

HEKA *impulse 01*

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Editorial

Established in 1974, HEKA Elektronik has provided the research community with cutting-edge products in electrophysiology and electrochemistry for over 25 years. As the first manufacturer of patch-clamp amplifiers in the world, the company's flagship, the digitally integrated EPC 9 patch-clamp amplifier, revolutionized patch-clamp technology by providing unprecedented accuracy, efficiency, versatility, and ease of use.

Today, the company has established itself as one of the leading hardware and software manufacturers in electrophysiology and electrochemistry worldwide. To ensure the highest degree of flexibility and compatibility, HEKA Elektronik supplies researchers with everything from signal and acquisition to analyzing and data backup systems. In order to perfect HEKA products, HEKA works closely with scientists in its user community.

With corporate headquarters in Germany and Canada, numerous distributors worldwide, and web-based access to products, HEKA guarantees quick and efficient customer support. This support can be addressed through email, telephone or through the multitude of tutorials, Q&A and tips&tricks (please visit us at www.heka.com) The launching of HEKA IMPULSE, the newsletter of HEKA Elektronik, adds a further dimension to the company's accessibility and expert customer support.

Scientific Note

The Stone Age of Patch Clamp Amplifiers

Erwin Neher

*Max Planck Institute for Biophysical Chemistry,
37077 Goettingen, Germany*

The first commercially available patch-clamp amplifier became known as the 'LM-EPC 5'. What does this combination of five letters and a numeral stand for? The 'EPC' is easy – it simply means 'Extracellular Patch Clamp'. 'LM' is slightly more ambiguous. It may be interpreted to represent the initials of Lothar Meyer, the head of the Neurobiology Electronic Workshop at our institute, whose small company built all of the EPC 5's and the first 50 or so of the EPC-7's. Alternatively, 'LM' may be taken to represent 'List Medical', the company that actually marketed the EPC 5's, most of the EPC 7's, and a few of the more recent EPC 9's. The number '5' is the most difficult to be certain about. As far as I remember, we were not quite sure how many different versions of EPC-type amplifiers we had built in the early 70's, before Fred Sigworth, Lothar Meyer and myself had to coin a brand name. We agreed that the number '5' would be about right. Now, 20 years later, it is even more difficult to reconstruct the genealogy of EPC's. Nevertheless, I will try to briefly summarize the history of EPC-type amplifiers in my laboratory that predate the first well-documented one, which is the EPC 5.

Common to all EPC amplifiers is the small head stage, equipped with a BNC-connector for attaching the pipette holder. This was a significant deviation from previous microelectrode amplifiers, where head stages were little boxes connected to pipette holders via short cables. The direct connection at the shortest distance became possible through the development of operational amplifiers that were light enough to be carried, along with a few more electronic components, by a micromanipulator. I built the first amplifier of this type for recording single-channel currents in lipid bilayers – Gramicidin and Alamethicin channels. This amplifier is still sitting in a cabinet in my laboratory (see Fig. 1).



Figure 1

Its head stage was originally equipped with an FMI 380K (Function Modules Inc.) amplifier and a 1 GW feedback resistor. Later the head stage was modified to use a Burr Brown 3523L amplifier and a 10 GW feedback resistor. The main unit consisted of little more than a few switches, BNC connectors, and a panel meter. The circuit (see Fig. 2) was almost as elementary as that depicted in Neher, Sandblom & Eisenman, 1978. Since it actually was not used as a patch-clamp amplifier - its 'pipette holder' carried a small polyethylene recording chamber for bilayer measurements - I would call it the 'EPC 0'.

For actual patch-clamp measurements, Bert Sakmann and I had to extend this circuit by a few more elements to create the 'EPC 1'. The head stage remained largely the same, except that it somewhat decreased in size. However, at that time, loose-seal recording (the Gigaseal had not yet been discovered) required careful balancing of the bath potential, requiring the main unit to adjust this potential. Furthermore, another operational amplifier was required to subtract this potential from the output signal of the first stage. The diagram of the 'EPC 1' is shown in almost full detail in Neher, Sakmann & Steinbach, 1978. Two or three amplifiers of this type were built between 1974 and 1977, including one at Yale University where I spent a year in Chuck Stevens' laboratory. These amplifiers were used for the very first single-channel recordings.

A new challenge surfaced when Franco Conti and I considered to record K^+ -channels from squid axon. Our plan was to do this by inserting an L-shaped patch pipette longitudinally into the axon and to approach the membrane from the inside. This was to be accomplished while the axon membrane was clamped by a conventional clamp circuit. We anticipated that this situation would create problems of excessive 'leak' currents across the pipette-membrane seal. I therefore designed an amplifier that would allow the patch potential to exactly follow the intra-axonal potential, as measured and controlled by the voltage-clamp circuit of the axon. Since I did not trust the accuracy of the

clamp, I incorporated a circuit that would automatically adjust the pipette potential (by slow feedback), to compensate for remaining DC-like current. A push-button allowed the shortening of the feedback's time constant into the sub-second range. This option turned out to be quite convenient for adjusting the pipette potential for zero current in less demanding cases, and it became the 'Search Mode' of later amplifier designs. Following Fred Sigworth's suggestion, I also incorporated a high-frequency boost into the second amplification stage to increase the bandwidth of the recording. That amplifier was built by Lothar Meyer in 1978 and it is quite appropriate to name it 'EPC 2', although several slightly modified versions of the 'EPC 1' were designed in the years between 1974 and 1978.

Experience gained with the EPC 2 and a keen interest for such amplifiers after the original publication of single-channel records (Neher & Sakmann, 1976) encouraged Lothar Meyer to slightly redesign and properly document the 'EPC 2', which made it possible to give our colleagues access to such amplifiers.

Susumu Hagiwara had approached us about the availability of a patch-clamp amplifier. As a result Lothar Meyer was eventually enticed (with some nudging by Joseph B. Patlak, who at that time was a postdoc in my laboratory after his PhD with Hagiwara) to privately build an instrument for sale. This instrument was shipped to Los Angeles in the second half of 1979. During my last visit to UCLA, I saw it on a shelf in the Jerry Lewis Center. A few more of these units were built and, I think, they deserve the name 'EPC 3'.

It should be noted that all of these instruments were 'pregigaseal' instruments. They had no more than 4 operational amplifiers in the main unit, had no capacitance compensation (since voltage steps could not be applied), and had feedback resistors of only 100 to 200 MW. Higher resistors were not necessary, because noise was dominated by the low value of the loose seal anyway.

Requirements for amplifier design dramatically changed with the first Gigaseal in January 1980, when suddenly biology no longer constituted the limiting factor. We first had to increase the feedback resistor to achieve lower noise; then Fred Sigworth added capacitance neutralization (both fast and slow). Eventually a feedback loop for a relatively fast current-clamp mode was also incorporated. By about the end of 1980 we had an amplifier that was relatively well adapted for Gigaseal recordings: the 'EPC 4'. It had most of the features of the EPC 5, except that it still used the Burr Brown 3523L as a current-voltage converter. Redesign of the main circuit board and chassis, replacement of the 3523L by Fred Sigworth's low noise current-voltage converter

and incorporation of a power supply turned this into the 'EPC 5', as it is known by the patch clampers' community. The subsequent models improved several of the key features of the 'EPC 5', adding larger gain ranges and more refined compensation circuitry. Today, HEKA's EPC 9 represents the latest (r)evolutionary EPC model, with full digital control and computerized data acquisition, making patch clamp a powerful and reliable technique for cell biology. EPC's sure have come a long way since the stone ages...

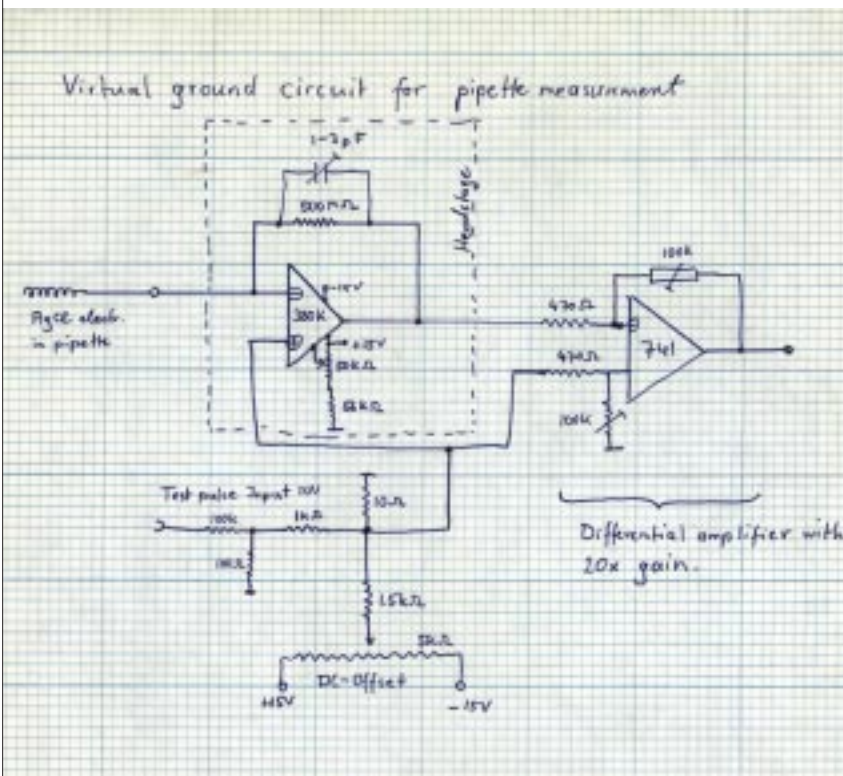


Figure 2

Literature:

Neher, E. and B. Sakmann (1976).
"Single-channel currents recorded from membrane of denervated frog muscle fibres."
Nature 260: 799-802.

Neher, E., B. Sakmann, and J.H. Steinbach (1978).

"The extracellular patch clamp: A method for resolving currents through individual open channels in biological membranes."
Pflügers Arch. 375: 219-228.

Neher, E., J. Sandblom, and G. Eisenman (1978).
"Ionic selectivity, saturation, and block in gramicidin A channels. II. Saturation behavior of single channel conductances and evidence for the existence of multiple binding sites in the channel."
J. Membr. Biol. 40: 97-116.

Scripting PULSE+PULSEFIT

There are two functions that allow writing your own, new controls in PULSE+PULSEFIT. From inside PULSE+PULSEFIT one can record "macros". A macro is a short script, which can comprise up to 40 calls to built-in functions of PULSE, including other macros. Making macros is by far the most popular way to extend the functionality of PULSE.

The second way to control PULSE+PULSEFIT is to send commands to it from an external program. This option, named "Batch Communication", gives the user more control over the behavior of PULSE. The user (i.e., the external program) can send commands to PULSE; PULSE will execute them and message back, if required.

Who might take advantage of this option to control PULSE? An example might be one who needs to integrate PULSE and/or the EPC 9, the computer controlled patch-clamp amplifier, in a more complex system. Here are some presently implemented applications:

- Automated patch-clamp systems. The controlling program handles the required robotics to establish a giga-seal. It uses the EPC 9 to measure the progress in seal resistance, and to determine the quality of the seal. Acquisition is then started according to the requirements defined in the total experimental protocol.
- Automated drug application system with complex perfusion protocols. The controlling program handles the drug application system and starts data acquisition in PULSE at the appropriate times.
- Simultaneous imaging and patch clamping. However, consider less sophisticated applications, e.g.:
 - Monitoring the progress of a long experiment remotely from another computer via the computer network system. Three modes are implemented: monitoring only, notification at defined times in the protocol sequence, and remote control (start/stop)
 - Conditional acquisition, i.e., the controlling program controls the acquisition based on the analysed results of the just acquired data.
 - Customized, automated generation of an experiment protocol comprising multiple analysis and print-out of the data and results

The communication protocol is simple and can be used from any programming language. The complete source code of a VisualBasic program demonstrating that feature is available from HEKA.

Commands are sent to PULSE by writing the corresponding text instructions into a "command" file. PULSE then reads from that command file, executes the commands, and feeds back to the user via a second, "response" file.

For a detailed description, please see the PULSE+PULSEFIT manual, Appendix VI, "Controlling PULSE".

HEKA's new holder takes the wiggle out of pipettes



Patch clampers know that successful Gigaseal formation, clean whole-cell break-ins, and low-noise recordings depend on a number of factors. While the choice of glass and the process of pipette fabrication are important factors, a vibration-free setting is a must. Air-buffered tables generally take care of inherent vibrations originating from buildings. However, movement of the glass pipette itself when applying suction can decrease the quality of high-resistance seals and establishing whole-cell with a pulse of suction is even trickier when pipettes vibrate. Such vibrations can arise from small air leaks and/or an imperfect "grip" of the holder, causing instability of the pipette.

Probably the major culprit in causing pipette movement or air leakage is the variation of the glass capillary's outer diameter. While this may only involve a tenth of a millimeter or so, the existing O-rings of a holder may not be able to completely compensate for such variations, especially if the O-rings are brittle or salt-encrusted. Cleaning the pipette holder or replacing O-rings frequently may reduce problems with pipette movement. However, a holder that offers higher pipette stability in the first place is high on the wish list of patch-clampers.

HEKA's new optimized pipette holder, made of extremely low-noise polycarbonate, offers two major improvements that virtually eliminate pipette movement and air leakage by elongation of the holder's cap and addition of a third O-ring. The longer cap allows for the insertion of a small polycarbonate cylinder, keeping the first O-ring firmly in place, even after removal of the cap for cleaning purposes. The second O-ring is nestled at the other end of the short cylinder featuring a precision mill cut that holds it in place. This new design provides the highest pipette stability, eliminates air leaks, and extends the life time of O-rings. Most importantly, this new holder will increase the rate of successful recordings and increase productivity.

TIB 14 digital trigger interface



Electrophysiological setups have to be increasingly flexible nowadays. More parameters than ever need to be acquired or altered simultaneously during data acquisition. Controlling bath perfusion or synchronizing video imaging systems with data acquisition are standard routines in many laboratories today. The EPC 9 offers 3 free DA outputs that are controllable via software and can be used for any desired purpose. However, some experimental approaches require significantly more DA channels.

We have addressed this need by developing the digital trigger interface TIB 14, which expands the EPC-9's capacity by 14 digital TTL outputs and 14 Open Collector outputs.

The TIB 14 is fully supported by the PULSE software, enabling the simultaneous control of numerous devices, such as magnetic valves, shutters, stimulators, etc. within one software application. The Open Collector outputs can optionally carry three different voltages which, in contrast to other digital I/O boards, eliminates the use of additional hardware to trigger external devices.

Users of EPC 9 Double or Triple will not need the TIB 14, since they can directly use the open collector outputs at the back of these amplifiers to control external devices (e.g. valves). However, the digital outputs of the EPC 9 should not be used without interfacing with the TIB 14, since part of their circuits (bit0...bit15) are used for internal control of the EPC 9.

| Triggers | 3 | #1 (+) | #2 (*) | #3 (x) |
|--------------|---|----------|----------|---------|
| DA channel | | dig-2 | dig-3 | dig-3 |
| Segment | | 1 | 2 | 2 |
| Time [ms] | | 50.00 | 50.00 | 450.00 |
| Length [ms] | | 2.50 | 2.50 | 2.50 |
| Voltage [mV] | | TTL-high | TTL-high | TTL-low |

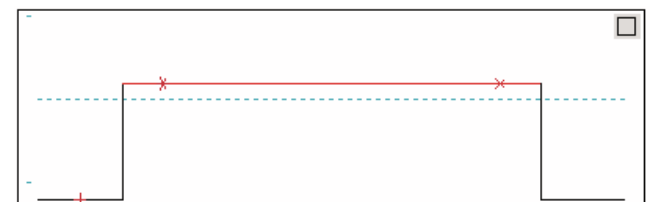


Figure 2

| | | |
|--|---|-----|
| <input type="checkbox"/> Dig0 | 0 | Ext |
| <input type="checkbox"/> Dig1 | 0 | Ext |
| <input checked="" type="checkbox"/> Dig2 | 0 | Ext |
| <input type="checkbox"/> Dig3 | 0 | Ext |
| <input type="checkbox"/> Dig4 | 0 | Ext |
| <input checked="" type="checkbox"/> Dig5 | 0 | Ext |
| <input type="checkbox"/> Dig6 | 0 | Ext |
| <input type="checkbox"/> Dig7 | 0 | Ext |
| Clear Digital Port | | |

Figure 1

The software allows TTL-signals either to be set manually or by a mouse-click on the corresponding check box in the amplifier window (see fig. 1) or at defined times within an acquisition sequence (see fig. 2).

The HEKA History

HEKA was founded in 1968 by three shareholders to produce controller units for production machinery. The company experienced a fresh start when Peter Schulze, an engineer with a Ph.D. in electrochemistry, joined HEKA in 1974. Dr. Schulze soon became president of HEKA and he maintains this function to this day. It manufactured and marketed new products such as parts of tape drives or three-dimensional CNC control units for punching machines. This allowed HEKA engineers to embark on the more risky endeavor developing research equipment for electrochemical, biological and physiological applications.

HEKA's growth has been closely linked to collaborations with some of the world's leading research laboratories. For example, quite a few Max-Planck Institutes became a part of HEKA's history. Instrumental in this process was Garching Innovation, the company of the Max-Planck Society responsible for technology transfer.

Developmental milestones included a cytometer measuring different parameters of cells such as volume and optical properties. Software to detect cell life time cycles in cancer research and potentiostats have been developed in cooperation with the Fritz Haber Institut in Berlin. The potentiostat proved to play a pivotal role in HEKA's collaboration with Dr. Erwin Neher and Dr. Fred Sigworth at the Max-Planck Institute in Göttingen. During a visit in Dr. Neher's laboratory, he described his newest technical developments. He explained to us that "he had a few colleagues interested in the equipment" and that "he wanted to make the new development available for everyone at a fair price". He estimated that all in all "perhaps 30 pieces could be sold...". This introduced HEKA to a newly established method to measure currents across membranes of living cells, which Erwin Neher had called the "Patch Clamp". Obviously, the story did not end after manufacturing thirty amplifiers, but instead the method revolutionized cellular physiology.

We started producing the EPC 5 and shortly thereafter a greatly improved version, the EPC 7. Fred Sigworth was at the forefront of these developments. He wrote a manual that for the longest time was referred to as the "Patcher's bible". At that time, List Medical managed the marketing since HEKA was new to the field of electrophysiology. The demand for EPC 7s was beyond all expectations, which kept HEKA busy. Soon other companies recognized the enormous potential of HEKA's newest patented amplifier system and introduced headstages that were curiously similar to the EPC 7 headstage. For many years, the joint venture between HEKA,

Jürgen List's company List Medical and John Adams' Medical Systems provided researchers worldwide with the highest quality equipment. A number of reliable companies joined HEKA, skillfully trading HEKA products. Their interaction with researchers around the world and the feedback to us was invaluable in the ongoing process to improve our products. HEKA's growing needs for more space resulted in the move of the company in 1984 from romantic, vineyard surrounded Forst to the city of Lambrecht, just 15 kilometers farther away.

Three years later Erwin Neher's laboratory started the development of the EPC 9. The simple recipe behind the idea was to maximize the efficiency of electrophysiological research by integrating the amplifier and the data acquisition systems such that data could be stored on a computer. This sprouted the idea to go all the way and actually develop a system that would control the amplifier itself by the same computer. We found a powerful partner in InstruTECH to help us alter the original design of a lab interface called LIH to the potent A/D-D/A converter ITC 16 with its 16 bit resolution. This step was crucial since 12 bit A/D-D/A converters proved inadequate to handle the requirements of a fully digitalized patch clamp amplifier.

Thus, the EPC 9 was born as the best patch clamp amplifier on the market. The amplifier was controlled by software written for Atari computers, which at that time were one of the leading personal computers that had enough RAM (4 Mbytes), excellent graphic features and an affordable price.

The digitalization of the data acquisition process allowed us to incorporate many superb features developed by Fred Sigworth. Most notably perhaps was the automated capacitance compensation. The unique design of the EPC 9 also allowed researchers to study exocytotic events with ease using the software "Lock-in Amplifier". The new software took advantage of the fact that - as another first - the EPC 9's headstage could be calibrated. The EPC 9 also permitted easy and gentle switching between current clamp and voltage clamp. Obviously, dial-controlled amplifiers could not and cannot offer features like the above. Furthermore, the overall noise performance of the EPC 9 continues to be outstanding. This is especially true when one is reminded that noise performance in the EPC 9's case includes the performance of both the amplifier and the digital interface.

Atari went out of business in 1992. Just in time, the equally powerful Macintosh computer dropped dramatically in price and was therefore chosen to replace the Atari system. However, these events led to challenging times for HEKA. Although we successfully introduced the new

software PULSE for the Macintosh, which was spearheaded by Dr. Hubert Affolter, many obstacles had to be dealt with, including the introduction of the power PC and the PCI bus system. Nevertheless, we were able to improve PULSE step by step, thanks to our customers and their patient reporting of bugs and problems with the software. There was simply no better software for electrophysiology on the market and HEKA worked very hard to meet the needs of its customers.

We introduced PULSE for MS Windows in collaboration with Dan Brown from Scalar in Seattle in 1995.

The ever-increasing performance of modern computer technology allowed us to equip the PULSE software with over 1000 new features, including Online Analysis, Solution Control and Data Base, defined Parameter Acquisition, Automated Calibration, as well as control of the amplifier by external software.

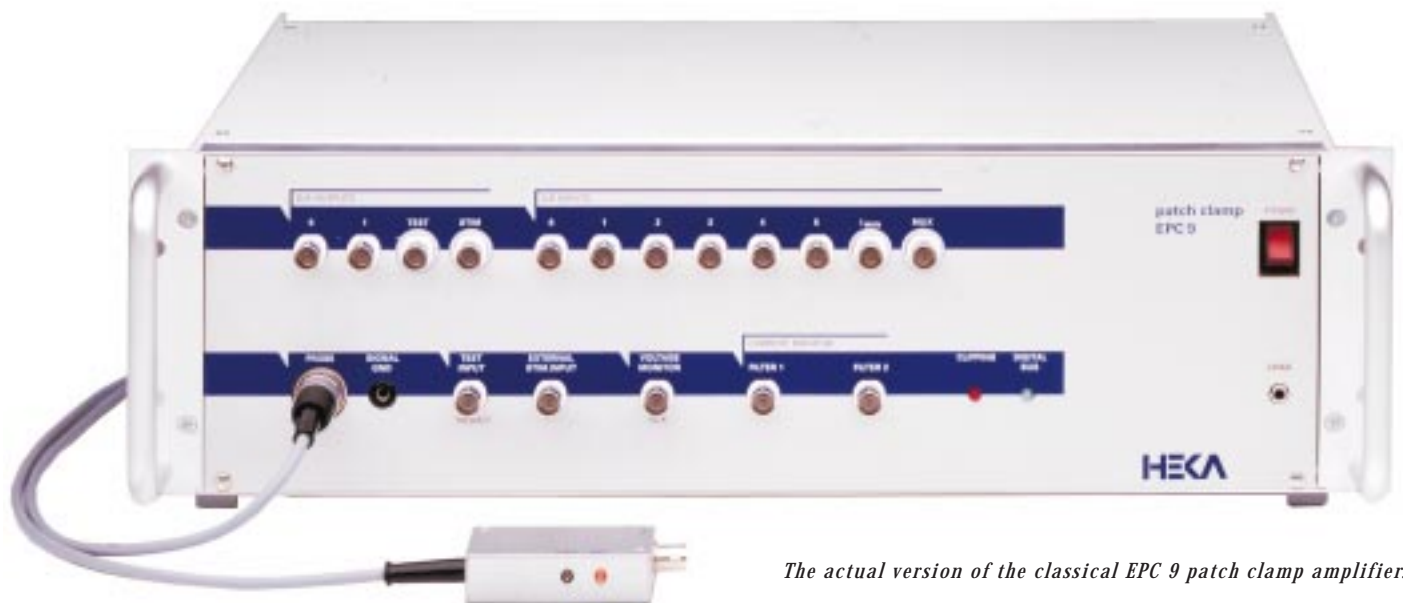
We knew from the very beginning that we had to push hard to be cutting-edge in the business and we succeeded. The competition was forced to emulate soft- and hardware features, but our software had been so carefully developed that it took competitors thirteen years to introduce their "new" amplifiers showing the same control

buttons, button by button, as we once introduced. HEKA is still proud that our customers following our ideas made the right decisions, the only right ones....what we promised year by year.. our customers got it.... We at HEKA are pleased that we have been providing digitally integrated acquisition/analysis systems for over a decade now.

Since dial-driven amplifiers were still being requested, HEKA introduced the successor of the EPC 7, the EPC 8. The new instrument had many new features and expanded capabilities. In 1995, we were able to offer the EPC 9 Double and EPC 9 Triple with either two or three independent amplifiers in one housing and optimized ground connections. Both the EPC 9 Double and EPC 9 Triple could still be controlled through one interface and one computer. The addition of a family of compatible software made the whole design very powerful.

Recently we founded 'HEKA Electronics' to promote the support for our customers in North America. The new company is located in Mahone Bay, Nova Scotia, Canada, a famous former privateer's place.

We believe that progress never ends and our friends can rest assured that HEKA will continue to provide the best research equipment available to the field.



The actual version of the classical EPC 9 patch clamp amplifier.

Q I am interested in measuring membrane capacitance as an assay for exocytosis. Can this be performed with your software controlled, EPC 9 patch-clamp amplifier ?

A Yes! HEKA's EPC 9 patch-clamp amplifier is completely computer controlled and includes a built-in data acquisition interface (ITC-16, InstruTECH, New York, USA). The stimulus and data acquisition capabilities of the ITC-16, as well as the instrumentation settings of the EPC-9, are controlled by HEKA's "PULSE" software package. One of the great advantages and unique features of this fully integrated hardware and software combination is the ability to implement a lock-in amplifier feature. HEKA has implemented the lock-in amplifier with the PULSE software and it was developed for the very raised proposed in your question, (i.e., it allows high resolution measurement of membrane capacitance (C_m) as a single-cell assay for such things as exocytosis or endocytosis).

Basically, changes in C_m are usually measured by applying a sinusoidal voltage stimulus and the resultant sinusoidal current is processed using a phase-sensitive detector or lock-in amplifier; it is the resulting phase shift that is used to estimate capacitance changes. The EPC 9 is the ideal device for measuring C_m because all the relevant parameters that can potentially affect C_m measurements are under software control, eliminating any manual input.

The digital control of filter settings, gains, and compensation networks enable the admittance of the signal source at the amplifier's input to be determined on the basis of the amplifiers own calibration.

Attenuation and phase shifts introduced within the EPC 9 by low-pass filters and other circuitry, which would affect capacitance estimation in other amplifiers, are modeled and automatically corrected by the PULSE software. In addition, changes in the measured signal introduced by whole-cell capacitance and series resistance compensation are accounted for. The noise of capacitance measurements is nearly optimal, and resistive parameters can vary over a large range without inducing artifactual changes in capacitance estimates.

We should point out that although the software lock-in amplifier is tailor made to be used with the EPC 9, it can be used with other amplifiers such as the EPC 8. Consider, however, that the phase shift caused by changing filter settings or any other critical parameters such as the sinusoid frequency, requires a new measured calibration to be made. There is no way for the lock-in amplifier feature to "know" if you changed a critical parameter, so it is up to the user to ensure that the calibration is valid. More details about HEKA's software lock-in amplifier can be found in the following paper.

Gillis, K.D. (2000). Admittance-based measurement of membrane capacitance using the EPC-9 patch-clamp amplifier. *Pflügers Arch.* 439: 655-664.

Q Upon startup of PULSE the following error message comes up: 'length conflict loading D_EPC9.dia. Loading aborted'. Nevertheless the program runs and seems to work fine. What is the meaning of this error message and how can I avoid it?

A This error message indicates a version conflict between the installed PULSE version and the file D_EPC9.dia. The files with the extension 'dia' are used to store dialog settings, like dialog position, colors and fonts. Each dialog has its own 'dia' file. The file D_EPC9.dia e.g. contains information about the EPC 9 amplifier dialog settings. When installing a new version of PULSE+PULSEFIT any customized dialogs are likely to become incompatible, because additional items will have been introduced in the new version. It is therefore best to trash these custom dialogs when upgrading. If you want to change the dialog settings, the new layout can be stored with the command Front Dialog -> Save. This command will create a new 'dia' file for the active dialog.

Q How can we print several traces from the screen, for example clamp data from several different command potentials?

A Set 'Export: ' in the Tree menu to 'Printer'. 'Export' in the same menu will print the data selected in the 'Replay' Dialog. If a series is selected (and the overlay button in the oscilloscope dialog is pressed), then PULSE will print all sweeps in the series.

If you want to print out only certain sweeps from a series, you could mark these sweeps ('Marks' menu) and use the 'Export' item in the 'Marks' menu. If the overlay button in the oscilloscope dialog is pressed, PULSE will print all marked sweeps (within one series) on one graph.

Q We understand that when a protocol is executed, the data are written to a temporary file and only written in final form at the termination of the experiment. Please advise:

1. what is the name and where is the file stored that is the temporary file of data. 2. is it possible to write, the data from each protocol (trace, manipulation of experiment) to a permanent file instead of waiting to the end of the experiment thus risking losing all the data if the computer should crash at that point?

A During acquisition the data is not stored to a temporary file, but kept in the computer's RAM. The raw data is written to the hard disk after

each sweep, but the file structure information is written at the end of the experiment.

Therefore, if the computer crashes before closing the file, the raw data (*.dat file) is stored, but cannot be opened in PULSE. Please refer to the chapter 'Troubleshooting' in the PULSE manual. Here you will find information on how to recreate a valid experiment from the raw data file if you loose the Stim and the Pulsed Tree due to a computer crash.

To minimize the risk of data loss, you should frequently use the menu option 'Update File' or enable the setting File -> Auto File Update.

Windows operating systems write all data into a file cache first. If you are using a Windows machine, you should disable 'Data File Caching'. Although disabling this feature slows the storing of data, it has the benefit of more safety.

Q I am using X-Chart 8.31. I have a problem opening exported X-chart data into IGOR. I have tried all the export possibilities but all I get is an error messages in Igor saying 'expected terminating quote'. Is there a trick to avoid this or is it fixed in the new version? The same situation occurs with the online analysis.

A There seems to have been a syntax error in older versions of X-Chart when creating an Igor ipx-file. This bug has been fixed in the recently released version 8.50. The newest version is available for free download at www.heka.com.

Q We just got a new Apple G4, and I was keen to run PULSE on it but realized that the dongle I used on the older PowerMacs no longer fits the USB ports of the G4. Is there a way around that problem? Maybe an adapter?

A Due to the new USB standard HEKA now offers dongles that are USB compatible. You can opt to send us your current PULSE ADB dongle and exchange it with a USB compatible dongle at a reduced net price of DM 300,-. An interim solution would be to acquire 'iMate' from Griffin Technology (www.griffintechology.com), which according to one of our customers works well as ADB/USB adapter when using PULSE on a Mac G4, 350 MHz.

Events Electrophysiology

- Meeting of the Society for Neuroscience
November 10th - 15th, 2001,
San Diego, California.
- Canadian Physiological Society Meeting
January 30th - February 3rd, 2002,
Silverstar Mountain, British Columbia.
- Meeting of the Biophysical Society
February 23rd - 27th, 2002,
San Francisco, California.
- Gemeinsame Tagung der Deutschen
Physiologischen Gesellschaft und
"The Physiological Society"
March 15th-19th, 2002,
Tübingen, Germany.
- 3rd Forum of European Neuroscience (FENS)
July 13th-17th, 2002,
Paris, France.

Events Electrochemistry

- 5. Vortragstagung des Arbeitskreises
"Chemische Analysemethoden" ELACH 5
Sep. 30 - Oct. 4, 2001,
Freiburg/Breisgau, Germany.

HEKA Courses

Introductory courses to PULSE and Software LockIn (for electrophysiologists) or POTPULSE (for electrochemists) will be held in Lambrecht/Germany. These courses will cover the basic principles of the program and show first steps involved in performing standard measurements. Please contact us for more detailed informations.

PULSE Tutorial: Stimulating with a complex pulse pattern

The stimulus template output by PULSE can either be computed by the program or loaded from a file by activating "Get File Template" in the timing section of the Pulse Generator window. This way you can stimulate any complex pulse pattern (i.e., a pre-recorded voltage trace such as an action potential) that PULSE otherwise could not calculate. Below is a step-by-step example demonstrating how the "File Template" works to stimulate a pre-recorded pulse pattern. This can easily be tested using a model circuit.

1. Generate a simple stimulus named "Get" with one sweep per series (No of sweeps =1) and three constant segments (Duration 20, 10, and 50 ms; Voltage -80, 0, and -80 mV). There should not be any triggers (Triggers = 0), the sample interval should be 0.1 ms, and only one input channel has to be acquired (Channels = 1). The sequence should not be linked with any others (Linked sequence = NIL) nor should there be any leak pulses (No of Leaks = 0). Stim DA and input channels are set to Default.

2. In the oscilloscope window, execute the "Get stimulus. You should see the current response with its corresponding "pulse" shape. Let us assume that the response is 50 pA in amplitude. Note: the Store button must be activated in the oscilloscope, otherwise the sweep will not be stored.

3. To generate a file template, first clear the "sweep buffer" (Buffer...Clear) and then add the acquired sweep into it (Buffer....Add).

4. The sweep data has been acquired as pA and now has to be scaled into mV. This is done by calling Buffer.....Scale and entering "1e9" as the scale factor.

5. Store the sweep buffer to disk as "Get_1" (Buffer...Save as binary file...)

6. Now, return to the pulse generator window, select the "Get" stimulus and activate "Get File Template".

7. Finally, execute the "Get" stimulus again in the oscilloscope window. The file template is read and used as the template, and you should see the corresponding current response.

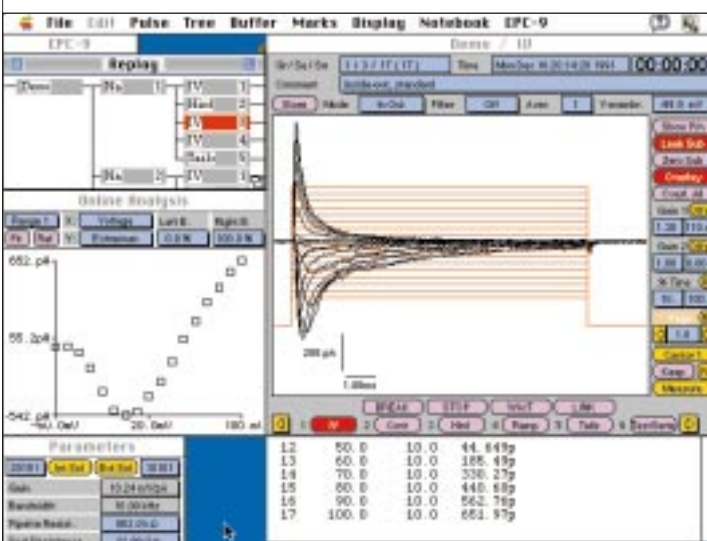
Alternatively, you can use third party programs such as IGOR Pro to generate the stimulus template file.

Please consider the following points when using the file template feature:

1. The template must be in a file in the same folder where the "pgf"-files are. You can also put the files into a sub-folder inside that folder, but the folder name must be the same as the stimulus.

2. The name of the template file must be "[stimulus name]_[sweep number]". Example: if the stimulus name is "IV", then PULSE looks for the template file "IV_1" to be the first sweep, "IV_2" for the second sweep, etc..

3. The file must contain one voltage value per stimulus point. The voltage value must be "short" (4 byte), binary IEEE-floating point format number. All values must be in volts, i.e., for a voltage output of -80 mV, the required value will be - 0.080.



HEKA Electronics Incorporated is the new North American arm of HEKA. HEKA began operations in Canada in 1999 and we are already planning an expansion of our facility. Activities in the Canadian office of HEKA include the assembly of all EPC 7, EPC 8 and EPC 9 single, double and triple amplifiers. Repairs for North American customers are also carried out in Canada.

The HEKA office is located in Mahone Bay, Nova Scotia. Mahone Bay is a beautiful seaside town located about a one hour drive South of the Halifax airport. Halifax is served by direct flights from Boston, New York, Toronto, Vancouver, Montreal and Europe. The South shore of Nova Scotia is a Mecca for both tourists and professional people alike. We are expanding our professional staff in order to conduct development and to be better able to advise our North American customers. We have recently added electronic, physiology and information system specialists to our Canadian staff.

Clients in the United States of America should contact our distributors, Instrutec Corp. or ALA for their equipment needs.

Australia and New Zealand:

SDR Clinical Technology
213 Eastern Valley Way
Middle Cove NSW 2068
Tel: +61 - (0) 2 9958 2688
Fax: +61 - (0) 2 9958 2655
Email: sdr@sdr.com.au

China:

IBB Instruments
School of Life Science
and Technology
Wuhan, Hubei 430074, P.R.China
Tel: +86 - (0) 27 - 87544375
Fax: +86 - (0) 27 - 87543104
Email: ibb@hust.edu.cn

Bioprobes Scientific &
Medical Supplies
RM. 22D, Jia Di Hua Yuan Bldg.
77-79 Shi You Xin Ma Lu
Guangzhou PR China 510600
Tel:
+86 (0) 20 - 8736 4699 / 8736 4606
Fax:
+86 (0) 20 - 8736 4482
Email:
bioprobe@public.guangzhou.gd.cn

France:

SEGA
Medical et Recherche
5, Rue Broussais
75014 Paris
Tel: +33 (0) 1 45-65-22-22
Fax: +33 (0) 1 45-80-44-54
Email:
segaelect@aol.com World Wide
Web:
<http://members.aol.com/segaelect>

Great Britain:

Digitimer Ltd.
37 Hydeway
Welwyn Garden City,
Hertfordshire, AL7 3BE
Tel: +44 (0) 1707 328347
Fax: +44 (0) 1707 373153
Email: bcooper@digitimer.com
World Wide Web:
<http://www.digitimer.com>

Hong Kong:

Bioprobes Scientific & Medical
Supplies
Rm 702 7/F., Knutsford Comm. Bldg.
4-5 Knutsford Terrace, Tsimshatsui
Kowloon, Hong Kong
Tel: +852 (0) 2723 9888
Fax: +852 (0) 2724 2633
Email: bioprobe@glink.net.hk

Israel:

N.B.T. New Biotechnology Ltd.
5 Nakdimon St.
POB. 8662 Jerusalem 91086
Tel: +972 (0) 2-5633 399
Fax: +972 (0) 2-5610 208
Email: nbtsales@inter.net.il

Japan:

Shoshin em Corp.
Shoshin Bldg.
1-14 Kuranishi, Akashibucho
Okazaki 444-02
Tel: +81 (0) 564 541231
Fax: +81 (0) 564 543207
Email: shoem@sun-inet.or.jp
World Wide Web:
<http://www.shoshin.com>

Korea:

Scitech Korea Inc.
40-5 Wooi-dong, Kangbuk-ku
Seoul 142-871, Korea
Tel: +82 02 999 4419
Fax: +82 02 999 4416
Email: scitech@kornet.net

USA:

ALA Scientific Instruments Inc.
1100 Shames Drive
Westbury, NY 11590
Tel: +1 (0) 516-997-5780
Fax: +1 (0) 516-997-0528
Email: staff@alascience.com
World Wide Web:
<http://www.alascience.com>

INSTRUTECH Corp.
20 Vanderventer Avenue
Suite 101E
Port Washington, NY 11050-3752
Tel: +1 (0) 516-883-1300
Fax: +1 (0) 516-883-1558
Email: sales@instrutech.com
World Wide Web:
<http://www.instrutech.com>

Taiwan:

Ying Sheng
Scientific Apparatus Co.
P.O.Box: 46-597
Taipei
Tel: + 886-2-2563 9858
Fax: + 886-2-2531 8699
Email: jinshen@ms15.hinet.net

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HEKA Elektronik
Dr. Schulze GmbH
Wiesenstraße 71
D-67466 Lambrecht/Pfalz
Germany

Phone +49 (0) 63 25 / 95 53-0
Fax +49 (0) 63 25 / 95 53-50
Web Site <http://www.heka.com>
Email sales@heka.com
support@heka.com

HEKA Electronics Incorporated
47 Keddy Bridge Road
R.R. #2, Mahone Bay
Nova Scotia, Canada
B0J-2E0

Phone +1 902 624 0606
Fax +1 902 624 0310
Web Site <http://www.heka.com>
Email heka@ns.sympatico.ca

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Editor

Prof. Dr. Andrea Fleig

Design

Zuerker Informationsdesign
info@zuerker-infodesign.de

HEKA Contributors

Dr. Peter Schulze
Dr. Bernd Letz
Dr. Hubert Affolter

Additional Contributors

Prof. Dr. Erwin Neher
Prof. Dr. Andrea Fleig