

# A New Era is Emerging for Ion Channel and Channelopathy Research

Tools:  
SyncroPatch 384

Alfred L. George, Jr., MD  
featured by Nanion Technologies



Alfred L. George, Jr., MD is the Magerstadt Professor and Chair of the Department of Pharmacology and Director of the Center for Pharmacogenomics at Northwestern University Feinberg School of Medicine. His work has contributed greatly to understanding the mechanisms by which ion channel mutations cause a variety of inherited disorders of membrane excitability.

The channelopathy field is an exciting branch of ion channel research that combines human genetics, cellular electrophysiology, and pharmacology. The main goals of this field are to elucidate the molecular mechanisms of diseases caused by ion channel dysfunction, and to develop more personalized treatment strategies based on an individual's DNA sequence.

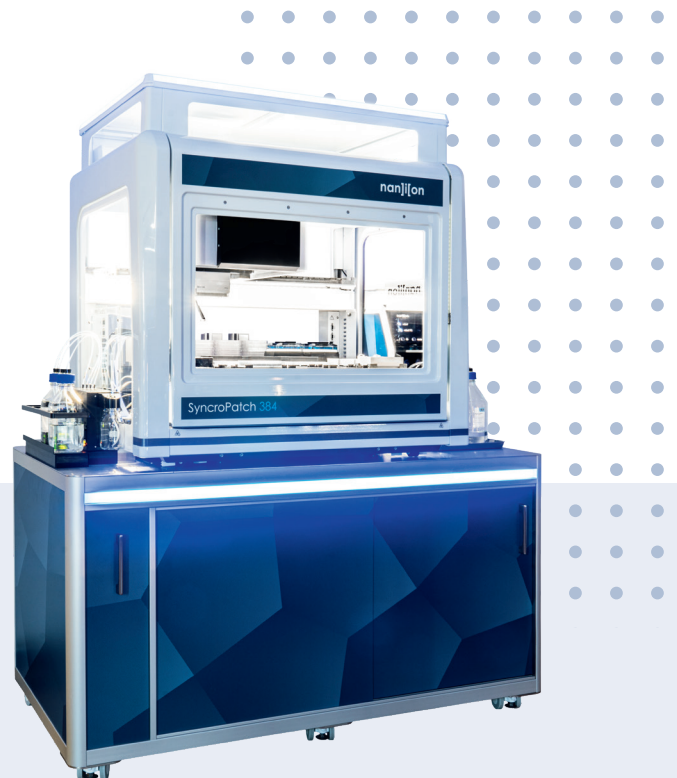
Ion channels are encoded by specific genes that exhibit nucleotide and amino acid sequence variation among individuals in the general population. In rare cases, specific mutations cause altered function or impaired trafficking of an ion channel protein. Channelopathy is the term used to describe those disorders associated with mutations in ion channel genes. In recent years, there has been accelerated effort to sequence genes and genomes in persons with all forms of genetic diseases as well as healthy people and this has led to an explosion in the number of genetic variants discovered. However, the interpretation of findings from genetic studies can be difficult when variants are classified by the designation 'variant of unknown significance' (VUS), which

is often the case when insufficient data exist to distinguish benign from pathogenic changes. This distinction is critically important in clinical medicine for making a proper diagnosis, planning appropriate genetic and reproductive counseling, and in some cases for devising a clinical management plan.

A widely used approach for classifying genetic variants enables inclusion of functional evidence demonstrating a damaging effect of a variant into the interpretation. Unfortunately for most genes, functional evidence to support or refute pathogenicity is challenging to obtain. Fortunately, this is not the case for genes encoding ion channels, which can be assayed with well-established experimental paradigms and methodologies. The gold standard approach for assessing functional effects of ion channel variants is patch clamp electro-physiology. But, this approach is tedious, time- and labor-intensive, and not scalable to match the rampant discovery of new ion channel variants.

## Nanion's SyncroPatch 384 platform

(successor of SyncroPatch 384PE and SyncroPatch 384i) allows effortless high-throughput ion channel screening coupled with high flexibility and reliability, due to the combination of its patch clamp module with Beckman Coulter's new i-Series liquid handling robot Biomek i5.

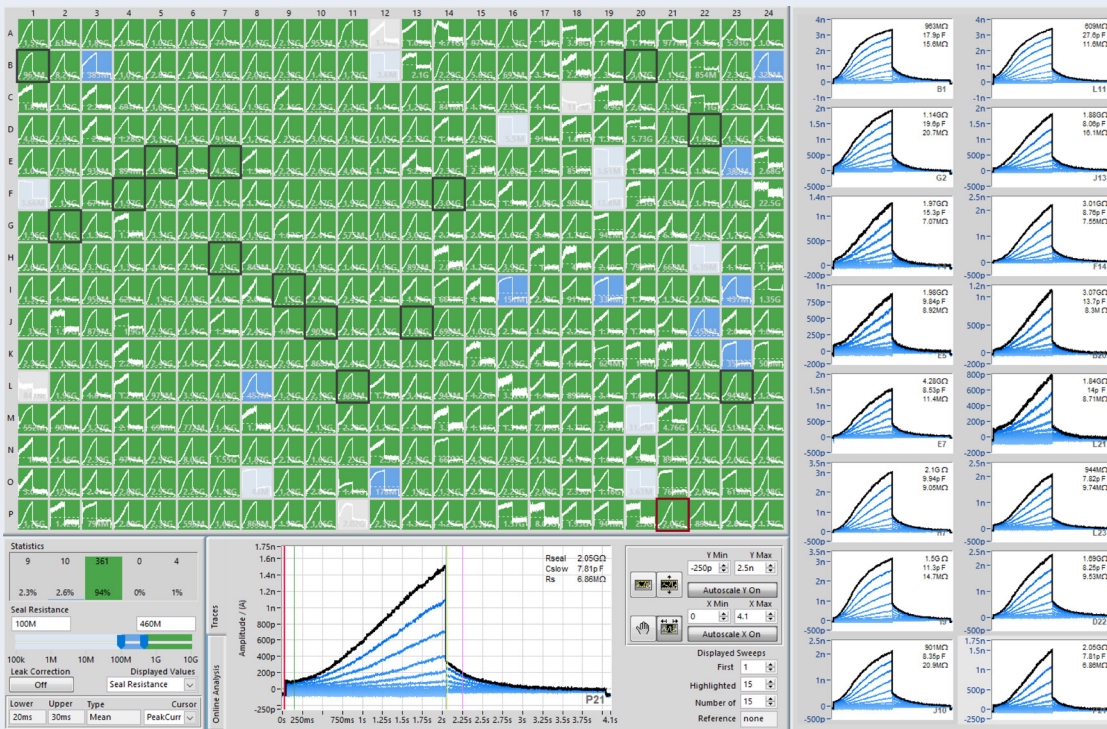


**“The SyncroPatch has revolutionized our ability to determine the functional consequences of hundreds of human ion channel variants, which could be considered one of the most significant recent advances in channelopathy research.”**

**Alfred L. George, Jr., MD,** Northwestern University Feinberg School of Medicine

In the past few years, an emerging group of pioneering researchers have utilized high throughput automated patch clamp instrumentation, originally designed for drug discovery, to study ion channel variants at an unprecedented scale. The SyncroPatch 384PE system, has emerged as the automated patch clamp system of choice among these groups. A pioneer in the study of ion channel variants has been Dr. Al George, Magerstadt Professor, Chair of the Department of Pharmacology, and the Director of the Center for Pharmacogenomics, Northwestern University Feinberg School of Medicine. He recognized the great potential of high throughput automated electrophysiology to determine the functional consequences of ion channel variants. His lab had the foresight to purchase a SyncroPatch 384PE system in 2014 to characterize cardiac and brain ion channel variants that

were associated with congenital long-QT syndrome or genetic epilepsy, respectively. One year later, they upgraded the instrument to have a second 384-well module (SyncroPatch 768PE) because of the tremendous initial success with this system. To date his lab has characterized the electrophysiological properties of more than 200 variants in voltage-gated sodium and potassium channels using the SyncroPatch 768PE system,<sup>1-4</sup> with many more in the pipeline. Determining the functional effects of ion channel variants can help with classification as potentially disease causing or not. In their recent study of long QT syndrome type 1, the George (cont.) laboratory was able to reclassify >65% of KCNQ1 (encoding  $K_v 7.1$ ) variants of uncertain significance as likely pathogenic.<sup>1</sup> Most recently, they presented a study of 80 KCNQ2 variants at the annual meeting of the American Epilepsy Society.



**The SyncroPatch 768PE recordings of the cardiac slow delayed rectifier current (IKs) from CHO cells expressing KCNQ1 and KCNE1.** The figure shows a snapshot of SyncroPatch 768PE software, measurements made at Northwestern University. Green fields represent successful recordings, indicating a success rate of approx. 95%.

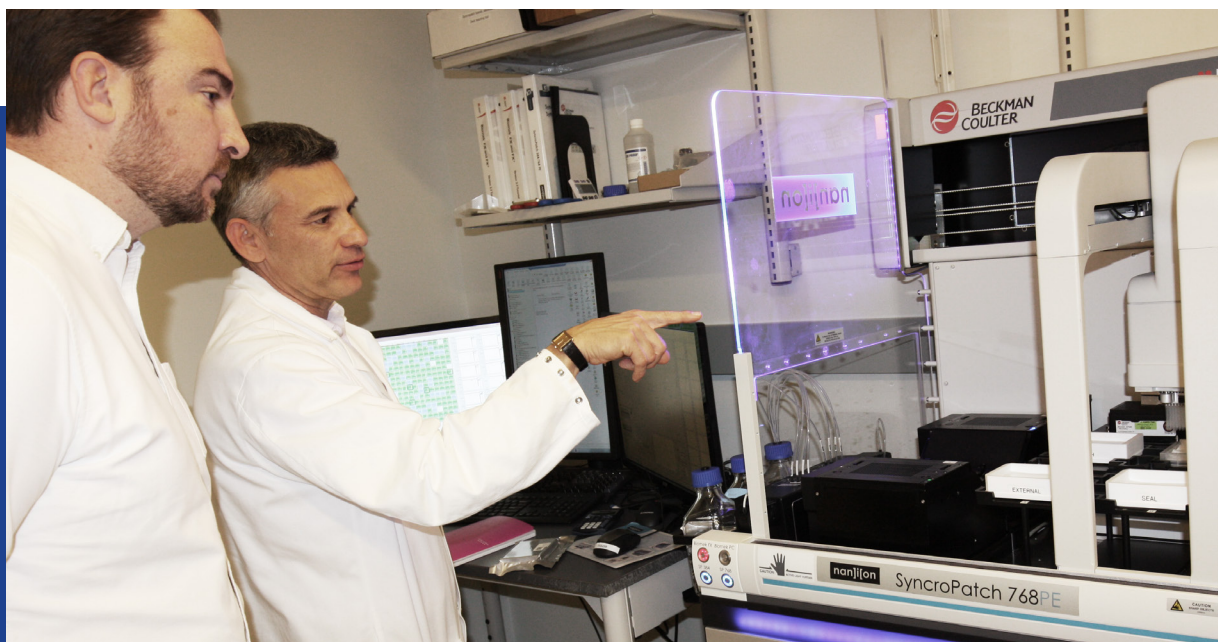
Once functional defects in ion channel variants are identified, researchers can evaluate whether existing drugs or investigational compounds can correct the dysfunction, and potentially translate this information to more precise pharmacotherapy in the clinic. The potential for this strategy to change the treatment of human channelopathies is remarkable and enables a new way of looking at ion channel diseases and their treatment.

While this pioneering work is still emerging and its potential is being rapidly realized, other labs have also embraced the SyncroPatch 384PE to perform similar studies. Jen Pan Ph.D., Director, Translational Neurobiology at the Stanley Center of the Broad Institute, is another early adopter of the SyncroPatch platform, working with it since early 2016.<sup>5-7</sup> Her laboratory and collaborators are using SyncroPatch recordings to study the mechanism of action of ion channel modulators identified from high throughput FLIPR assays. In addition, her group characterized genetic variants of genes that encode voltage-gated sodium and calcium channels that are implicated in schizophrenia and neurological disorders. They developed novel methods for noise analyses using the

SyncroPatch that can estimate single channel conductance and surface expression (Nanion User meeting 2019). They presented a methodology to predict gain-of-function (GOF) and loss-of-function (LOF) and then validated the model by studying 50 variants on the SyncroPatch 384 system.<sup>7</sup>

Researchers at the Victor Chang institute in Australia published a paper recently assessing the functional properties of 30 variants of the KCNH2 gene encoding the  $K_v$  11.1 (or hERG) channel, which is a major gene responsible for congenital long QT syndrome.<sup>8,9</sup> Using the SyncroPatch 384PE system they examined the full range of functional attributes of heterologously expressed hERG channels and were able to distinguish benign from pathogenic variants.

In another recent paper, researchers at Vanderbilt University studied cardiac sodium channel (SCN5A gene encoding NaV 1.5) variants associated with Brugada and long QT syndromes.<sup>10</sup> They studied 83 SCN5A variants including 10 positive and 10 negative controls. They identified 45 new or partial loss-of-function variants, and were able to reclassify 50 of 60 variants of uncertain significance using the SyncroPatch 384PE system.



*Dr. Carlos Vanoye explains the Northwestern University SyncroPatch 768PE to Will Hutson and Leah Schust from the Familie SCN2A Foundation. Together with the members of the foundation, researchers at Northwestern University are collaborating with focus on a sodium channelopathy involved in neuronal disorders (SCN2A encodes one of the brain sodium channels involved with epilepsy and autism). For more information, visit [www.scn2a.org](http://www.scn2a.org).*

**“We believe the SyncroPatch platform will transform our approach to determining the most effective pharmacological remedy for certain channelopathies, and enable a precision medicine approach to these rare genetic conditions.”**

**Alfred L. George, Jr., MD, Northwestern University Feinberg School of Medicine**



Many more variants associated with ion channel diseases are identified every year through clinical genetic testing and genomic research studies. A large proportion of the identified variants get classified as variants of unknown significance, but the use of automated electrophysiology on platforms such as the SyncroPatch 384PE can provide much needed functional data to better classify these variants.

Automated patch clamp systems such as the SyncroPatch 384PE are poised to further transform the channelopathy field enabling researchers the throughput needed to determine the functional consequences of newly discovered ion channel variants, collect novel data on structure-function relationships, ascertain molecular mechanisms of disease, and to elucidate variant-specific pharmacological profiles that may improve clinical management.

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