the test compounds. The next day an antibody for the epitope is added along with a second antibody that produces a chemiluminescent signal. "If you don't permeabilize the membrane you can count the number of channels at the cell surface — it's simple and fast," says Brown. ChanTest's FAST & Lite service runs the antibody-based test alongside automated patch clamping to assess channel function. Assays for other channels are being developed and ChanTest has been awarded a small-business innovation grant from the National Institutes of Health to automate its system.

Patch clamping goes automated

Although indicator-based methods are fast and inexpensive, the gold-standard for assessing ion channels is the Nobel prize-winning technique of patch clamping developed by Erwin Neher and Bert Sakmann in the 1970s. The conventional manual method involves a glass micropipette filled with an ionic solution that electrically connects a silver–silver chloride electrode wire to a small patch of cell membrane. A vital part of the procedure is to get an electrical seal of at least 1 gigaohm between the pipette tip and the membrane; without this seal the tiny currents that pass through the channels in the membrane patch cannot be measured. The drawback is that the technique requires considerable expertise, hours are spent poring over a microscope, and recordings can only be taken from one cell at a time. But over the past few years automation has entered this green-fingered science.

A major player in the automated patch-clamp market is Molecular Devices of Sunnyvale, California, which merged last year with imaging specialists Axon Instruments. Molecular Devices has two high-throughput automated patch-clamping systems that can collect between 100 and 2,000 patch-clamping data points a day, depending on configuration.

IonWorks Quattro from Molecular Devices.

MOLECULAR DEVICES

Both instruments work by sucking cells down against 1–2 μm diameter pores in the base of multi-well plates. The PatchXpress 7000A uses 16-well, glass SealChip plates made by Aviva Biosciences of San Diego, California. The machine places cells in each well and suction holds one cell that falls on the pore in place with sufficient strength to create an electrical seal of 1 gigaohm. The machine uses suction to disrupt the cell membrane to access the interior of the cell, and currents are measured across the entire cell surface. "You are, in effect, reversing traditional patch clamping by having the ground electrode measuring from the inside of the cell rather than from the outside," says Steve Davenport, vice-president of Europe for Molecular Devices. Each well is controlled and monitored individually and cells can be sealed for 30 minutes or more — during which time test compounds can be added to and flushed from the well. A single run takes around 45 minutes. The PatchXpress platform works well for both voltage-gated and ligand-gated ion channels and yields high-quality data comparable to the conventional manual patch-clamp method.

IonWorks Quattro from the same company uses a 384-well Patch Plate, but wells share electronics. "This makes sense for a screening instrument where you need the highest throughput possible without compromising the pharmacology," explains Davenport. The system uses a new technology developed by Molecular Devices called Population Patch Clamp (PPC). PPC uses 64 holes versus a single hole in each well of the Patch Plate. This enables the signal from up to 64 cells in each well to be averaged. "The advantage of PPC over conventional single-hole planar patch-clamp is the reduction in biological variability and substantial increase in the success rate of obtaining a data point from each measurement," says Davenport. Using IonWorks, scientists can measure up to 2,000 data points per day.

This speed doesn't come cheap. Both

STEM-CELL OPTIONS

It's easy to focus on the kit and forget the really important part of the system — the cell. Cells of most interest with respect to ion channels include neurons and heart cells, which cannot be grown for long in culture and do not divide.

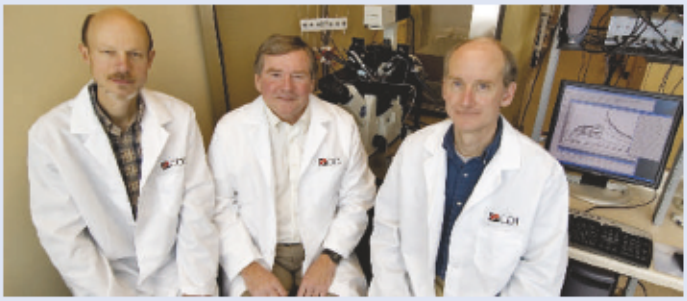
Many of the cell lines used in ion-channel work are, therefore, stem cells and cell lines engineered to express specific channels. These include human embryonic kidney (HEK293) and Chinese hamster ovary (CHO) lines. bSys of Basel, Switzerland, offers a wide range of screening techniques, but chief executive officer Daniel Konrad believes that one of the company's chief advantages is their skill in selecting and fine-tuning cells.

"Each clone of cells is subtly different, and only trialling with many different sources can show which expression system is ideal," he says. bSys also works hard to find the right suspension protocol. This can make the difference between cells that generate 200 picoamp currents and those that can generate 500–1,000 picoamps and can be used in robotic screening systems, says Konrad.

A new company moving into the designer-cell niche is Cellular Dynamics International (CDI) of Madison, Wisconsin, founded by noted human embryonic stem-cell researchers James Thomson, Craig January and Timothy Kamp of the University of Wisconsin. CDI will initially focus on developing HEK cell and cardiomyocyte-based screening services to the pharmaceutical and biotechnology industries, and plans to have a drug-screening service running by the first quarter of 2006.

On the other side of the Atlantic, in Edinburgh, UK, the European arm of Stem Cell Sciences, founded by Peter Mountford in Melbourne, Australia, is developing neural stem (NS) cell lines from the Universities of Edinburgh and Milan. These cells are thought to be phenotypically similar to the NS cells found *in vivo*. Derived from human and animal embryonic stem (ES) cells and from fetal and adult brain tissue, NS cells have great potential in biomedical research because of their homogeneity, their ability to self-renew indefinitely, and their relative ease of manipulation. Stem Cell Sciences is establishing a service for generating specifically mutated NS cells from engineered ES cells and transgenic animals. NS cells are attractive candidates for *in vitro* drug screening and may also be useful for cellular therapy for conditions such as Parkinson's disease and epilepsy.

R&D Systems of Minneapolis, Minnesota offer ready-to-use primary cortical stem cells derived from rat embryos and the kits to grow them. The cells are validated for differentiation into astrocytes, neurons and oligodendrocytes. P.M.



B. FRITZ

Cellular Dynamics International: James Thomson (right), Timothy Kamp (left) and Craig January.