlier this year at Johns Hopkins and the respective proceedings and a *Baltimore Declaration toward OI* shall be published in early 2023. Notably, the Cortical Lab group, who published their work in this issue, was part of this process. The OI community is very aware of the possible ethical challenges and follows a concept of embedded ethics,[9] in which ethicists and developers together address ethical issues via an iterative and continuous process from the outset of development. Studies surveying public perception of such technologies are on the way in order to understand levels of concern.

The possible gain of implementing learning-in-a-dish (or other cognitive

functions) is enormous. First, reductionistic experimental models of human cognitive functions could allow the experimental exploration and verification of concepts of the underlying physiology on molecular and cellular levels. Second, such test systems could allow tackling neurodevelopmental and neurodegenerative diseases in human models and perturbed cognition as a functional readout—with respect to both the identification of genetic risk factors and contributing exposures on the one hand and possible interventions such as drugs on the other hand. This could enhance cell models in the context of diseases with cognitive impairment such as autism or Alzheimer's disease, where patient's stem cells can be used to generate neural cell cultures or organoids. We see a potential to use such a system to address not only individual differences in learning and memory capacities but also how to booster these cognitive functions. For example, we might use such a system to test or develop drugs that improve learning and memory.

Finally, this type of technology—as exemplified by Kagan and colleagues—might open up an entire avenue for actual biological computing. It is certainly pie-in-the-sky at this moment but an attractive suggestion, considering the energy efficiency of the brain. The human brain is still largely unmatched as a computational asset: the most advanced supercomputer, the Hewlett Packard Enterprise Frontier, or OLCF-5, at Oak Ridge National Laboratory and US Department of Energy in Tennessee, in June of 2022 just surpassed the estimated computing power of a single human brain of one exaFLOPS. It is tempting to compare the effort of $600 million, installation on 680 $m^2$ (7,300 sq. ft) and energy consumption of 21 MW with a human brain of 1.4 kg and 20 W power need. The computer industry is already embracing the dreams of biological computing: "Biology will undoubtedly fuel computing," Eric Schmidt, former Google CEO, stated at SynBioBeta October 2019. He expanded: "Taking biology, which I'd always viewed as squishy and analog, and turning it into something that can be digitally manipulated, is an enormous accelerator. … There's lot of evidence that biology is in that golden period right now." Kagan et al. have helped us bring this dream a little closer to reality.

## DECLARATION OF INTERESTS

T.H. is named inventor on a patent by Johns Hopkins University on the production of mini-brains (also called BrainSpheres), which is licensed to AxoSim, New Orleans, Louisiana, USA. T.H. and L.S. are consultants for AxoSim, New Orleans, and T.H. is also a consultant for AstraZeneca and American Type Culture Collection (ATCC) on advanced cell culture methods.

## REFERENCES

1. Kagan, B.J., Kitchen, A.C., Tran, N.T., Habibollahi, F., Khajehnejad, M., Parker, B.J., Bhat, A., Rollo, B., Razi, A., and Friston, K.J. (2022). In vitro neurons learn and exhibit sentience when embodied in a simulated game-world. Neuron *110*, 3952–3969. https://doi.org/10.1016/j.neuron.2022.09.001.

2. Roth, A.; MPS-WS Berlin 2019 (2021). Human microphysiological systems for drug development. Science *373*, 1304–1306. https://doi.org/10.1126/science.abc3734.

3. Shahaf, G., and Marom, S. (2001). Learning in networks of cortical neurons. J. Neurosci. *21*, 8782–8788. https://doi.org/10.1523/jneurosci.21-22-08782.2001.

4. Marom, S., and Shahaf, G. (2002). Development, learning and memory in large random networks of cortical neurons: lessons beyond anatomy. Q. Rev. Biophys. *35*, 63–87. https://doi.org/10.1017/S0033583501003742.

5. Demarse, T.B., Wagenaar, D.A., Blau, A.W., and Potter, S.M. (2001). The Neurally Controlled Animat: Biological Brains Acting with Simulated Bodies. Auton. Robots *11*, 305–310. https://doi.org/10.1023/a:1012407611130.

6. Bakkum, D.J., Chao, Z.C., and Potter, S.M. (2008). Spatio-temporal electrical stimuli shape behavior of an embodied cortical network in a goal-directed learning task. J. Neural. Eng. *5*, 310–323. https://doi.org/10.1088/1741-2560/5/3/004.

7. Buchanan, M. (2018). Organoids of intelligence. Nat. Phys. *14*, 634. https://doi.org/10.1038/s41567-018-0200-2.

8. Reardon, S. (2020). Can lab-grown brains become conscious? Nature *586*, 658–661. https://doi.org/10.1038/d41586-020-02986-y.

9. McLennan, S., Fiske, A., Tigard, D., Müller, R., Haddadin, S., and Buyx, A. (2022). Embedded ethics: a proposal for integrating ethics into the development of medical AI. BMC Med. Ethics *23*, 6. https://doi.org/10.1186/s12910-022-00746-3.