Case study

Advancing cardiac research: 32 wells at a time

Tools:
SyncroPatch 384
32-well mode

Professor Niels Voigt and Dr. Fitzwilliam Seibertz at UMG featured by Nanion Technologies

Professor Niels Voigt is Professor of Molecular Pharmacology at the University Medical Center Göttingen. His research focuses on investigating the pathophysiology of cardiac arrhythmias. Using the SyncroPatch 384, Professor Voigt's lab has used a variety of cells including stem-cell derived atrial and ventricular cardiomyocytes and adult primary cardiomyocytes.

The study of cellular electrophysiology involves in-depth functional characterization of ion channels and transporters bound within cellular membranes. In excitable cells, such as cardiomyocytes and neurons, physiological ion channel function is crucial for the formation of action potentials, which serve as an elegant means of organized electrical signal propagation across cardiac muscle or throughout the brain. Encoded by specific genes, ion channels are susceptible to mutations which can alter their protein structure and function. On a system level, this may translate into severe medical conditions such as cardiac arrhythmias or neurological disorders. It is therefore of vital importance to study the functional abnormalities of these channels and the resultant modifications of action potential morphology at a cellular level. Understanding the underlying functional pathologies then allow medical researchers to identify molecular drug targets that could be utilized for pharmacological treatment and clinical management of these diseases.

In order to accurately understand the impact of ion channel mutations on human organ function, cellular models should ideally be employed that accurately represent the structure and function of endogenous organ-specific cells. In contrast to heterologous expression systems which may not completely reflect cardiomyocyte or neuron physiology, primary cells from humans or large animal models are desirable. In addition, induced pluripotent stem cell (iPSC) technology has emerged over the last decade to provide wider access to cardiac and neuronal material directly derived from human patients.

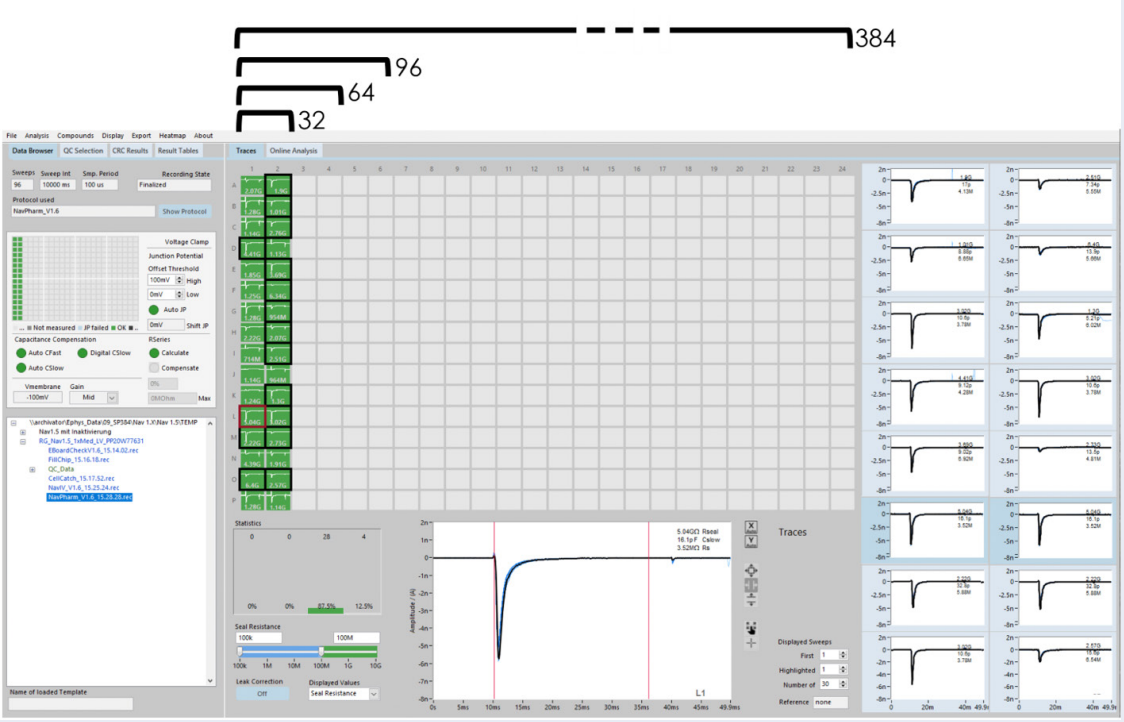
Traditional methods for the acquisition of ion currents, action potentials and analysis of ion channel biophysics involve the patch-clamp technique. This approach is complex, requires extensive user expertise and is highly laborious. It is, by definition, a low-throughput method. New developments in automated patch-clamp (APC) instrumentation have removed this limitation, with high throughput devices such as the new SyncroPatch 384 which can measure up to 384 cells simultaneously during single experiment. This is achieved by depositing cells in suspension via robotic liquid handler across a precision engineered 384-well measurement chip.

Nanion's SyncroPatch 384 platform *is highly flexible, making it ideal for academic research labs. The 32-well mode optimizes assay development and maximizes NPC-384 economy. Alternatively, the SyncroPatch 384 can be run in fully automated, unattended mode making it both a high and medium throughput instrument in one.*

> **"Together with Nanion's dedicated researchers, we performed novel experiments on primary cardiomyocytes using the SyncroPatch. These experiments laid the foundation for our exciting future collaboration with Nanion and multiple new projects."**
>
> **Professor Niels Voigt**, Principle Investigator, UMG

Originally designed for drug discovery, APC systems are amenable to a huge range of measurements from different cellular substrates. Such scalability in the methodological domain has been invaluable, however sometimes may surpass the requirements of those in academia working with highly limited and valuable cellular models. In order to introduce more flexibility and cost economy into the academic workflow of high throughput measurements, the SyncroPatch 384 offers a reduced means of measurement, the 32-well mode. This new feature allows the experimenter to use only a fraction of the chip per experiment through customizable constellations of wells in multiples of 32. Afterwards, the chip can then be stored and used again at later time points with no impact on future measurement quality or success rate.

Newly launched in 2021, the SyncroPatch 384 equipped with 32-well mode has been invaluable in recent studies from basic science research groups such as the group of Professor Niels Voigt at the University of Göttingen. His lab primarily aims to unravel the molecular mechanisms that contribute to atrial fibrillation and other cardiac arrhythmias in human patients. The 32-well mode played a pivotal role in the successful first-time measurements of primary cardiomyocytes using APC[1]. Here, cost-effective measurements of action potentials and ionic currents were successful even when cellular isolation yielded very low numbers of viable cellular material (~7000 cells per 5 ml). For these sparsely populated isolates, it was vital to concentrate cellular numbers with low solution volumes. The 32-well mode of operation accommodated this requirement with ease while still delivering high throughput results. The 32-well



The SyncroPatch can be used in 32-well mode for assay development, smaller screening projects and where the number of cells is limited. *The amplifier and pipettes can be used in columns of 32-wells at a time and the rest of the chip can be used at a later time point without any reduction in success rate.*

mode also opens the door for direct assessment of human cardiomyocyte electrophysiology from small biopsies that can be acquired from human patients undergoing open heart surgery. Operating under reduced tissue requirements is also particularly important for decreasing the amount of animals needed for scientific use. In the aforementioned study, the 32-well mode allowed for a massive amount of functional data to be collected from very few animals, indicating that APC paired with this approach may represent a desirable step towards reducing the number of animals sacrificed for disease modeling purposes.

Advents such as induced pluripotent stem cell technology also acknowledge the ethical responsibility of reducing animal use by providing a theoretically infinite amount of patient specific cellular material. iPSC-derived cells pair very well with APC and a number of modeling, screening and methodological studies have been released over recent years[1–6]. However, it is clear that regular cell differentiation and maintenance of iPSC lines is an expensive exercise requiring large amounts of reagents and personnel. The 32-well mode can be used to screen a limited number of precious cells based on their availability, or as a means to test and optimize voltage protocols and solution components without wasting cellular material, reagents and consumables. A recent study from the Voigt lab made use of the 32-well mode to optimize a novel protocol to measure I_{Kr} in iPSC-derived cardiomyocytes using equimolar caesium concentrations[7]. The findings of this study provide deep insights into the maturation of distinct ion currents in iPSC-derived cardiomyocytes over long-term culture to address their often controversial applicability and usefulness as a model of human cardiomyocyte function.

In a recent symposium (Nanion Cardiac Safety Symposium 2023), Fitzwilliam Seibertz described his use of the 32-well mode as an alternative screening platform for a rapid determination of batch-based purity of iPSC constructs. iPSC-derived cardiomyocytes often display heterogeneous subtype specificity ranging from nodal, to atrial or ventricular characteristics. In the study of cardiac chamber-specific diseases such as atrial fibrillation, it is necessary to verify the cells under investigation are of the desired subtype. Following the deployment of a depolarizing ramp protocol to measure inward rectifier activity, compounds such as the M2 receptor agonist carbachol can be applied

Prof. Niels Voigt (left), Aistė Liutkutė, Dr. Fitzwilliam Seibertz and Dr. Funsho Fakuade with the SyncroPatch 384 at the UMG. *The SyncroPatch 384 is routinely used in the lab to record hiPSC-derived cardiomyocytes to investigate the physiology and pathophysiology of cardiac arrhythmias.*

"If I'm experimenting with new protocols on cardiomyocytes, the 32 well mode really puts my mind at ease knowing that I can still optimise my recordings and save most cells for later when everything is ready to go."

Dr. Fitzwilliam Seibertz, Postdoctoral Scientist, UMG

to unmask acetylcholine-activated inward rectifiers that are only present in atrial-specific cells[8]. 32-well mode can be used to easily and economically test each batch "on the side" after crucial measurements to avoid the often difficult and time consuming molecular techniques to test for atrial or ventricular-specific genes.

APC systems such as the SyncroPatch 384 offer insights into cellular electrophysiology at an unprecedented scale. In the domain of pharmacological screening and drug discovery, high throughput electrophysiology using APC has proven invaluable over the last two decades. As these techniques traverse into academia, where constant funding often remains an obstacle, the 32-well mode of the SyncroPatch 384 offers a highly flexible and economic modus operandi, reducing the cost per data point to 10% of lower throughput APC instruments. A smooth gear change from high-to-medium throughput (or vice versa) on the same device ensures that costs and valuable cell material are well conserved, allowing for exciting future disease modeling studies and drug development initiatives.

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