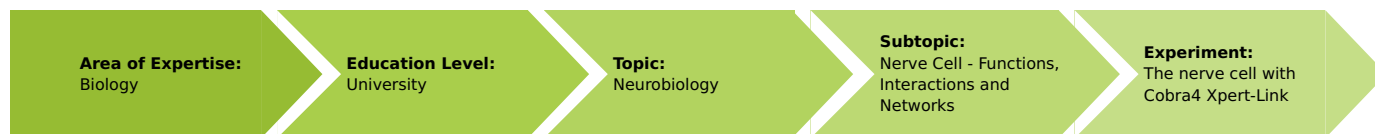


The nerve cell with Cobra4 Xpert-Link (Item No.: P4010764)

Curricular Relevance



Difficulty



Difficult

Preparation Time



10 Minutes

Execution Time



2 Hours

Recommended Group Size



2 Students

Additional Requirements:

- PC

Experiment Variations:

Keywords:

Membrane potential, Action potential, Types of synapses, Depolarisation, Summation, Hyperpolarisation

Task and equipment

Introduction

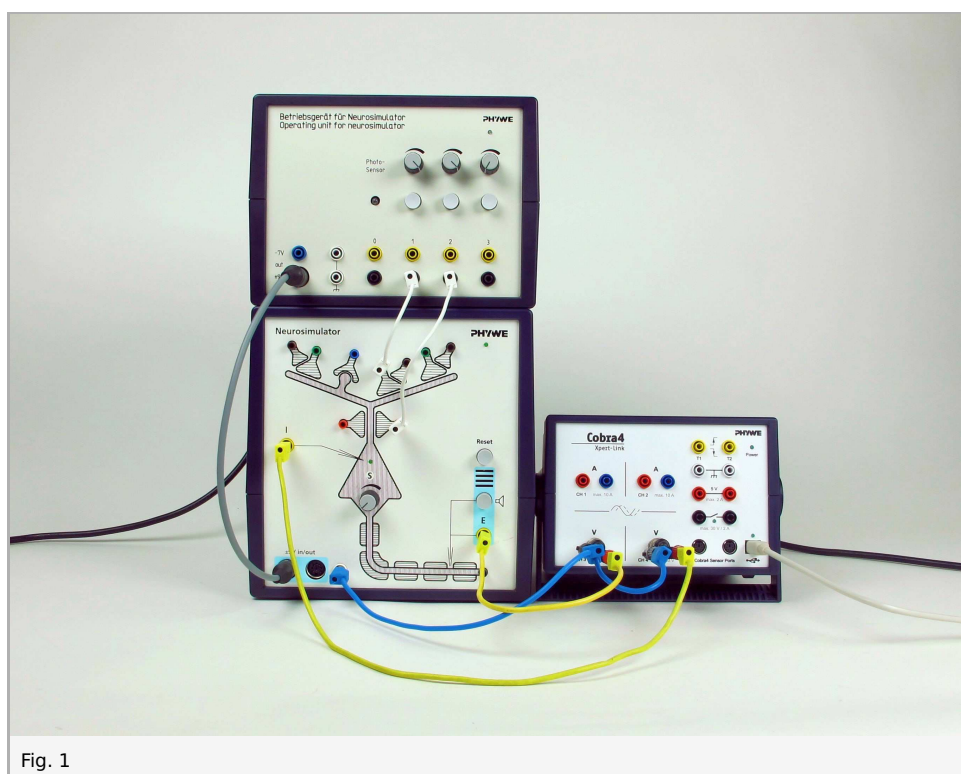


Fig. 1

Function and Applications

The neuron unit Neurosimulator simulates a generalised nerve cell with an apical dendrite and its synaptic contacts, a cell body (soma) and a nerve fibre (axon) with myelin sheathes and a Ranvier's ring.

The operating unit comprises the power supply of up to four neuron units, three touch simulators with a variable stimulating intensity and an optical sensor.

The Cobra4 Xpert-Link is the perfect measurement device for all experiments. Up to 3 values can be measured: 2 values using the two internal voltage channels and optionally, for even more sophisticated experiments not documented here, 1 value via the Cobra4 Sensor Ports. (To use the Sensor Port, a Cobra4 Sensor-Unit Electricity (12644-00) and a Cobra4 Xpert-Connect (12625-01) are needed additionally.)

Benefits

- The dendrite comprises exciting, inhibiting, presynaptic and Hebbian synapses which are marked by the corresponding colours of the sockets
- here the axons end in presynaptic buttons
- these are represented together with a part of the (afferent) fibre providing the signal
- the connection between the (efferent) axon of a neuron unit which leads away or the stimulus output socket of the operating unit and a synapse is established by means of a white cable which is inserted into the desired synapse socket
- the yellow sockets serve for the derivation of the state of excitement of the simulated neuron
- they must be connected to the computer interface Xpert-Link
- the action potentials can be made audible with the aid of the integrated acoustic monitor
- the turning knob "S" serves for setting the "firing threshold" of the neuron.

Equipment

Position No.	Material	Order No.	Quantity
1	Cobra4 Xpert-Link	12625-99	1
2	Neuro-simulator	65963-00	1
3	Neuro-simulator, power supply	65963-93	1
4	Software measureLAB	14580-61	1
5	Adapter, BNC-plug/socket 4 mm	07542-26	2

Instructions for measureLAB

Create chart

Start measureLAB. You will see:

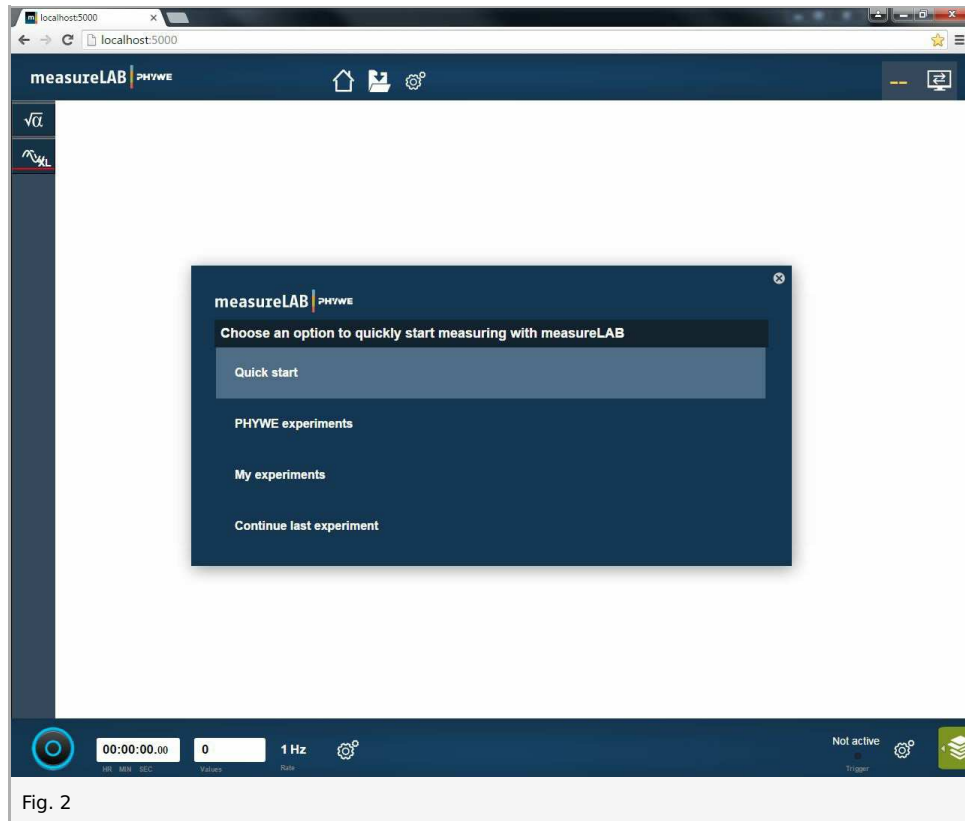


Fig. 2

The measured values of the measured channels can be recorded together in one diagram or individually in two separate diagram, one for each measurement channel (this is necessary if different voltage scales are used). You can choose between the two options when you start measureLAB (fig. 3 and fig. 9), or you can change the windows later (fig. 7, 8, 10).

Select "Quick start" and then the channels that you want to measure (below, instruction for recording into one diagram from the outset):

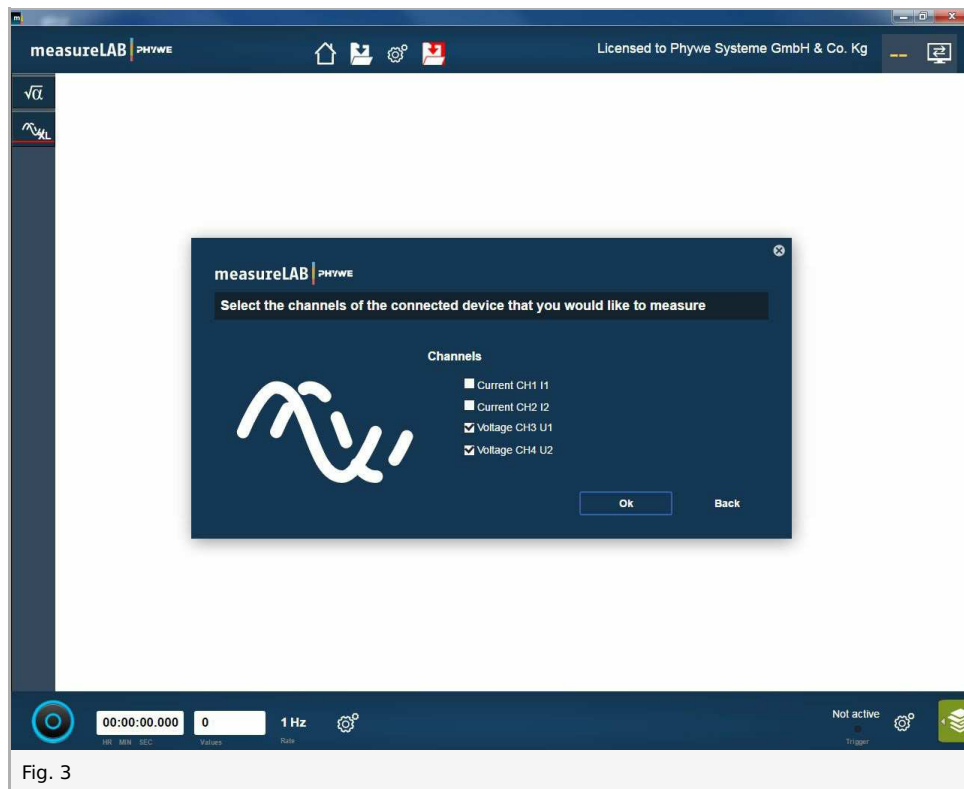


Fig. 3

You will see:

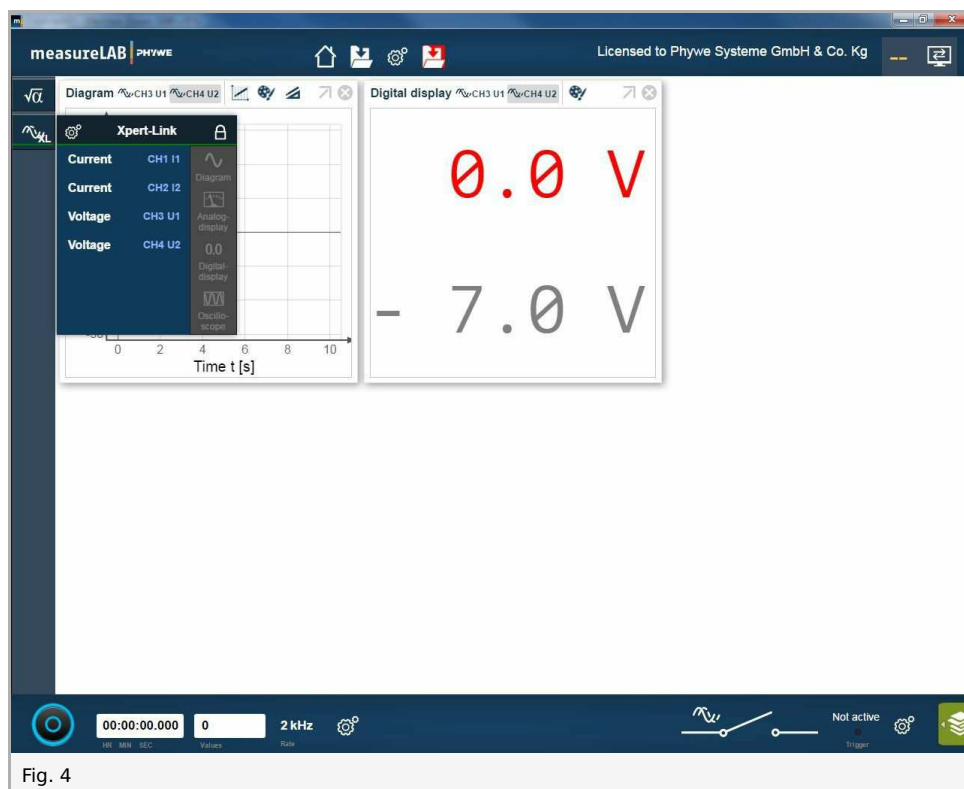


Fig. 4

Move the diagram into a better position and enlarge them by clicking on the chart button: press and hold the left mouse button and move the mouse.

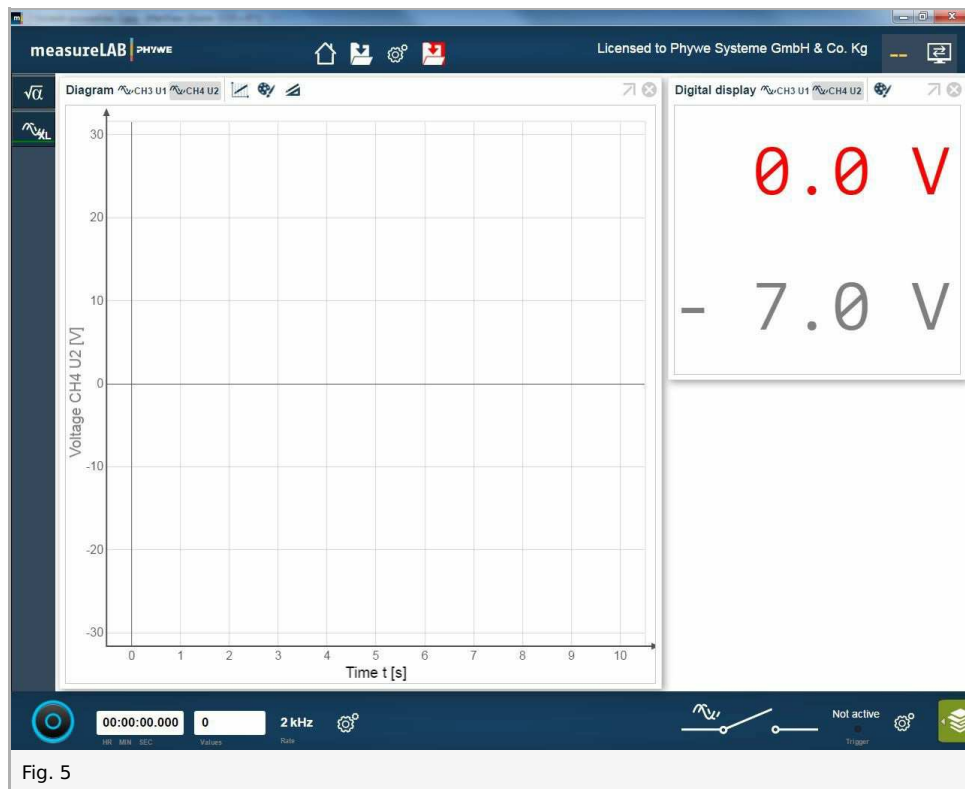




Fig. 5

By clicking on the grey arrow button , you can maximize the diagram.
Now, by clicking on the grey arrow field right , you can minimize the diagram.

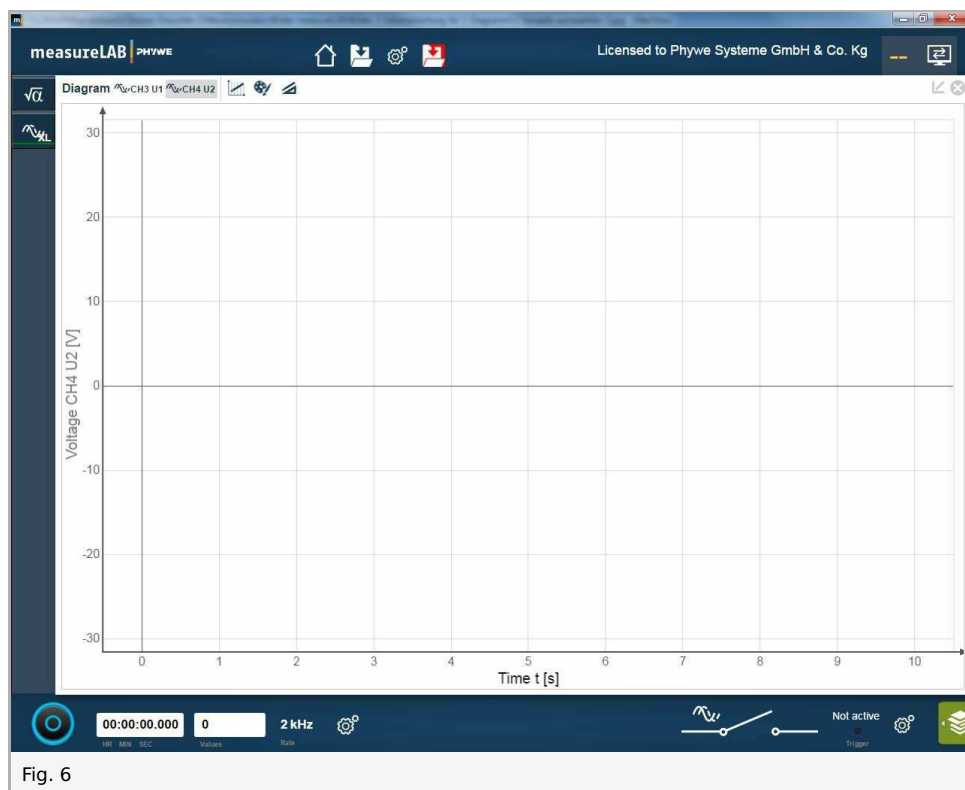


Fig. 6

Change in measured channels in the diagram:
By clicking on the button of the channel with the **right** mouse button and on the field "remove" with the **left** mouse button you can delete this channel.

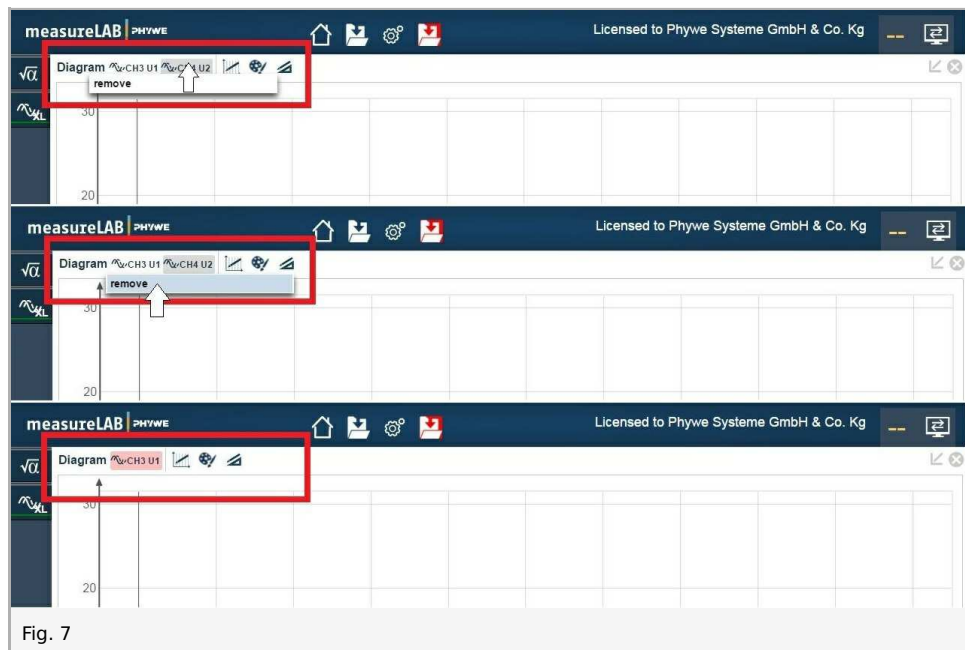



Fig. 7

By clicking on the button  you can add more measuring channels.

Select the channel by clicking on the channel button with the **left** mouse button, hold this button and move the symbol into white chart field.

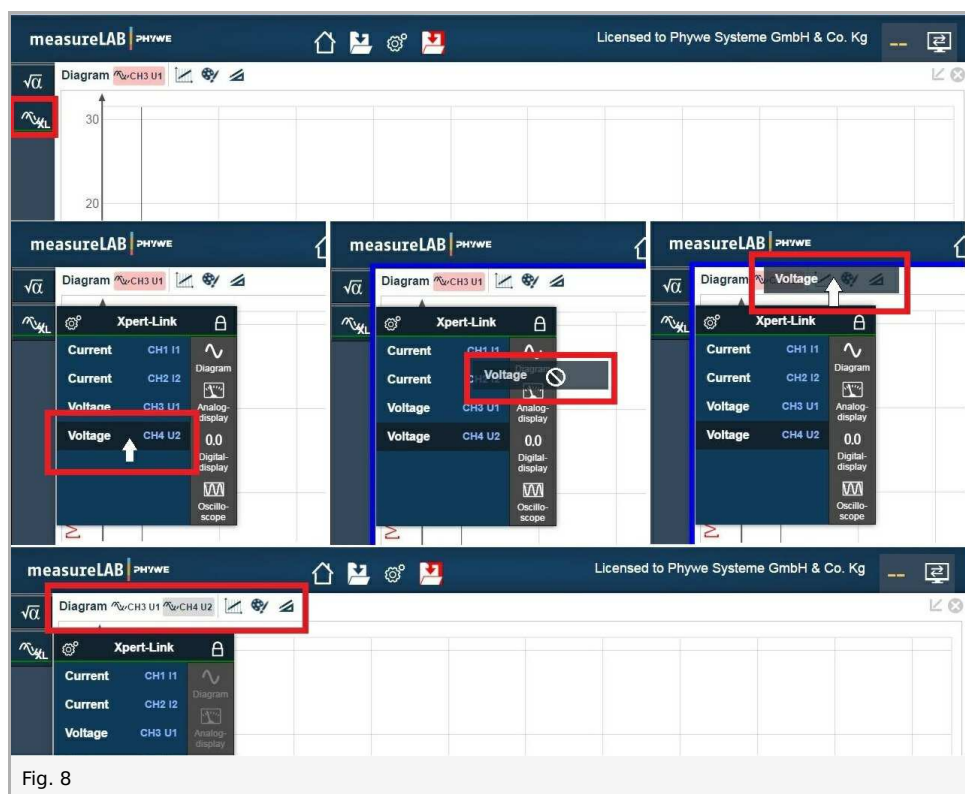




Fig. 8

Select the colors of the channel:

Click with the **left** mouse button on the channel button (for example ) and then on the button painter's palette  . Follow the instructions to fig. 11.

If you start with the recording of the values of a channel in a diagram...

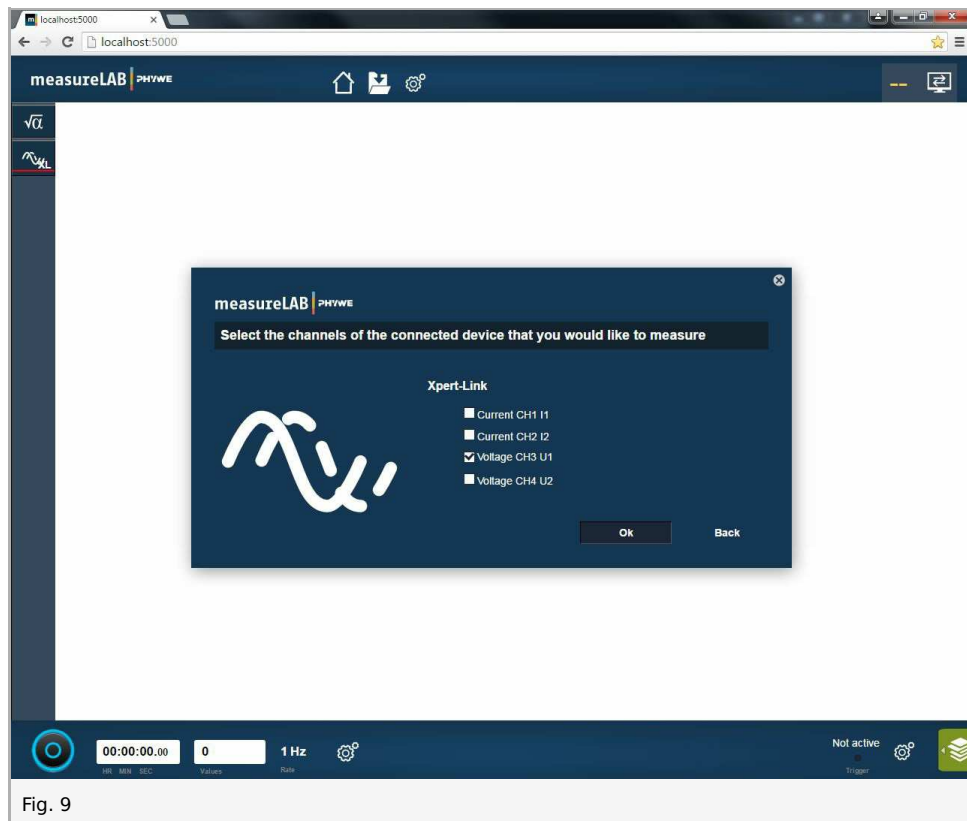




Fig. 9

... you can add more measuring channels and diagrams by clicking on the button .

Select the channel by clicking on the channel button (fig. 10: example with **Voltage CH4 U2**) and click on the button .

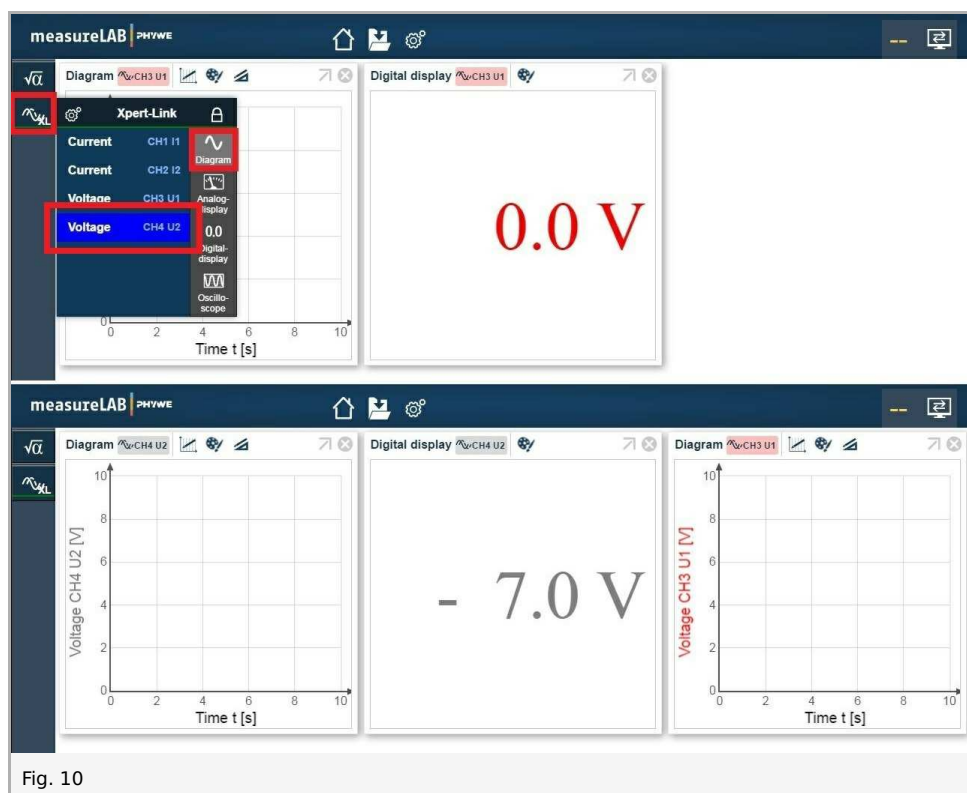


Fig. 10

Hint: You can change the measured channels in any diagram button as explained above.

Move the diagrams into a better position and maximize them by clicking in the chart button: press and hold the left mouse button and move the mouse.
Select the colours of the charts by clicking on the button painter's palette .

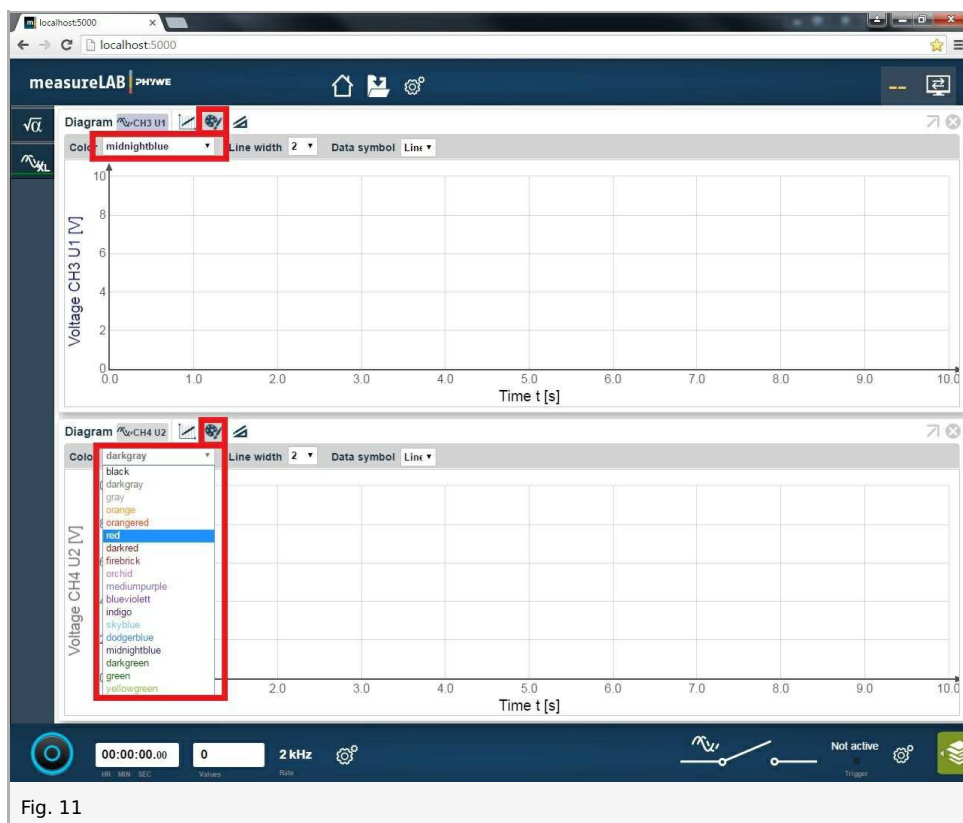


Fig. 11

You will see:

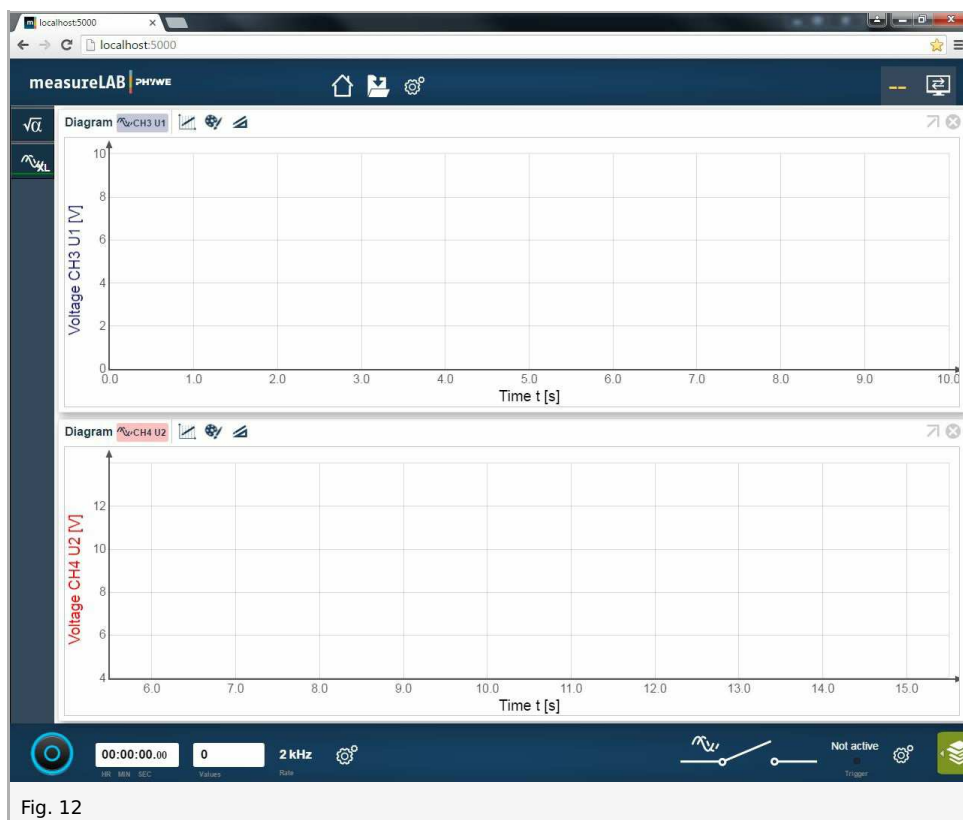





Fig. 12

Start the measurement by clicking on the button below left: 
 Stop the measurement by clicking on the button below left again: 



Settings

Open the settings by clicking on the cogwheel button 
 Select the language and click on Apply:

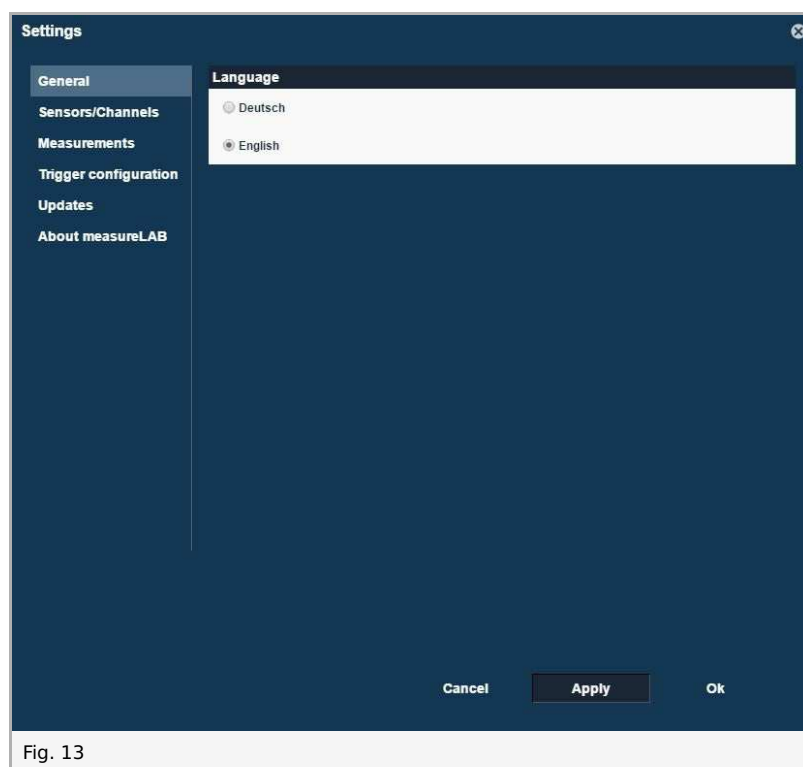


Fig. 13

Select the channel by clicking on the grey channel button. Select the range of the measured channels and **click ever on Apply each time you change channel settings.**

Notes:

- Any signal can be measured on any channel.
- AP (yellow socket E) will be measured in the range of 100 mV.
- For any other values (EPSP, IPSP, stimulation - yellow sockets 0, 1, 2, 3 and I) select the range 10 V.
- For averaging always use 10 values.

Below: an example with the AP-signal on channel 3 (fig. 14) and the EPSP-signal on channel 4 (fig. 15).

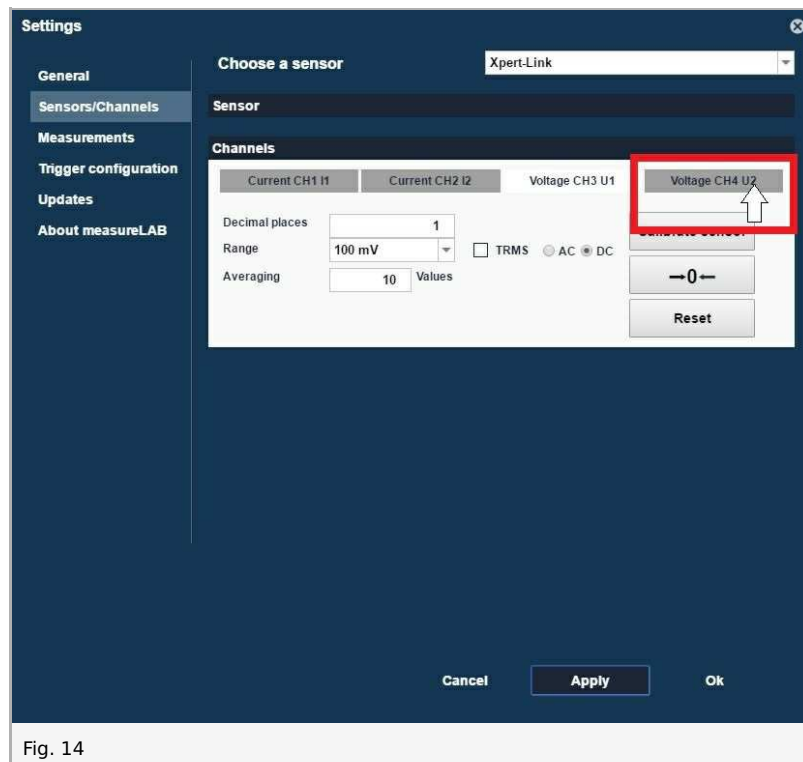


Fig. 14

You will see:

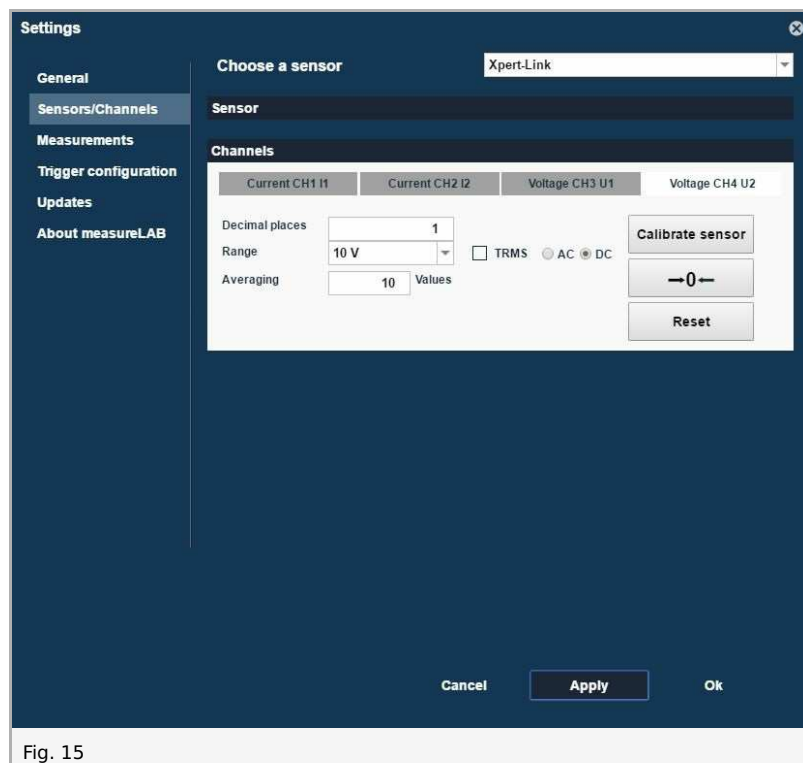


Fig. 15

Perform the experiments with Neurosimulator at 10 kHz when EPSP, IPSP and signal intensity are measured. When **AP** is measured perform the experiment at **100 kHz** to get a better resolution. Performance at 50 kHz and 10 kHz is possible, but a magnification and accurate observation of action potentials is not possible. Select the frequency and click on Apply:

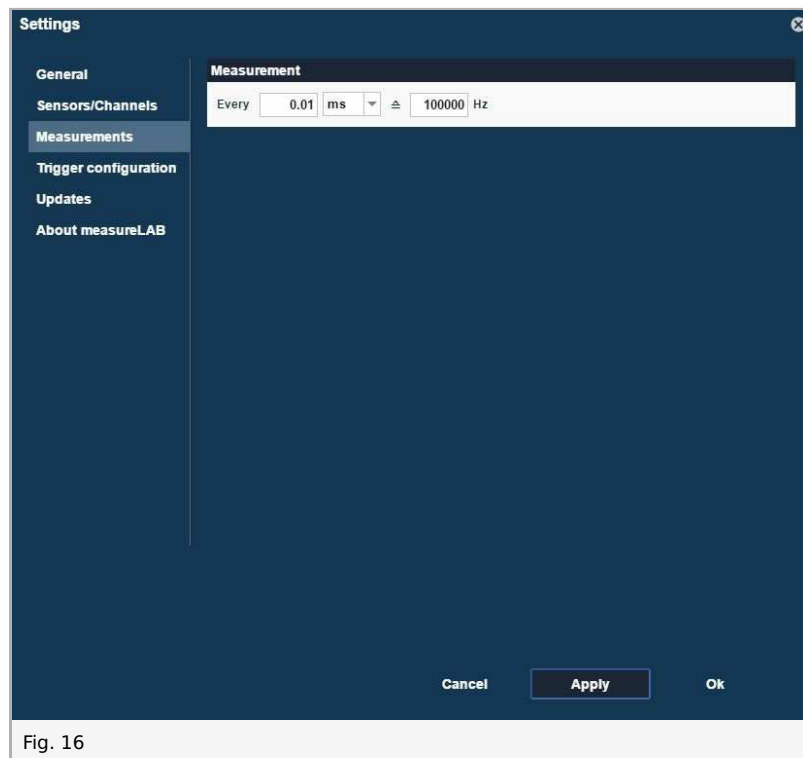


Fig. 16

Confirm with OK:

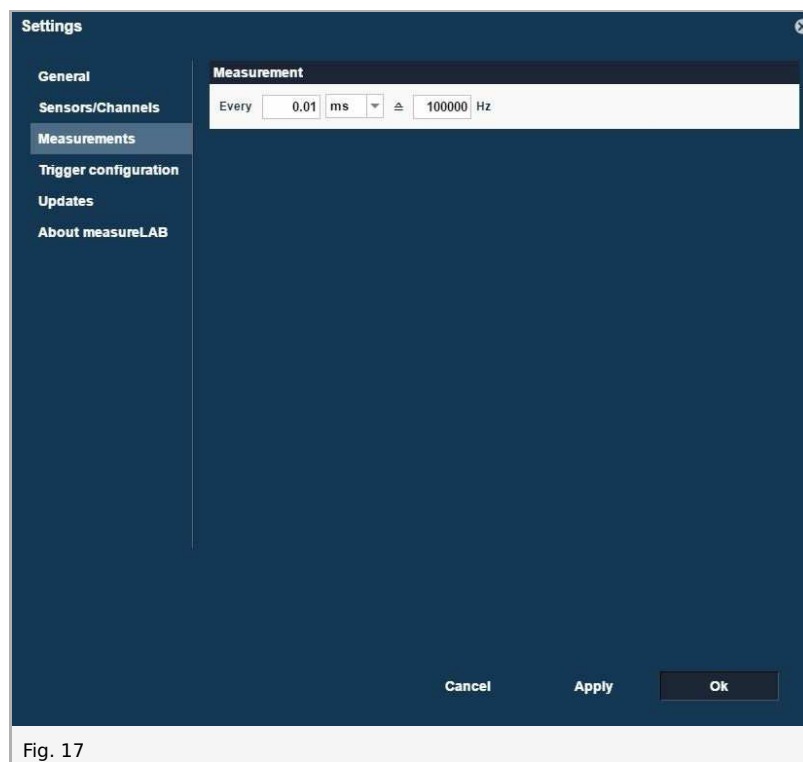
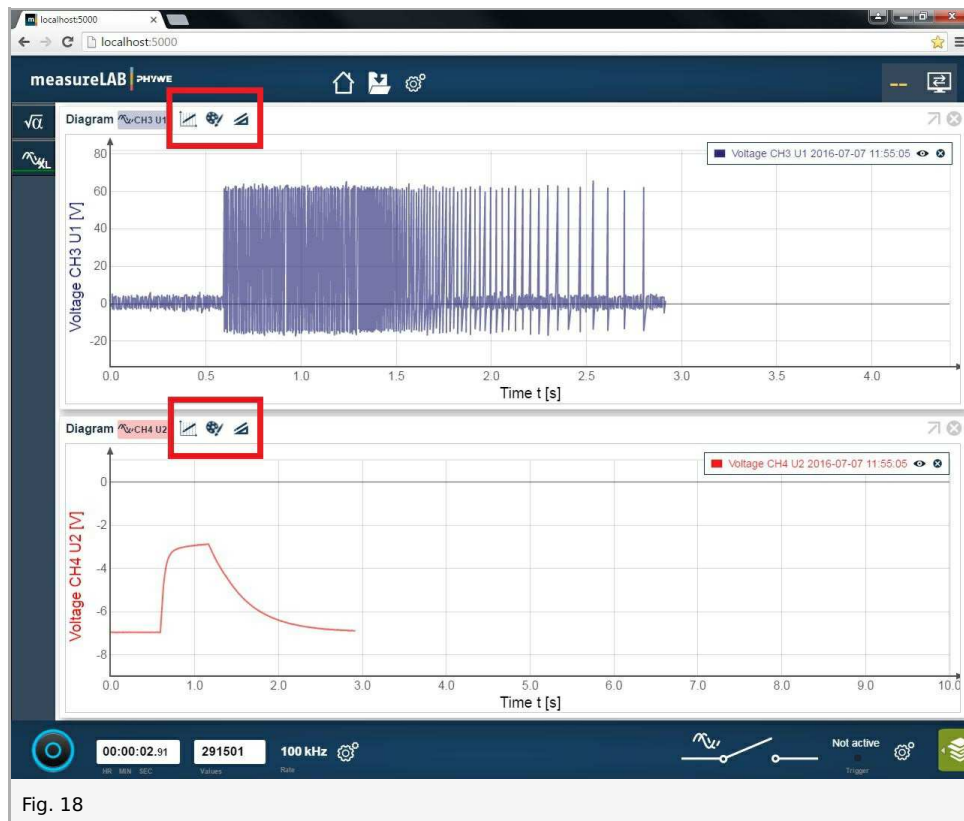


Fig. 17

Edit chart

1. Edit diagram

You can edit the diagram with the tool functions:



The arrow symbol optimizes the visible area:

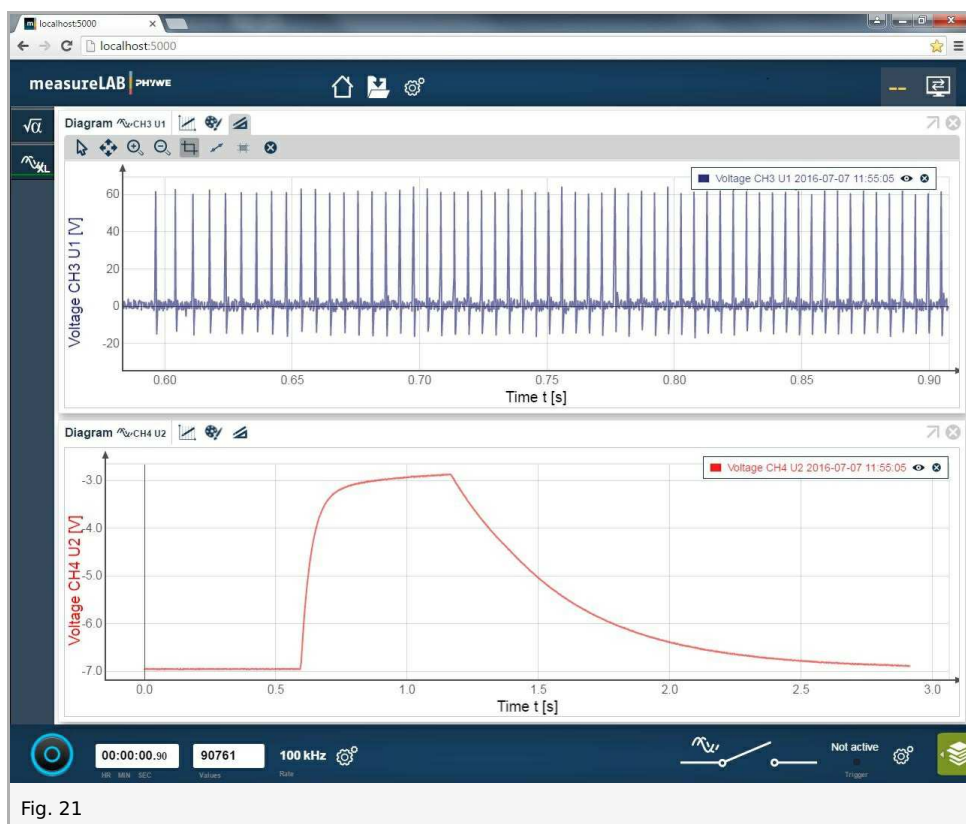
You will receive the following diagram:



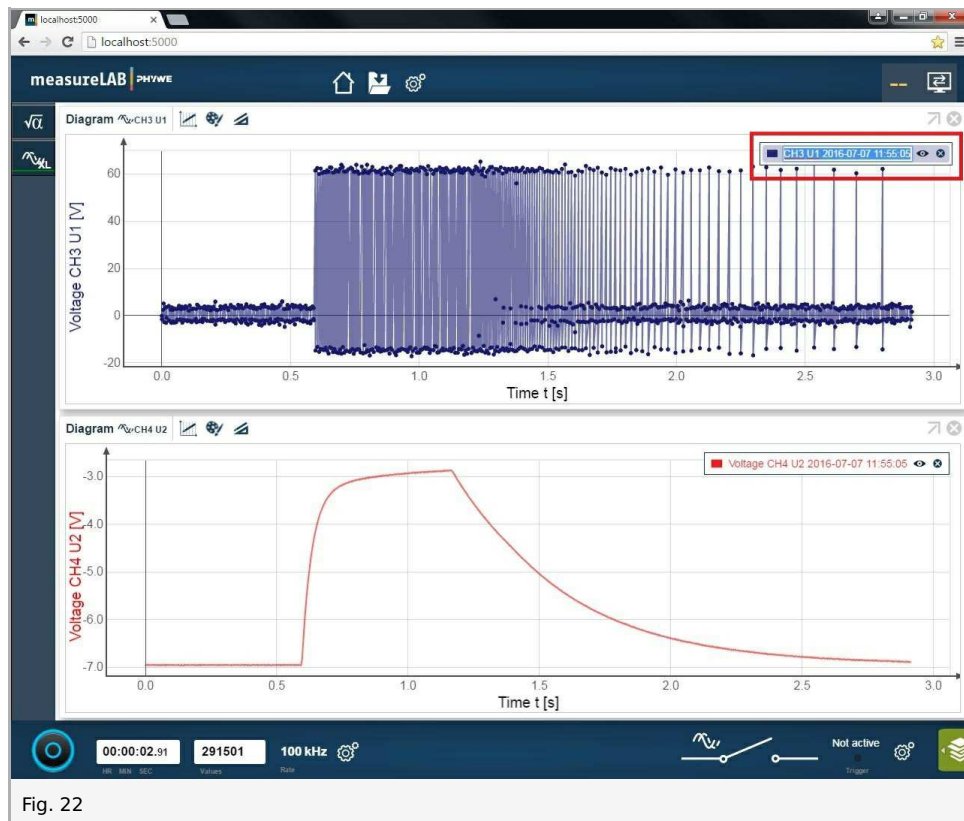
If you click on the rectangle button, you can select and enlarge an area:



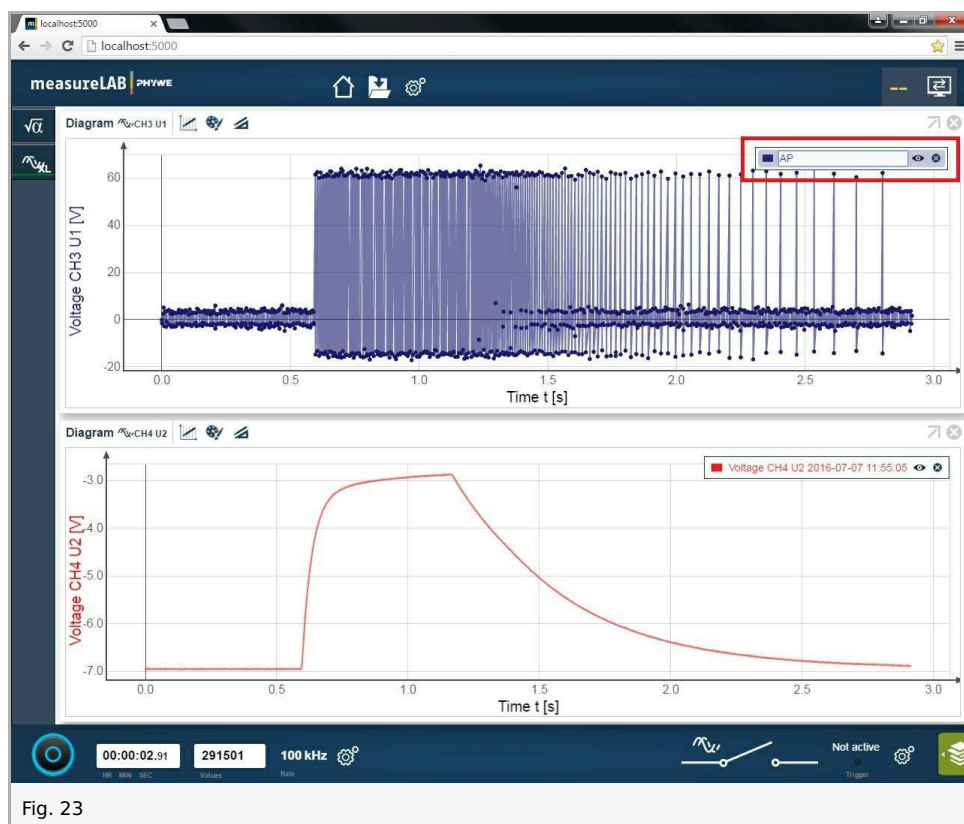
You will receive the following diagram:



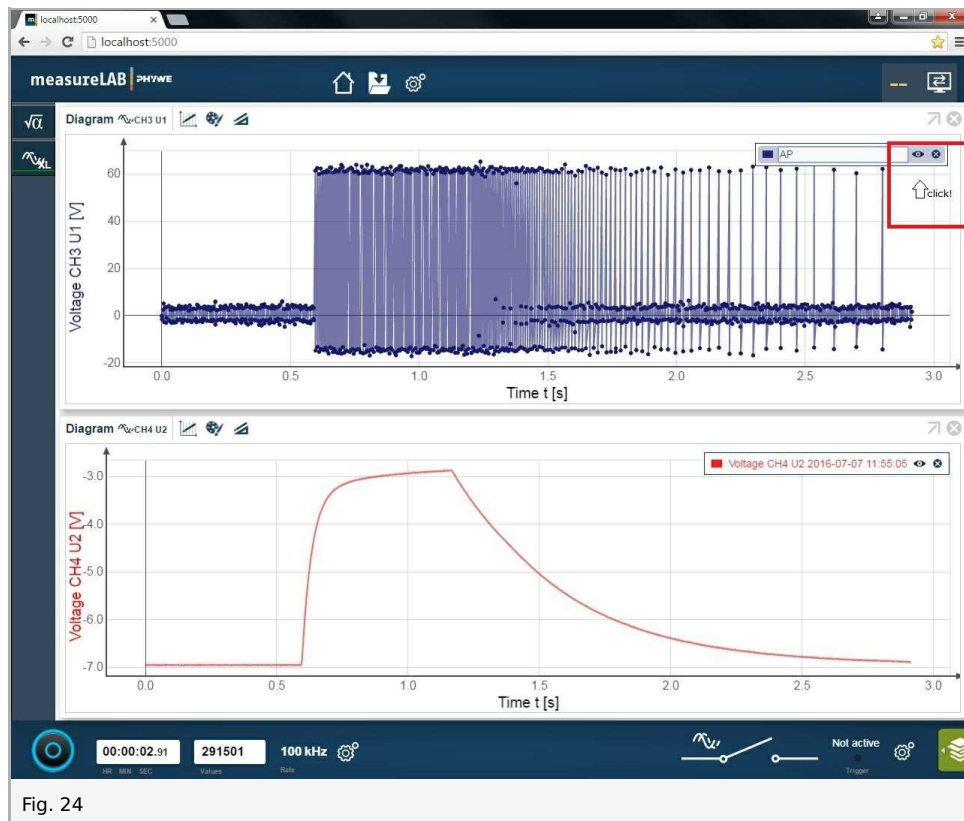
You change the caption by double clicking on the caption field:



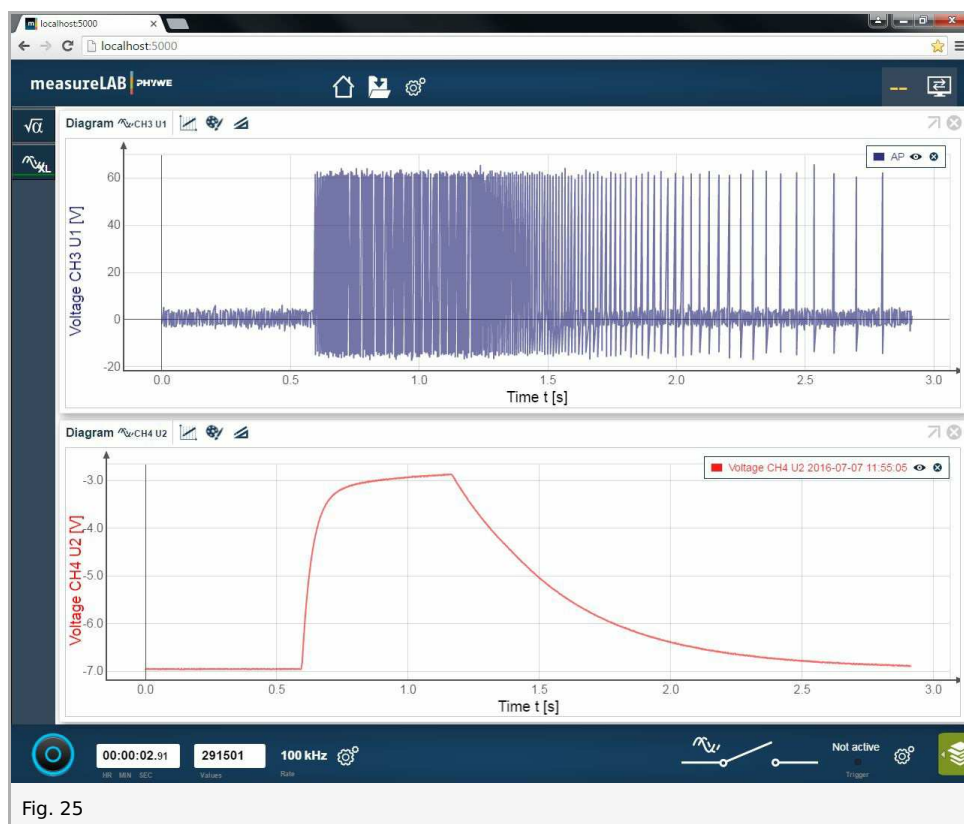
Enter the name manually:



Click on the chart field to save the new name:



You will receive the following diagram:



Change the caption of the second diagram in the same way.

2. How to read values exactly?

The most convenient way to read the values is when you use the mouse pointer. Move the mouse pointer along the graph line:



Virtual Channel

With the Virtual Channel will you can set the values which you expect when you would measure using a live nerve cell. Open Virtual Channel under settings:

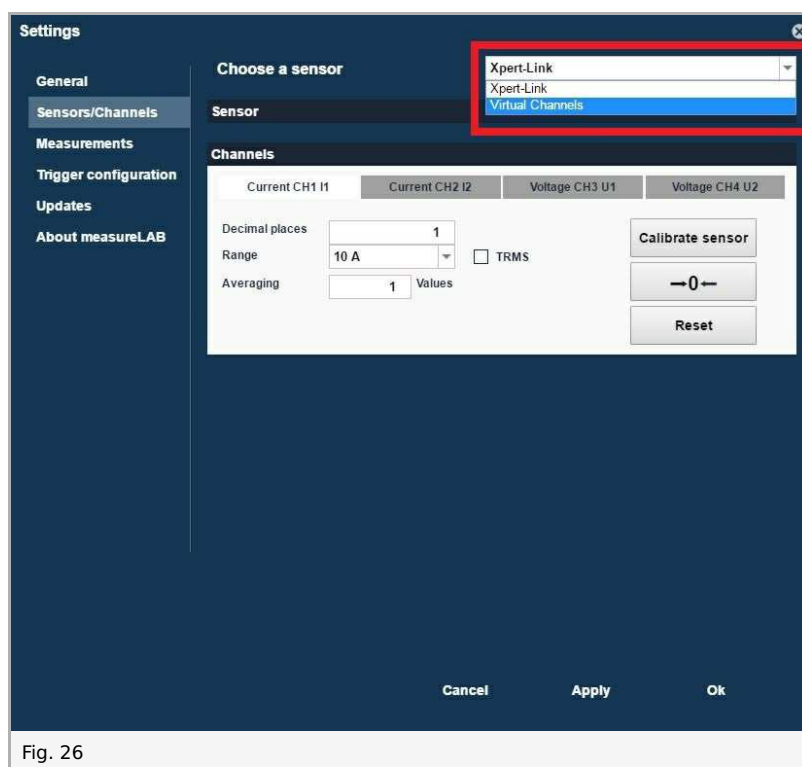


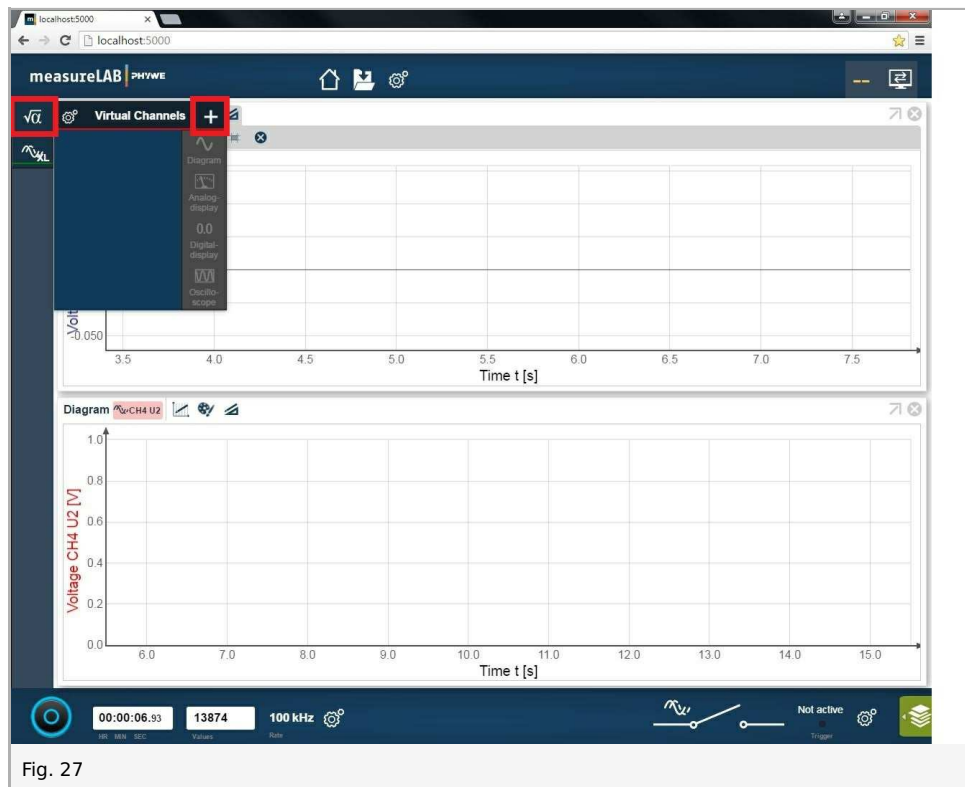
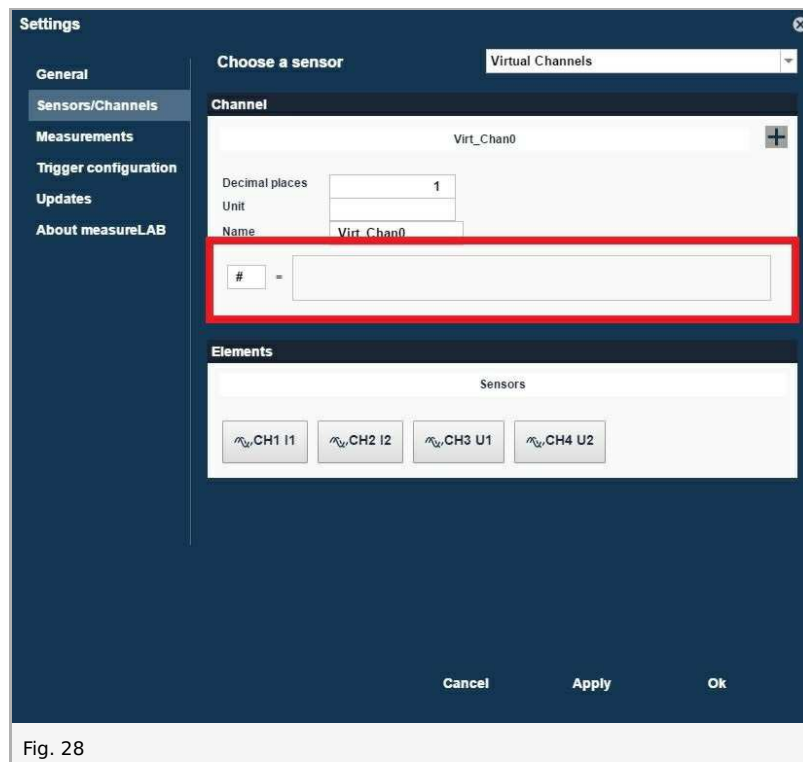


Fig. 26

... or by clicking on button  and then on button .



You create formulae in the Virtual Channel:



Enter the values and click on Apply:

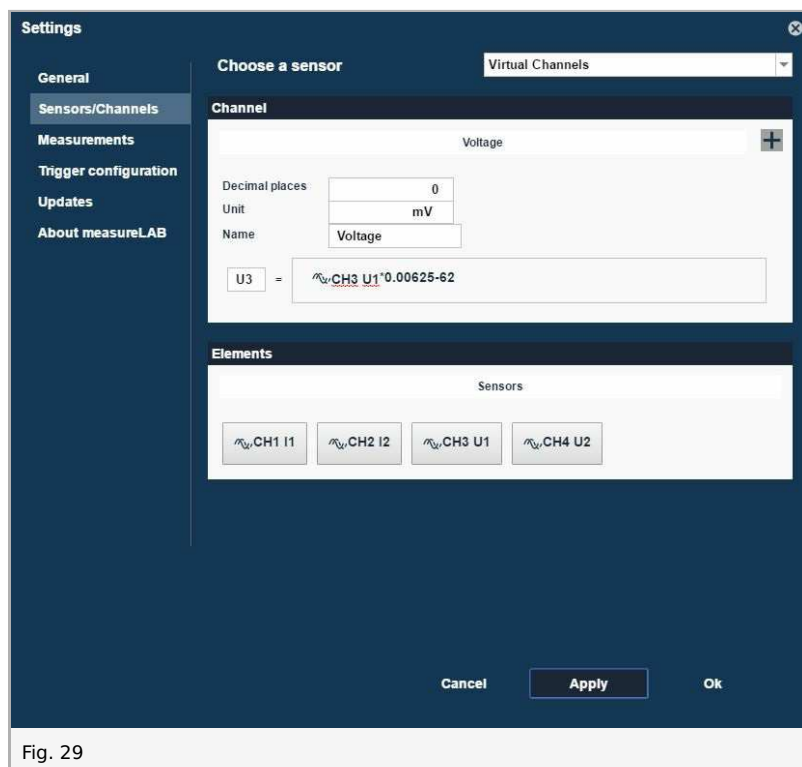



Fig. 29

You can define a second virtual channel. Click on , enter the values and click on Apply. You will receive the following window:

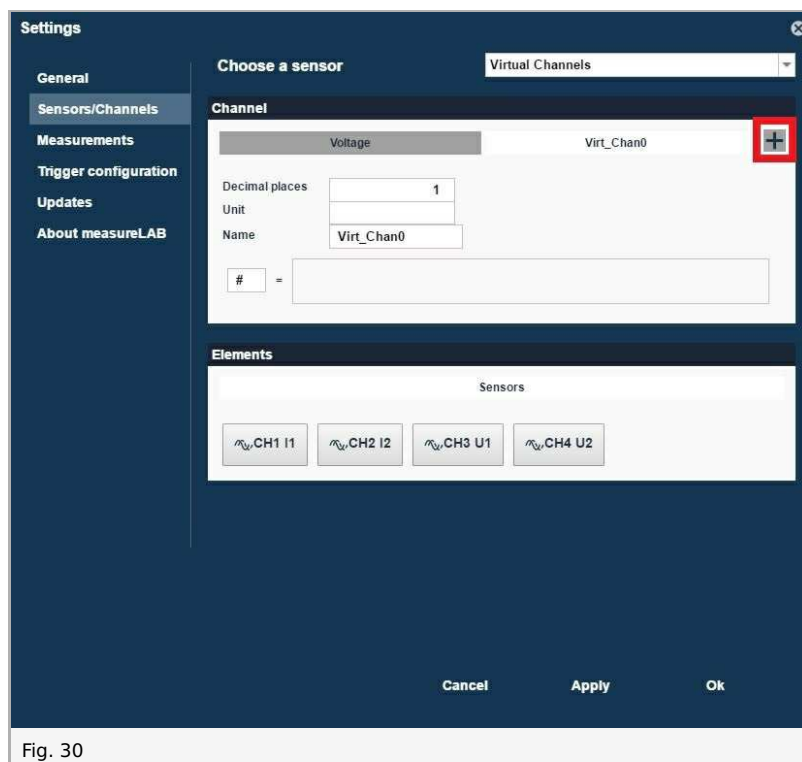


Fig. 30

Additional channel with additional material

1 value can be measured additionally by using a Cobra4 Sensor Port. To use the sensor port, a Cobra4 Sensor-Unit Electricity (12644-00) and a Cobra4 Xpert-Connect (12625-01) are needed additionally.

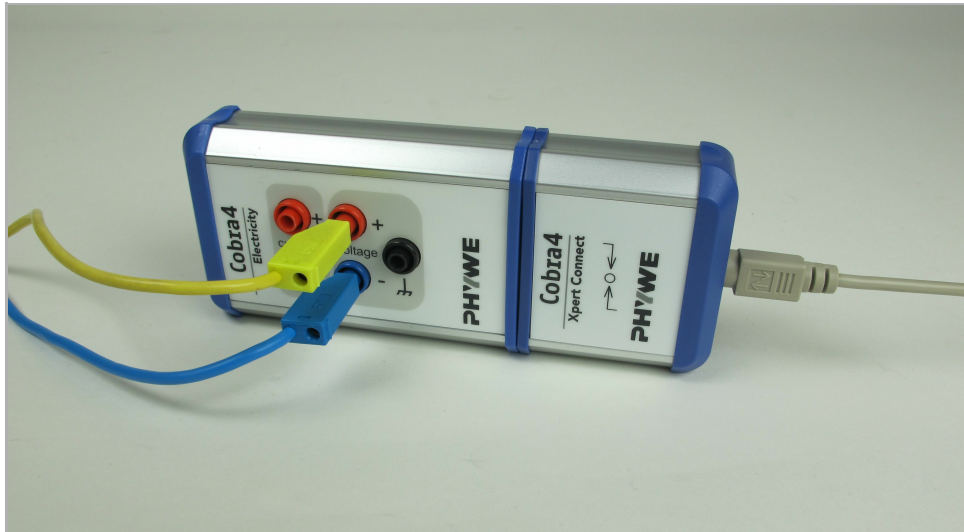


Fig. 31

Setup

Attach the Sensor-Unit on Xpert-Connect as per figure below:

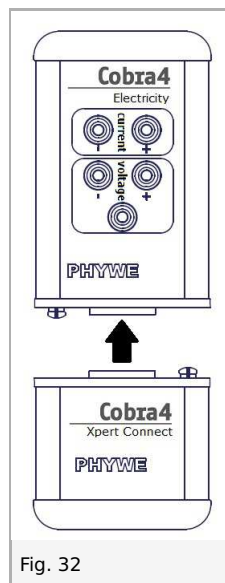


Fig. 32

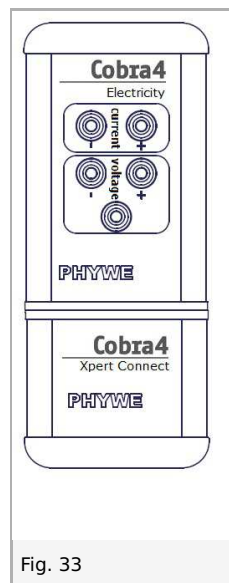


Fig. 33

Plug the Xpert-Connect into the Cobra4 Sensor Port of Xpert-Link using the cable provided with the Xpert-Connect. Plug a yellow and a blue cable into the sockets for voltage measurement:

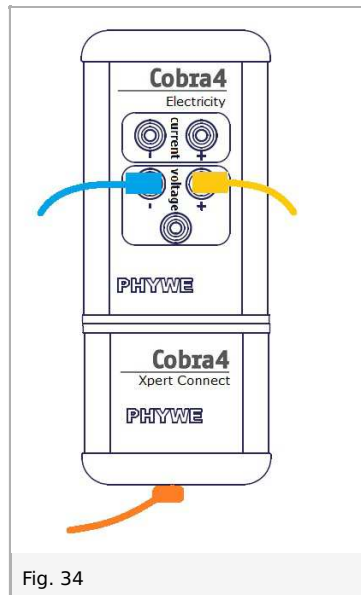


Fig. 34

The signal is measured with the yellow and the blue cable. Plug the the free end of the yellow cable into the yellow socket of your choice (Neurosimulator 0, 1, 2, 3 or Operating unit for neurosimulator I, E). Plug the free end of the blue cable in the ground (white socket of the Neurosimulator / socket on other blue cable).

For synchronising the Xpert-Connect with the Xpert-Link use the trigger at the Xpert-Link. It is set up as per figure below (black cable; applies to one and two additional channels).

You see an example with one additional channel below (in the left Cobra Sensor Port).

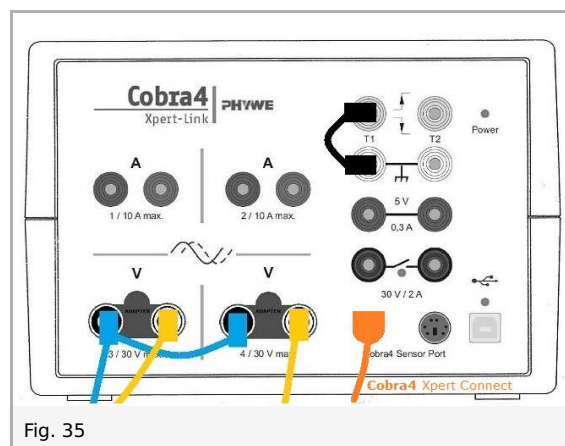


Fig. 35

Instructions for measureLAB

Open Sensor-Unit Electricity Virtual Channel under settings (see chapter Virtual Channel).
Select the range of the measured channel Voltage U and click on Apply for any channel modification.

Notes:

- AP (yellow socket E) will be measured in the range of -30000..30000 mV.
- For any other values (EPSP, IPSP, stimulation - yellow sockets 0, 1, 2, 3 and I) select the range -30..30 V:

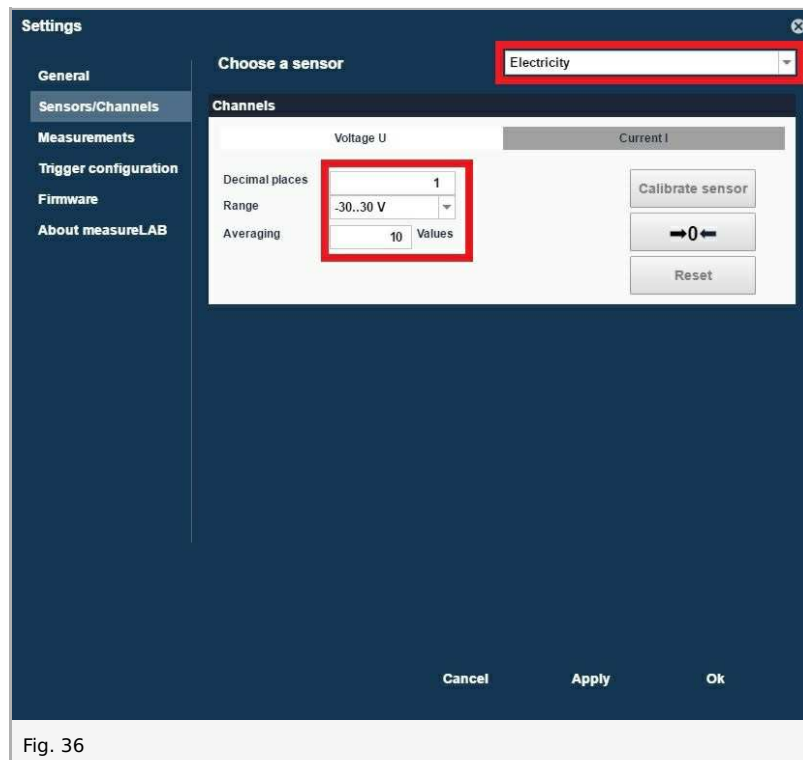


Fig. 36

Select the trigger configuration rising for T1:

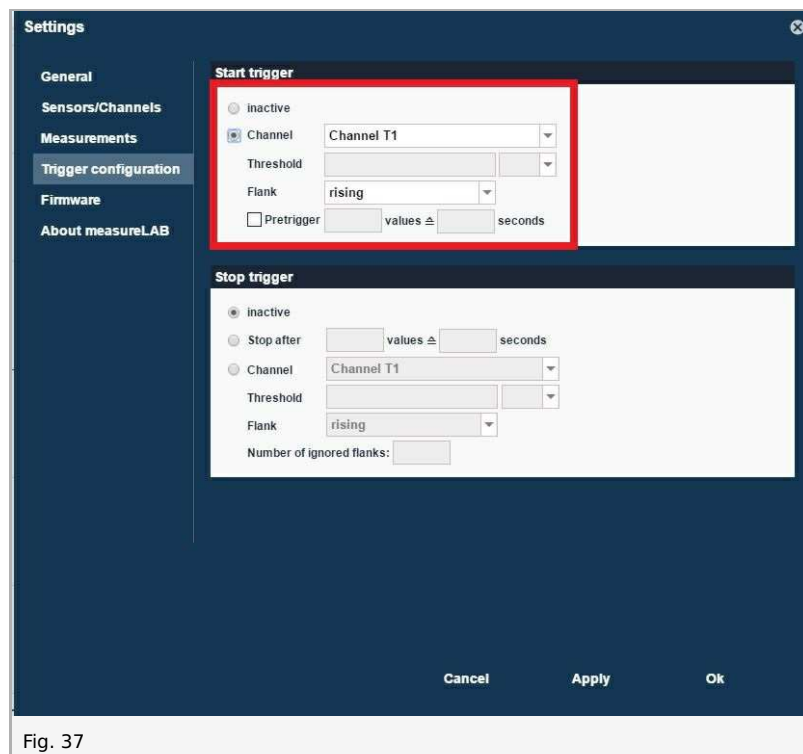

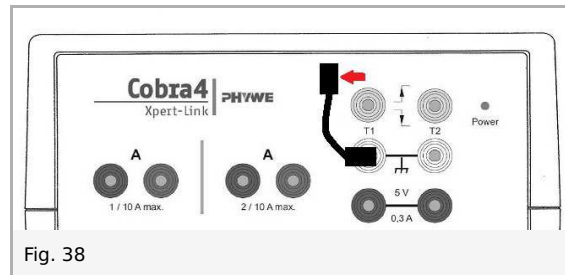


Fig. 37

Measurement

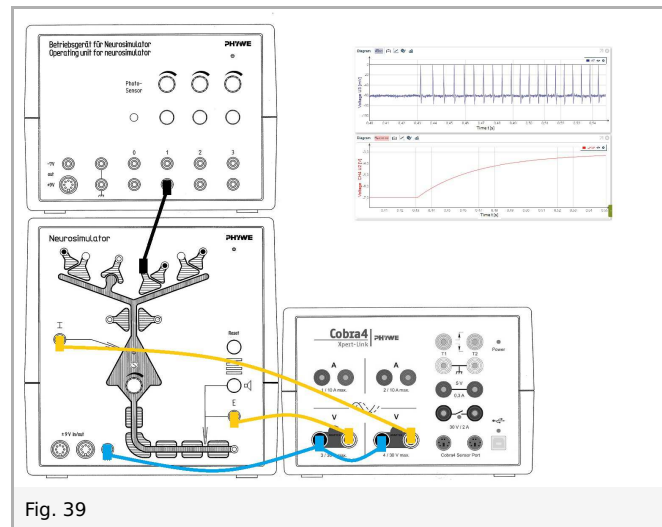
- Start the measurement by clicking on the start button: 
- Before the start of the experiment: remove the black cable on T1 (Xpert-Link) because Xpert-Connect and Xpert-Link are now synchronized.
- Start the experiment according to the instructions.



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1. Membrane potential and action potential, membrane time constant

Introduction



In this set of experiments membrane potential and action potential as well as membrane time constant are studied. Membrane depolarization must reach or exceed a certain value for action potentials to be released – the firing threshold. This therefore corresponds to a limiting potential value. With many nerve cells, the value of the firing threshold is around -50 mV, but it differs from cell type to cell type. Even with individual cells, it does not have a constant value. On the contrary, the value of it depends on a series of intracellular and extracellular factors.

Immediately after the release of an action potential, for example, the firing threshold is greatly increased and only goes back to its original value after a time, typically within 2-10 ms (refractory period). Further to this, the threshold can be influenced by external modulators that act directly or indirectly on the membrane pores, as well as by changes in the internal or external environment.

Action potential arises by influx of sodium ions through the sodium channels of the nerve cells. Stimulus movement along the axon occurs due to the consecutive influx of sodium ions along its cell membrane.

The measurement method of this experiment differs from that of the other experiments so that action potential can be displayed (together with membrane potential). In the other experiments (with the exception of the experiments dealing with the excitatory synapse) presynaptic signal strength is shown instead, along with membrane potential.

Set-up and procedure

At first we will measure that the action potential (AP) depends on the excitatory postsynaptic potential (EPSP) by varying different parameters.

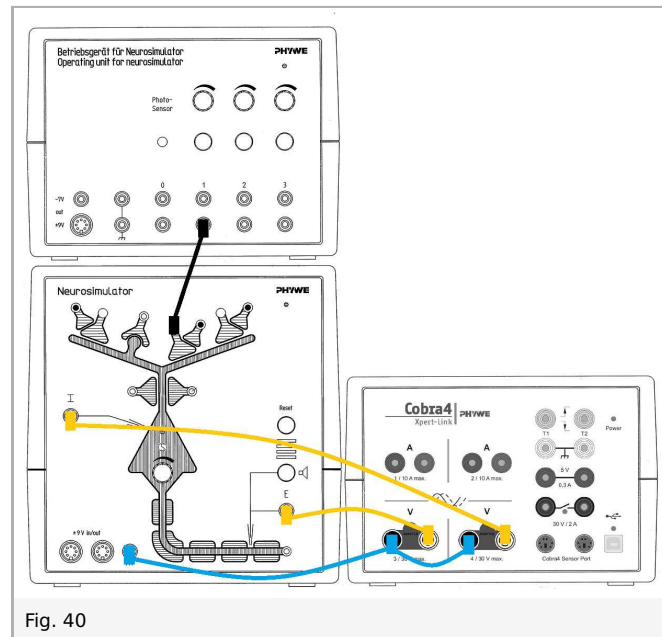
Notes: The knobs are infinitely adjustable. However, specific values (e.g. 0%, 33%, 66%, 100%) are given for clear description of the experiments. Knobs for stimulation intensity 2 and 3 are not needed in this experiment.

Voltage measurements can be taken on both Xpert-Link voltage channels (CH3 and CH4). Wiring diagrams are given for the experiments.

Part 1 of experiment 1 and 2 are identical and therefore do not need to be performed twice.

Connect the Xpert-Link to the PC.

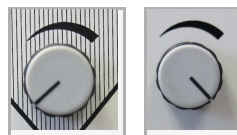
The experiment is set up as per Fig. 40. Two BNC-adapters (plug/socket 4 mm) are needed for voltage measurement.



1.1. Experiment 1, Part 1: Different threshold

Neurostimulator, knob threshold: 0%

Operating unit, knob stimulation intensity 1: 100%

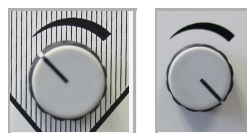


- Start measurement in the measurement window.
- Press the stimulation button 1 for approximately one second.
- Finish the measurement as soon as the voltage of EPSP (now CH4) has reached the initial value.
- Save and evaluate the results. (See examples in "Evaluation".)

1.2. Experiment 1, Part 2: Different threshold

Neurostimulator, knob threshold: 33%

Operating unit, knob stimulation intensity 1: 100%

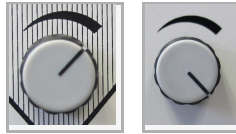


Repeat the measurement and evaluation of part 1.

1.3. Experiment 1, Part 3: Different threshold

Neurostimulator, knob threshold: 66%

Operating unit, knob stimulation intensity 1: 100%



Repeat the measurement and evaluation of part 1.

2.1. Experiment 2, Part 1: Different stimulation intensity

See above: Part 1 of experiment 1 and 2 are identical.

You can use the same diagram.

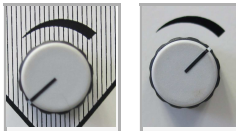
Neurosimulator, knob threshold: 0%

Operating unit, knob stimulation intensity 1: 100%

2.2. Experiment 2, Part 2: Different stimulation intensity

Neurosimulator, knob threshold: 0%

Operating unit, knob stimulation intensity 1: 66%

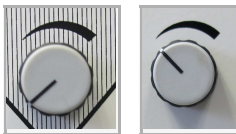


Repeat the measurement and evaluation of part 1.

2.3. Experiment 2, Part 3: Different stimulation intensity

Neurosimulator, knob threshold: 0%

Operating unit, knob stimulation intensity 1: 33%



Repeat the measurement and evaluation of part 1.

Results and evaluation

Results

The threshold potential is the critical level to which a membrane potential must be depolarized to initiate an action potential. The threshold value can vary according to numerous factors like the ion conductances of sodium or potassium, the diameter of the axon as well as the density and properties of the sodium channels.

Shape of typical action potentials (enlarged section of a diagram, see below):
Measurement at 100 kHz in the range of 100 mV.

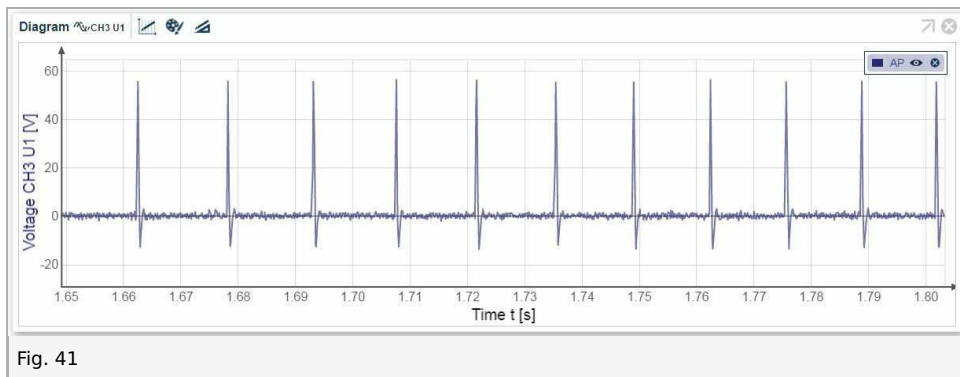


Fig. 41

You will receive the following diagrams:

1.1. Experiment 1, Part 1: Different threshold (0%)

1.1.1. In case you have recorded data of both channels together in a diagram:

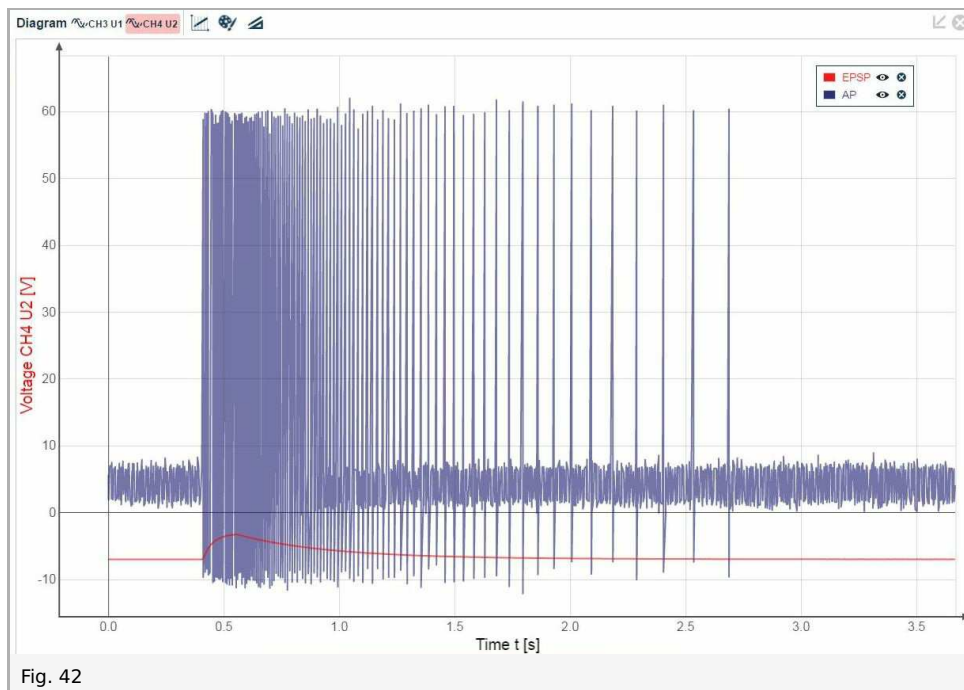


Fig. 42

1.1.2. In case you have recorded data of each channel in separate diagrams (example with Virtual Channel for CH3):

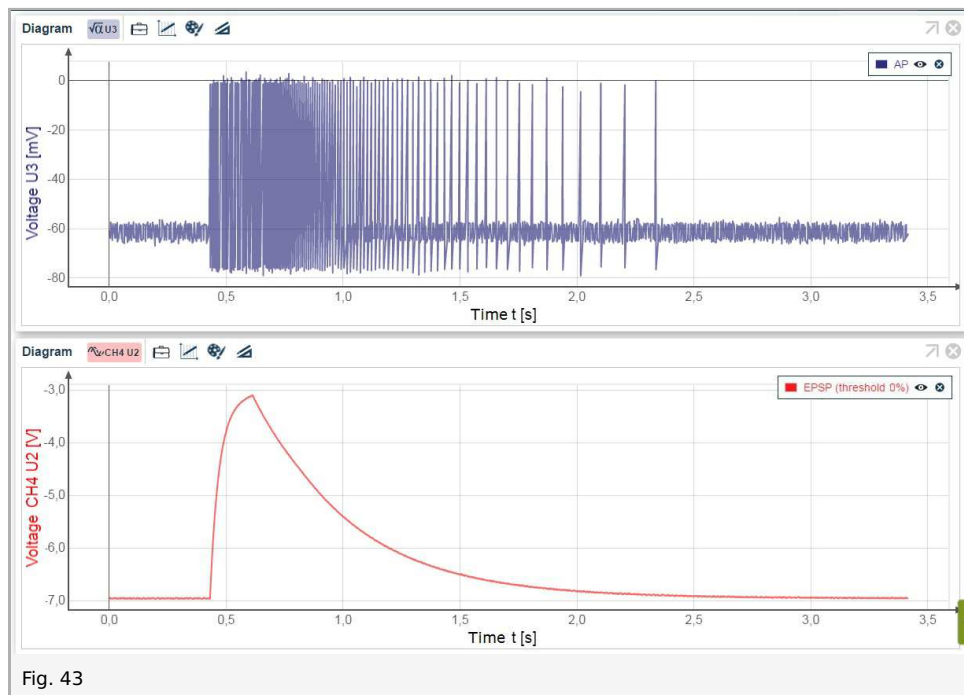


Fig. 43

1.2. Experiment 1, Part 2: Different threshold (33%)

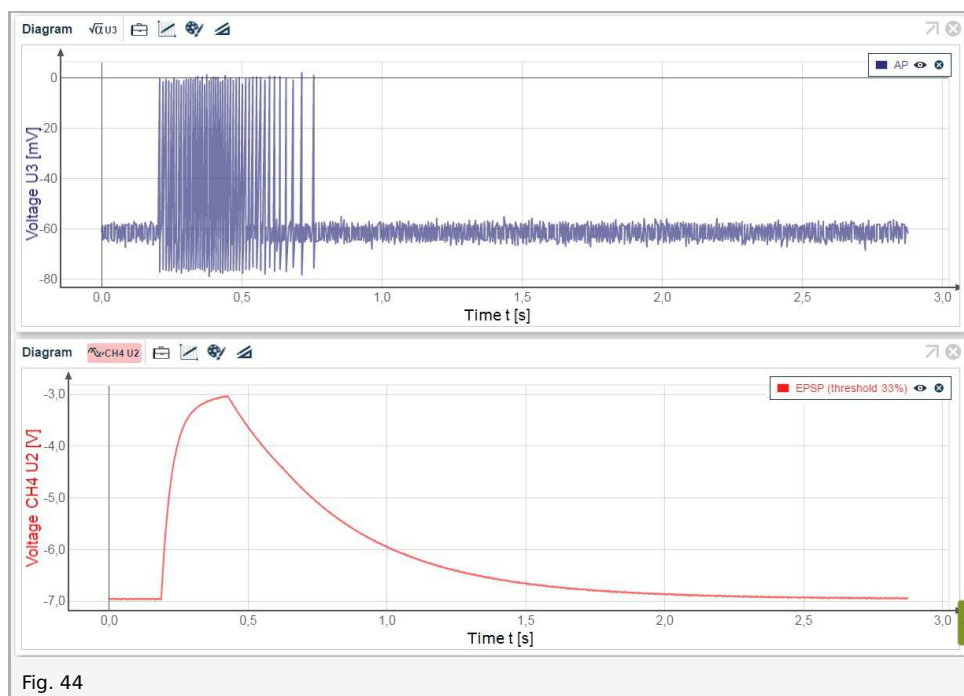


Fig. 44

1.3. Experiment 1, Part 3: Different threshold (66%)

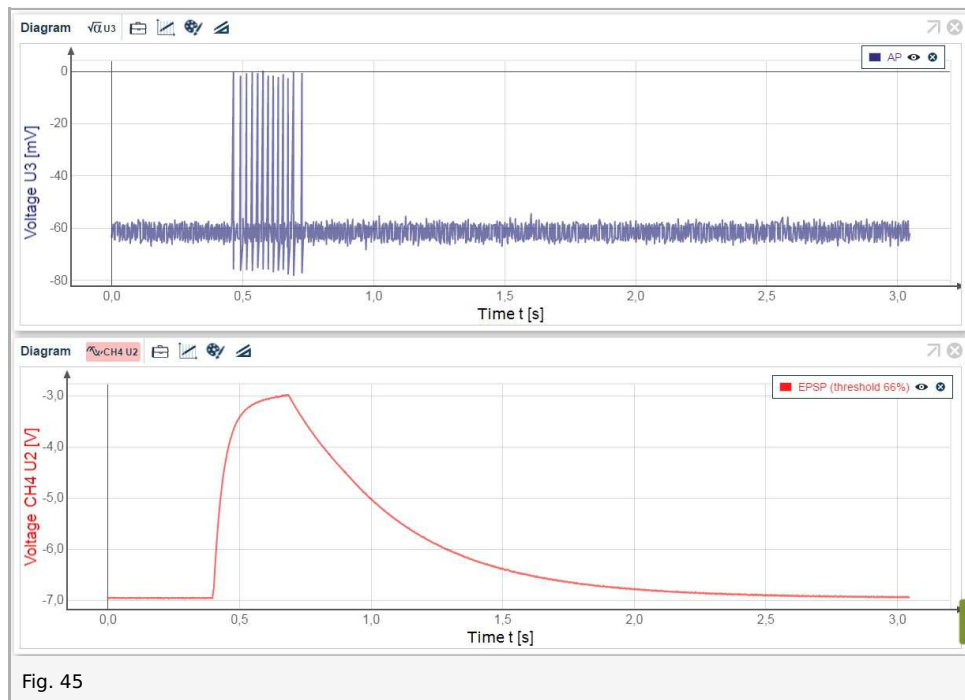


Fig. 45

2.1. Experiment 2, Part 1: Different stimulation intensity (100%)

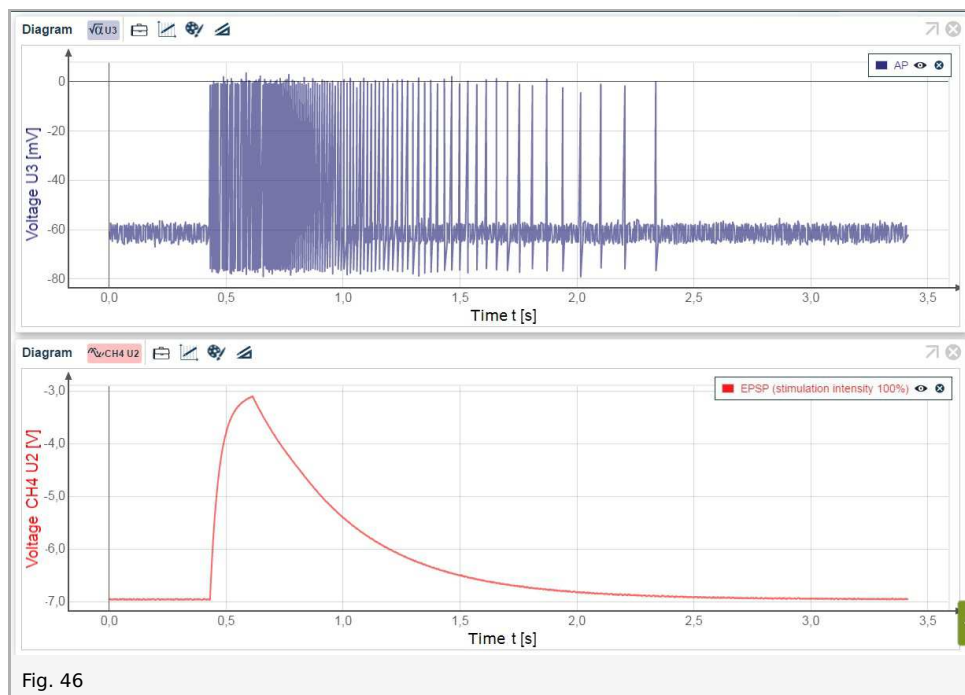
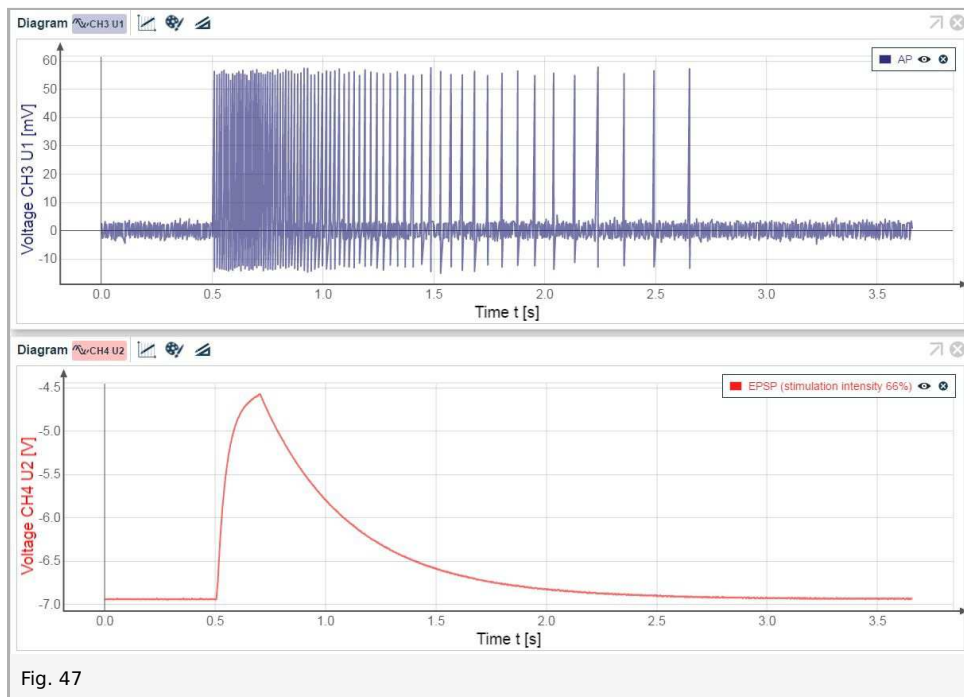
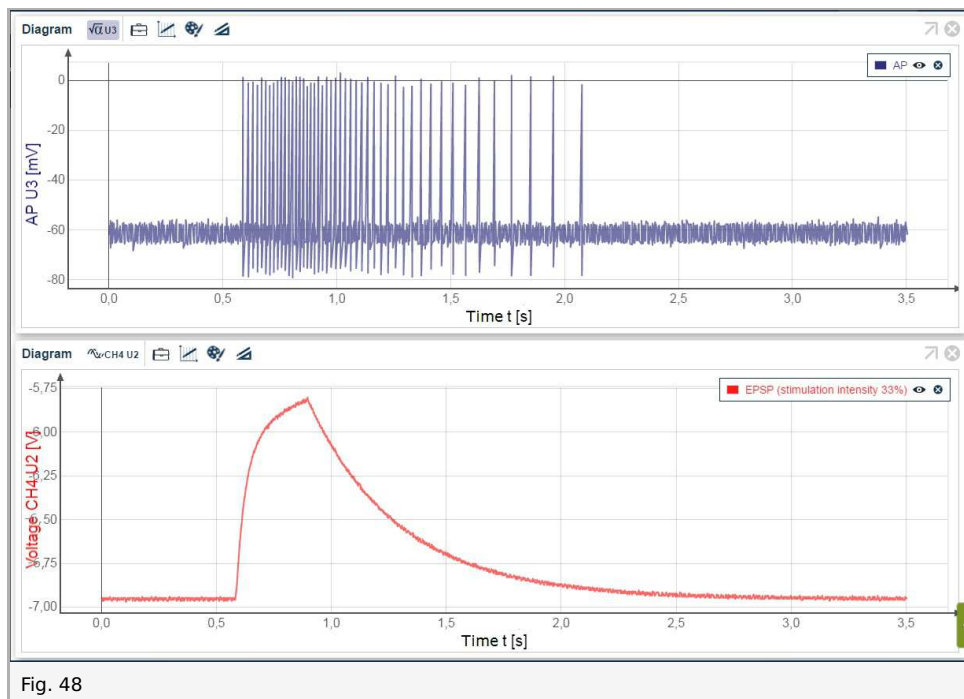


Fig. 46

2.2. Experiment 2, Part 2: Different stimulation intensity (66%)



2.3. Experiment 2, Part 3: Different stimulation intensity (33%)



Evaluation: 1. Threshold

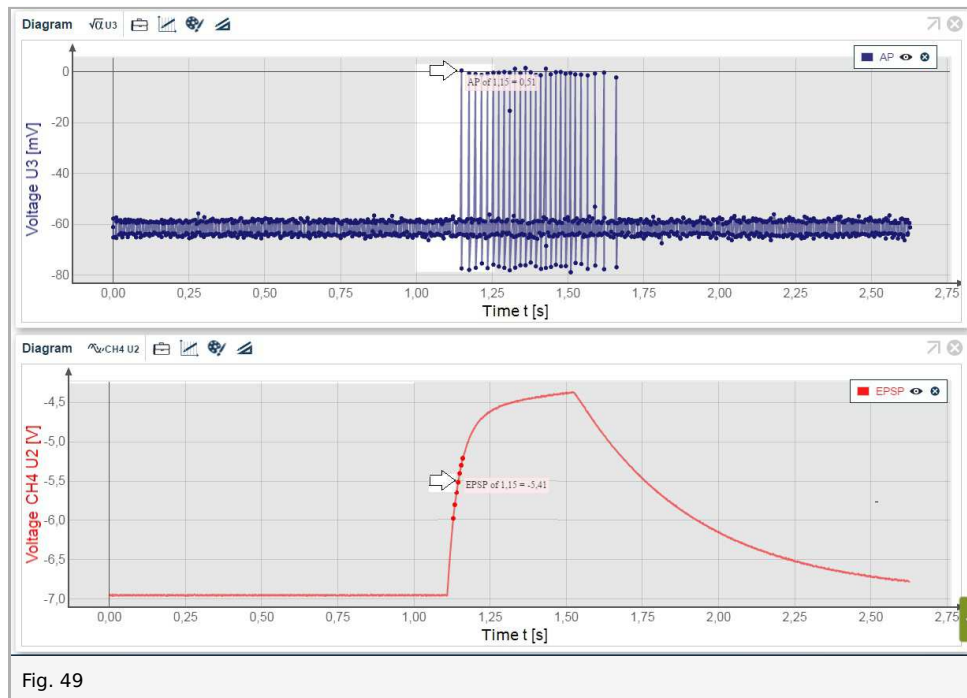
Hint: The most convenient way to read the values is when you use the mouse pointer. Move the mouse pointer along the graph line, as shown in fig. 49.

1. Threshold

Determine the threshold in any diagram as follows:

- Read the time of the peak of the first AP.
- Read the voltage of EPSP at this time.

Example:



Time of the first AP = 1.15 s. Voltage at time 1.15 s = -5.41 V (see diagram above).

The Neurosimulator measures an intracellular potential which is 100 times the potential in a real nerve cell:

$$\text{Threshold} = -5.41 \text{ V} / 100 = -54.1 \text{ mV}$$

Compare the threshold of all measurements.

Evaluation: 2. Membrane time constant for the rise sets

Hint: The most convenient way to read the values is when you use the mouse pointer. Move the mouse pointer along the graph line, as shown in Fig. 50.

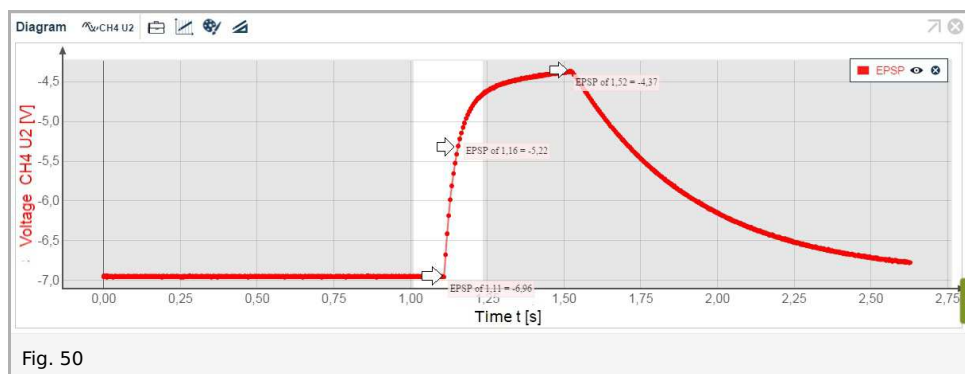
2. Membrane time constant for the rise sets

This time constant is the time elapsed after 63% of U_{\max} has been reached.

Determine the membrane time constant in any diagram as follows:

- Read the voltage and the time of $EPSP_{\max}$ and $EPSP_{\min}$.
- Calculate the voltage of membrane time constant ($EPSP_{\text{constant}}$).
- Read the time at $EPSP_{\text{constant}}$.
- Calculate the membrane time constant from the time of $EPSP_{\max}$ and $EPSP_{\text{constant}}$.

Example:



$EPSP_{\max}$	1.11 s	-6.96 V	-69.6 mV
$EPSP_{\min}$	1.52 s	-4.37 V	-43.7 mV
$EPSP_{\text{constant}}$	1.16 s	-5.22 V	-52.2 mV

$$(-69.6) - (-43.7) = -25.9 \text{ mV} = 100\%$$

$$66\% = 17.01 \text{ mV}$$

$$(-69.6) - (-17.01) = -52.59 \text{ mV} = EPSP_{\text{constant}}$$

Nearest measurement point: EPSP of 1.16 s = -5.22 V which is equivalent to -52.2 mV in a real nerve cell.

$$\text{Membrane time constant for the rise sets} = 1.16 \text{ s} - 1.11 \text{ s} = 0.05 \text{ s} = 50 \text{ ms}$$

Compare the membrane time constants of all measurements.

Evaluation: 3. Membrane time constant for the fall sets

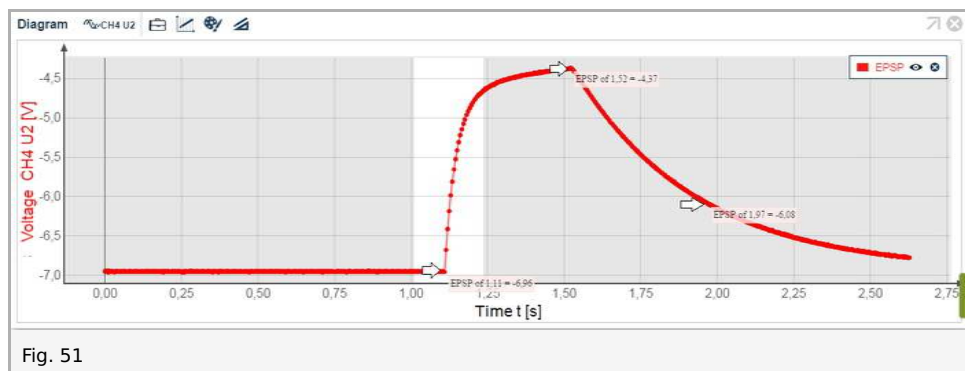
Hint: The most convenient way to read the values is when you use the mouse pointer. Move the mouse pointer along the graph line, as shown in Fig. 51.

3. Membrane time constant for the fall sets

Determine the membrane time constant in any diagram as follows:

- Read the voltage and the time of EPSP_{max} and EPSP_{min}.
- Calculate the voltage of membrane time constant (EPSP_{constant}).
- Read the time at EPSP_{constant}.
- Calculate the membrane time constant from the time of EPSP_{max} and EPSP_{constant}.

Example:



EPSP _{max}	3.50 s	-6.96 V	-69.6 mV
EPSP _{min}	1.52 s	-4.37 V	-43.7 mV
EPSP _{constant}	1.97 s	-6.08 V	-60.8 mV

$$(-69.6) - (-43.7) = -25.9 \text{ mV} = 100\%$$

$$66\% = 17.01 \text{ mV}$$

$$(-43.7) + (-17.01) = -60.71 \text{ mV} = \text{EPSP}_{\text{constant}}$$

Nearest measurement point: EPSP of 1.97 s = -6.08 V which is equivalent to -60.8 mV in a real nerve cell.

$$\text{Membrane time constant for the fall sets} = 1.97 \text{ s} - 1.52 \text{ s} = 0.45 \text{ s} = 450 \text{ ms}$$

Compare the membrane time constants of all measurements.

2. Excitatory synapse

Introduction

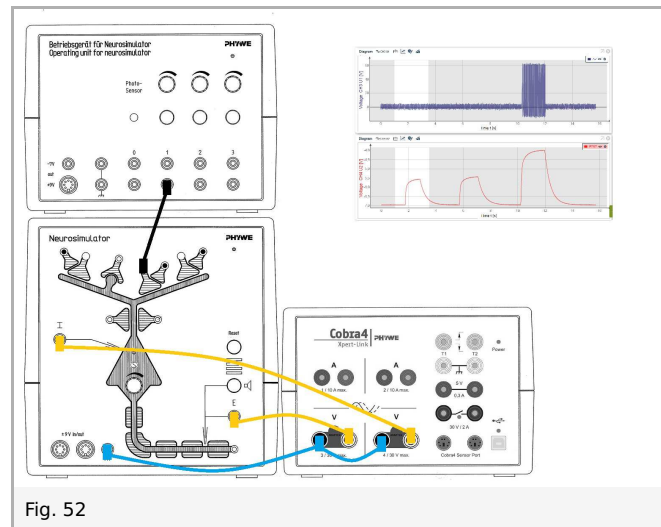


Fig. 52

Excitatory synapses are typically localized at the distal (away from the cell body) sections of dendrites. Afferent signals that reach the cell via an excitatory synapse depolarize the neuron, i.e. the membrane potential is changed to a more positive value. As the resting potential is negative relative to the cell environment, it decreases. This occurs mainly by a flow of sodium ions into the cell. The stimulating effect of depolarization is based on the increase in the membrane potential up to near the firing threshold or even above it. Transmitter substances of excitatory synapses are, for example, acetylcholine or glutamate.

Stimulations via excitatory synapses depolarize the cell membrane of the intracellular potential, i.e. the voltage gradient between inside and outside the nerve cell membrane becomes less negative.

The following experiments can be performed:

1. Effect of the excitatory synapse
2. Temporal summation
3. Spatial summation
4. Synaptic amplification by terminal branches

Set-up and procedure

Notes: Connect the Xpert-Link to the PC.

The knobs are infinitely adjustable. However, specific values (e.g. 50%, 33%, 66%, 100%) are given for the clear description of the experiments.

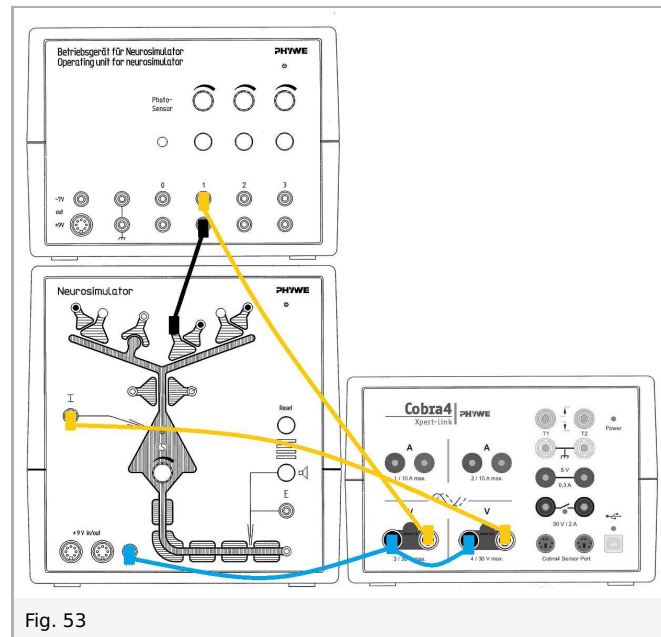
Voltage measurements can be taken on both Xpert-Link voltage channels (CH3 and CH4). Wiring diagrams are given for the experiments.

1. Effect of the excitatory synapse

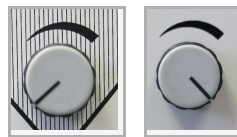
Connect the Xpert-Link to the PC. Two BNC-adapters (plug/socket 4 mm) are needed for voltage measurement.

1.1. Effect of stimulation of the excitatory synapse on the membrane potential (EPSP)

The experiment is set up as per fig. 53.



- Neurostimulator, knob threshold: 0%
- Operating unit, knob stimulation intensity 1: 100%



- Start measurement in the measurement window.
- Press the stimulation button 1 for approximately one second.
- Finish the measurement as soon as the voltage of EPSP (now CH4) has reached the initial value.
- Save and evaluate the results.

1.2. Effect of the membrane potential (EPSP) on the action potential (AP)

The experiment is set up as per fig. 54.

(This experiment and part 1 of experiment "1. Membrane potential and action potential" are identical. You can use the same diagram.)

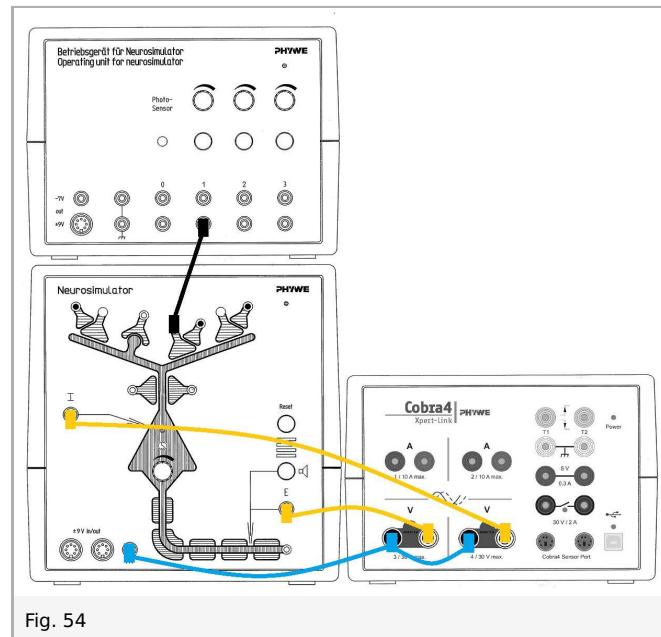


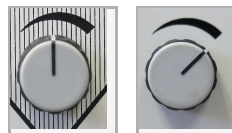
Fig. 54

- Start measurement in the measurement window.
- Press the stimulation button 1 for approximately one second.
- Finish the measurement as soon as the voltage of EPSP (now CH4) has reached the initial value.
- Save and evaluate the results.

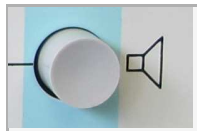
2. Temporal summation

The experiment is set up as per Fig. 54.

- Neurosimulator, knob threshold: 50%
- Operating unit, knob stimulation intensity 1: 66%



Temporal summation makes use of the integrated loudspeaker of the operating unit (acoustic monitor).

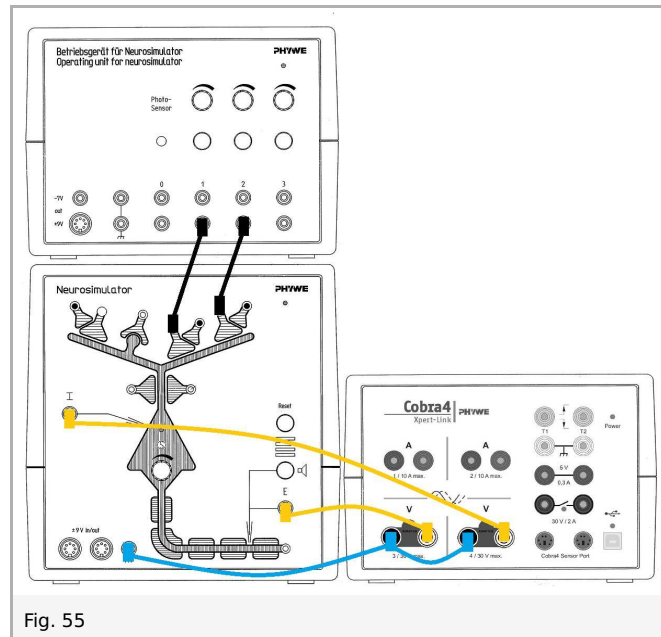


The signal helps find the level of stimulus at which no acoustic signal is emitted when pressing the stimulus button **very briefly** – calibrate carefully by turning the stimulus knob counterclockwise and by pressing the button.

- Start measurement in the measurement window.
- Press the stimulation button 20 times: each button press should be very brief, but between each press the voltage of EPSP should reach the initial value (approx. -7V).
- Press the stimulation button 20 times in a row.
- Finish the measurement as soon as the voltage of EPSP has reached the initial value.
- Save and evaluate the results.

3. Spatial summation

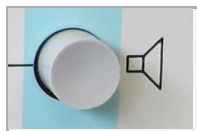
The experiment is set up as per fig. 55.



- Neurosimulator, knob threshold: 50%
- Operating unit, knob stimulation intensity 1: 33%
- Operating unit, knob stimulation intensity 2: 33%



Spatial summation makes use of the integrated loudspeaker of the operating unit (acoustic monitor). The signal helps find the level of stimulus at which no acoustic signal is emitted when pressing only one of the two stimulus buttons.

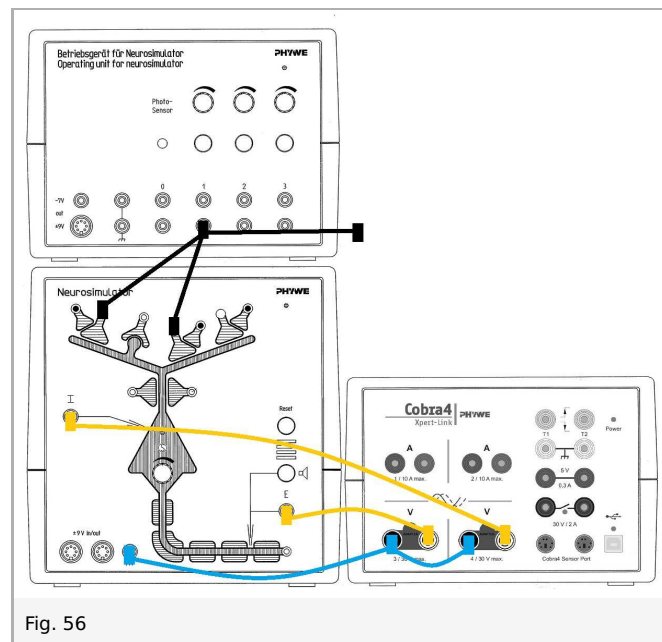


- Start measurement in the measurement window.
- Press the stimulation button 1 for approximately one second. Wait until the voltage of EPSP has reached the initial value.
- Press the stimulation button 2 for approximately one second. Wait until the voltage of EPSP has reached the initial value.
- Press the stimulation button 1 and 2 for approximately one second.
- Finish the measurement as soon as the voltage of EPSP has reached the initial value.
- Save and evaluate the results.

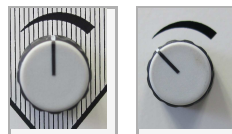
4. Synaptic amplification by terminal branches

The experiment is set up as per fig. 56.

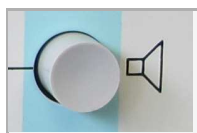
Connect two of the three excitatory synapses with stimulation output 1 with two cables. Leave the third cable unplugged.



- Neurosimulator, knob threshold: 50%
- Operating unit, knob stimulation intensity 1: 33%



Again here, determine action potentials acoustically. As in the two previous experiments, the position of the stimulus knob is determined at which no signal is emitted, i.e. no action potential is created.



Now pull one of the cables out of one green socket (Fig. 57) of one of the excitatory synapses.

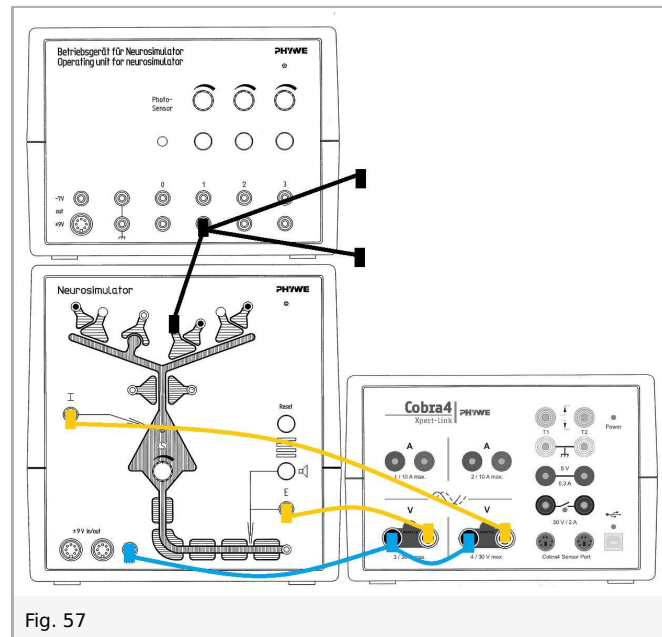


Fig. 57

- Start measurement in the measurement window.
- Press the stimulation button 1 for approximately one second. Wait until the voltage of EPSP has reached the initial value.
- Plug one loose cable back into a green socket (fig. 58).

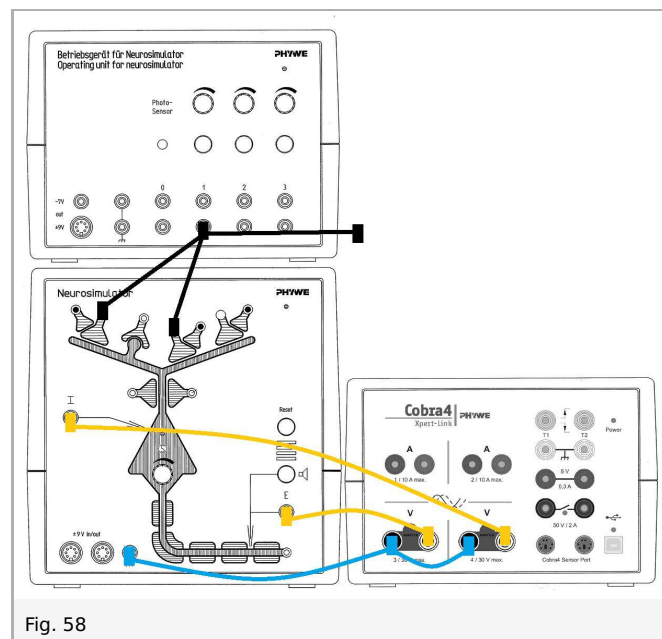
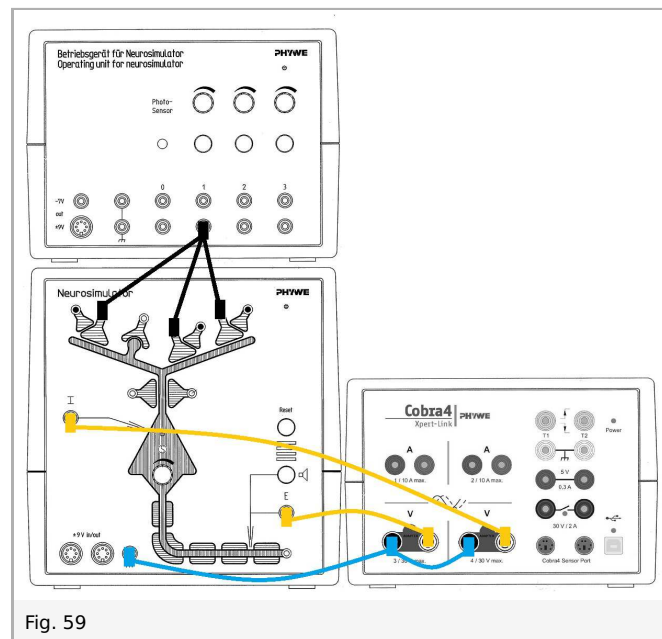


Fig. 58

- Press the stimulation button 1 for approximately one second. Wait until the voltage of EPSP has reached the initial value.
- Plug all three cables into the green sockets of the three excitatory synapses (fig. 59).



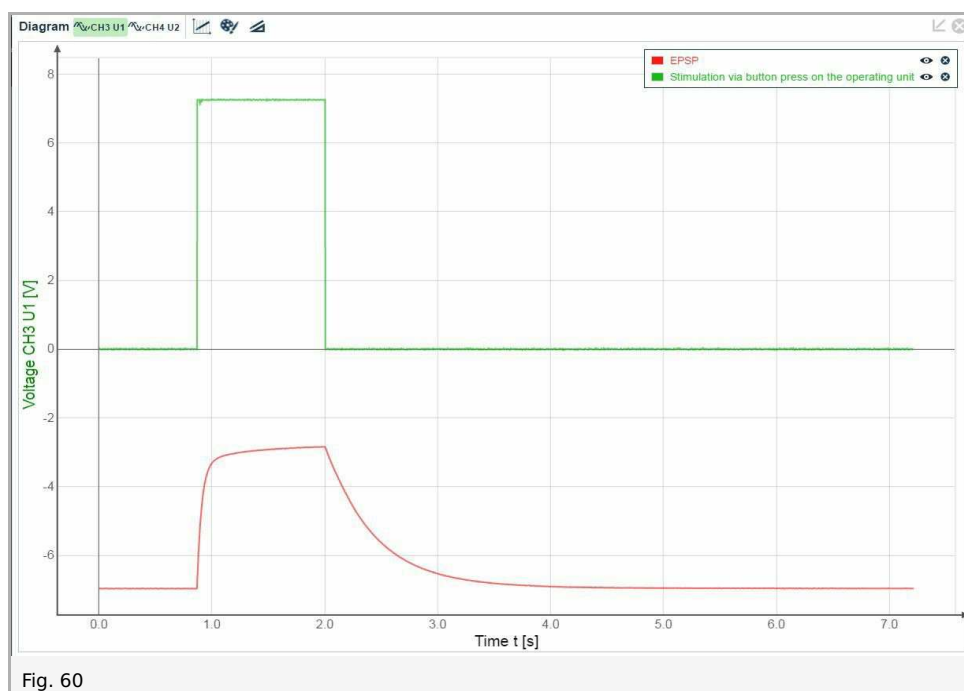
- Press the stimulation button 1 for approximately one second.
- Finish the measurement as soon as the voltage of EPSP has reached the initial value.

Results and evaluation

The threshold potential is the critical level at which a membrane potential must be depolarized to initiate an action potential. You will receive the following diagrams:

1. Effect of the excitatory synapse

1.1. Effect of stimulation of the excitatory synapse on the membrane potential (EPSP)



1.2. Effect of the membrane potential (EPSP) on the action potential (AP)

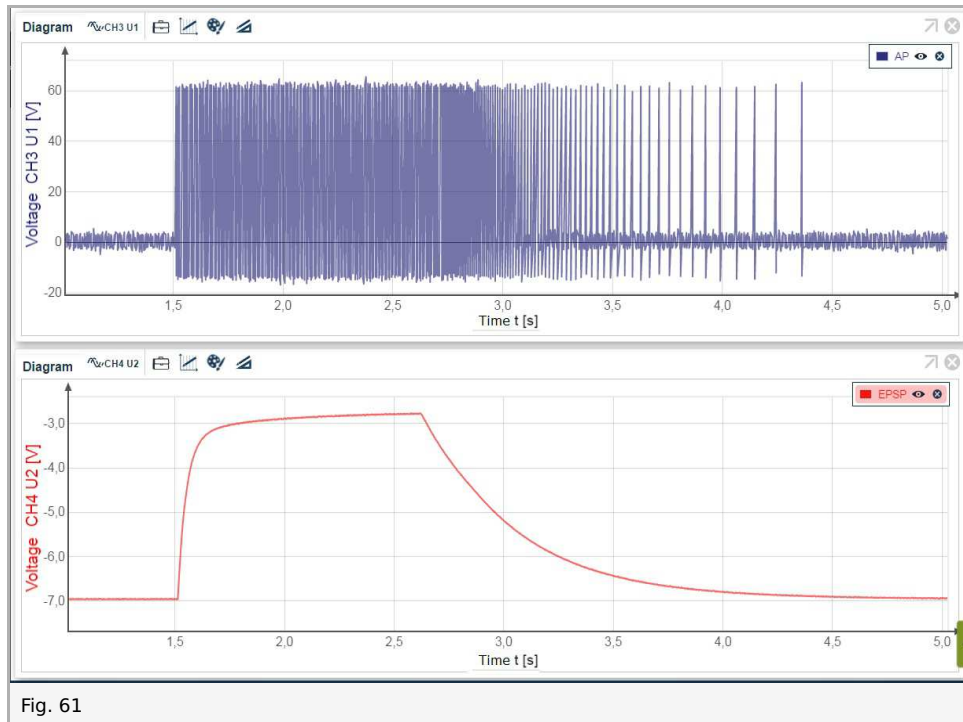


Fig. 61

2. Temporal summation

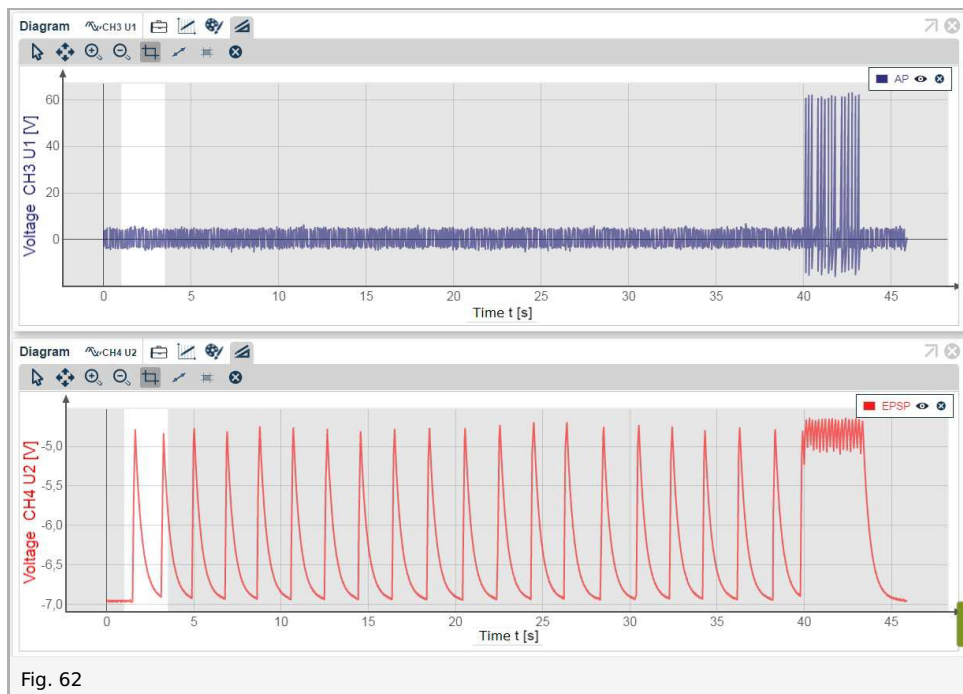


Fig. 62

3. Spatial summation

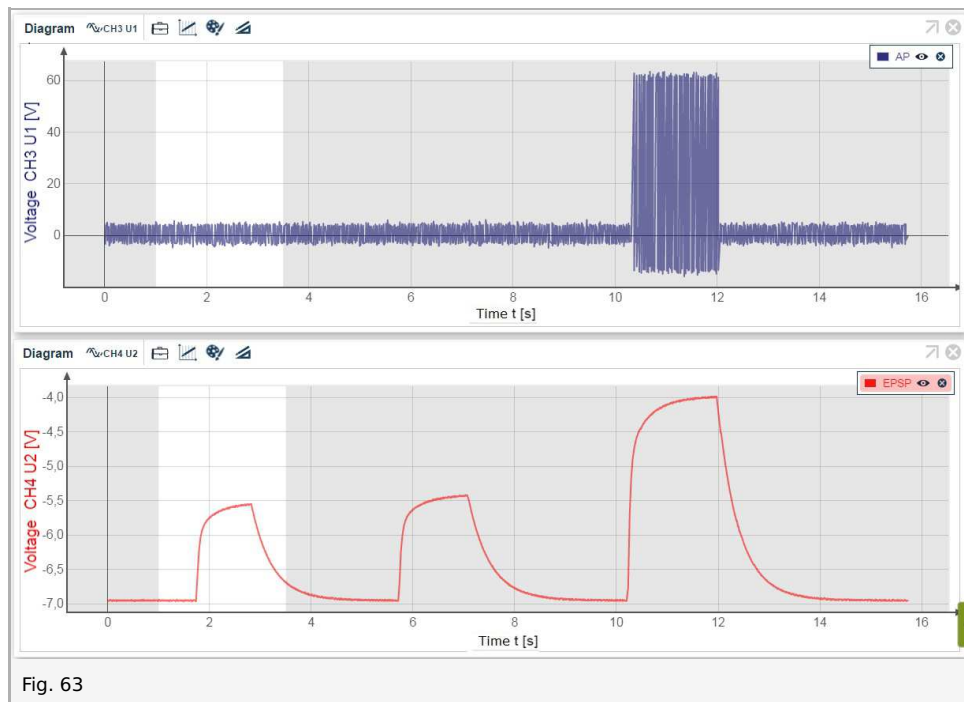


Fig. 63

4. Synaptic amplification by terminal branches

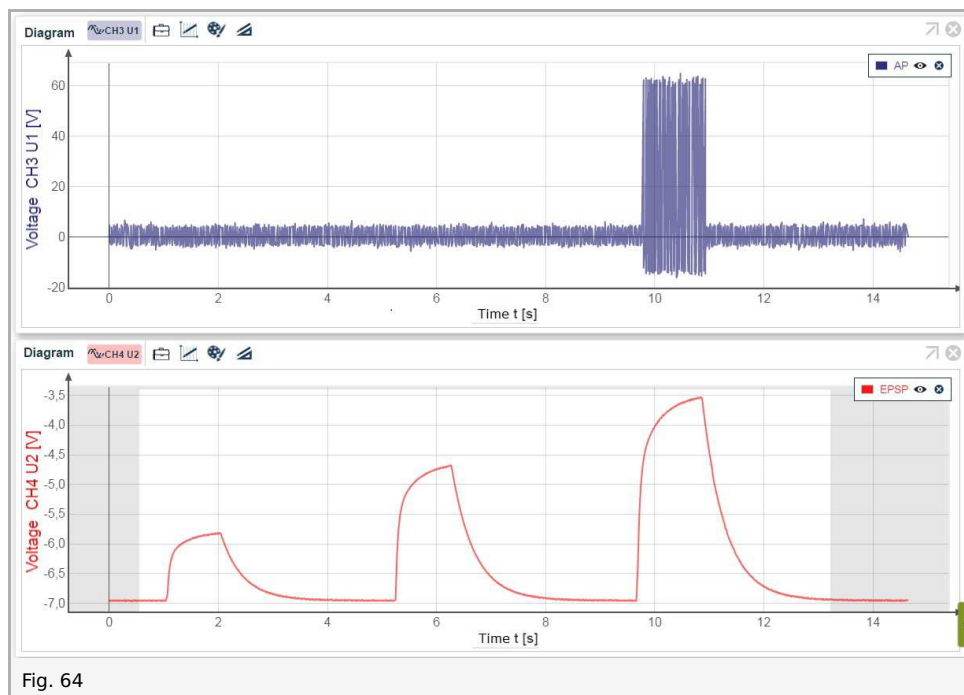
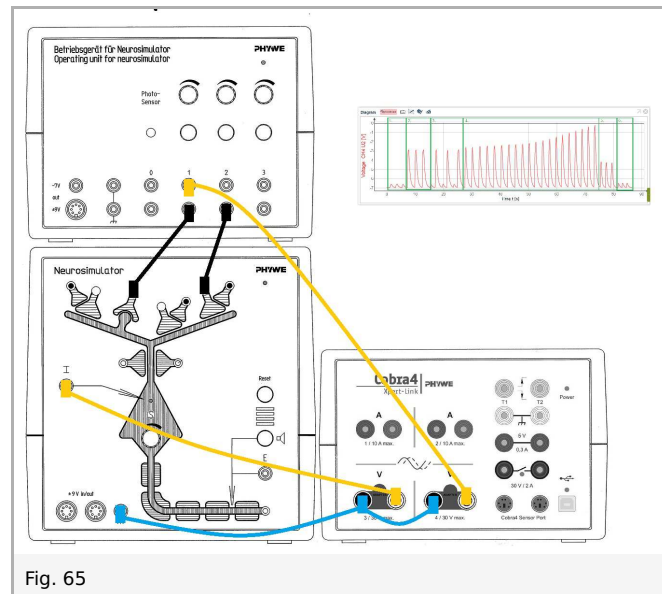


Fig. 64

3. Hebbian synapse

Introduction



In the anatomical sample the Hebbian synapse is located at the end of dendritic spines. It is an excitatory synapse with variable transmission behavior.

Synapses of Hebb's type (named after the Canadian behavioural and neurobiologist Donald Hebb) are excitatory synapses that change their transmission characteristics relative to chronological excitation conditions. Starting from a very low transmission strength, the relationship of the output signal to the input signal of the synapse, the transmission factor, increases when two conditions are simultaneously fulfilled: Firstly, an afferent signal must be present, secondly, intracellular depolarization must exist that is not the result of that signal. When these two conditions are fulfilled for a sufficient length of time, a long sustained amplification of the transmission factor occurs (long term potentiation = LTP), which is said to be the basis for memory processes. "Forgetting", i.e. attenuation of the transmission factor, occurs when these conditions are not fulfilled over a longer period of time.

The amplifying effect is probably attained by an increased postsynaptic inflow of calcium ions as a result of the opening of additional voltage-dependent calcium channels during synaptic potentiation. The experimental results suggest that glutamate, as transmitter of the afferent fiber, attaches to the N-methyl-D-aspartate (NMDA) receptor of the postsynaptic membrane. Should the dendrite not be sufficiently polarized because of too low an activity of other excitatory afferents, then magnesium cations block the receptor and LTP cannot occur. With sufficiently large polarization, magnesium can no longer bind to the NMDA receptor. Glutamate takes its place and results in an opening of calcium channels: Calcium ions can enter into the cell. The increase in the postsynaptic calcium concentration in the dendritic spine is the prerequisite for the stabilization of the LTP. It is assumed that the increased calcium concentration leads to activation of the protein kinase enzyme, which transfers a phosphate group to other proteins and so activates them. When these are membrane proteins, then the properties of the ion channels can be changed and the activity of the cell increased.

The following experiment can be performed:

- Effect of the Hebbian synapse - a basic mechanism for synaptic plasticity

All processes are measured in one measurement:

Process 1: Activation of the Hebbian synapse.

Process 2: Activation of the excitatory synapse.

Process 3: Consecutive activation of the Hebbian and excitatory synapses (no learning process).

Process 4: Simultaneous activation of the Hebbian and excitatory synapses (learning process).

Process 5: Activation of the Hebbian synapse after learning process.

Process 6: Activation of the Hebbian synapse after synaptic forgetting.

Set-up and procedure

Connect the Xpert-Link to the PC.

The experiment is set up as per fig. 66. Two BNC-adapters (plug/socket 4 mm) are needed for voltage measurement.

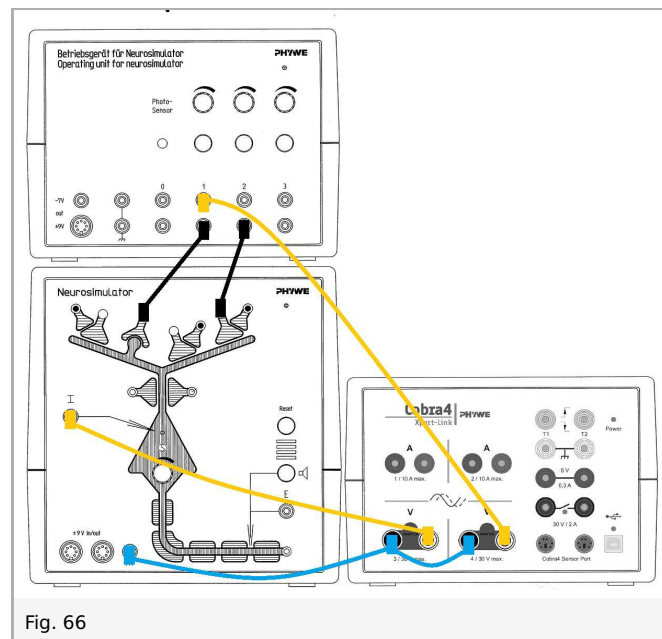


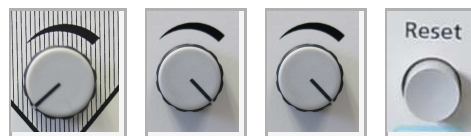
Fig. 66

Neurostimulator, knob threshold: 0%

Operating unit, knob stimulation intensity 1: 100%

Operating unit, knob stimulation intensity 2: 100%

Press the reset button on the Neurostimulator. The reset button initiates **synaptic forgetting**.



- Start measurement in the measurement window.

Notes: Between each button press the voltage of EPSP (now CH3) should reach the initial value (approx. -7V).

1. Press the stimulation button 1 three times for approximately one second (activation of the Hebbian synapse).
 2. Press the stimulation button 2 three times for approximately one second (activation of the excitatory synapse).
 3. Press the stimulation buttons 1 and 2 alternately, each button three times for approximately one second (consecutive activation of the Hebbian and excitatory synapses).
 4. Press the stimulation buttons 1 and 2 simultaneous 20 times for approximately one second (simultaneous activation of the Hebbian and excitatory synapses).
 5. Press the stimulation button 1 three times for approximately one second (activation of the Hebbian synapse).
 6. Press the reset button on the Neurostimulator (synaptic forgetting). Press the stimulation button 1 three times for approximately one second again (activation of the Hebbian synapse).
- Finish the measurement as soon as the voltage has reached the initial value.
 - Save and evaluate the results.

Results and evaluation

You will receive the following diagram:

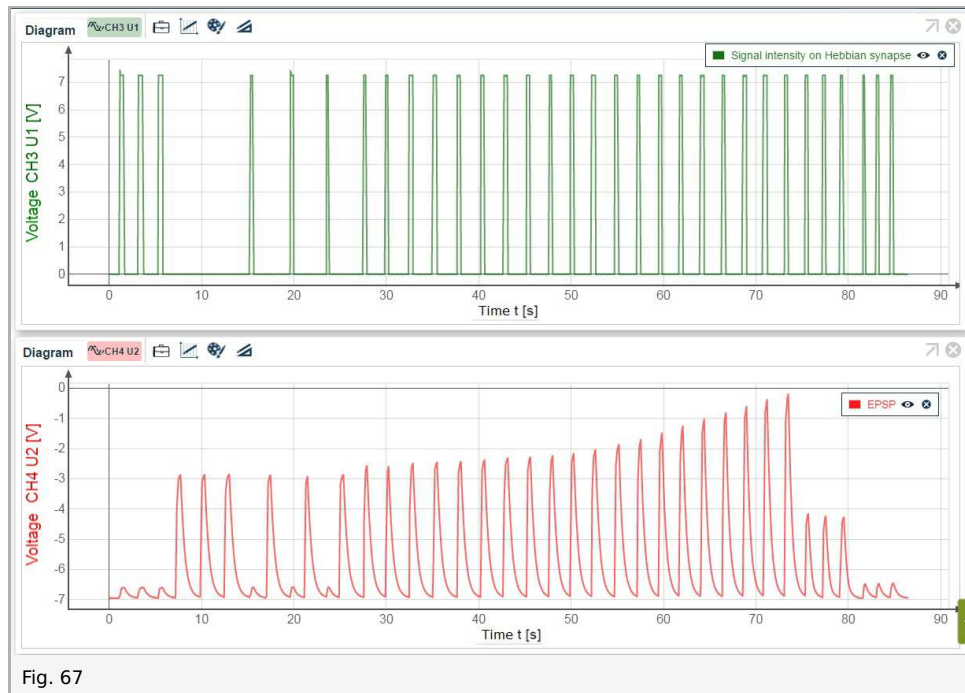


Fig. 67

The diagram shows the following experiment parts:

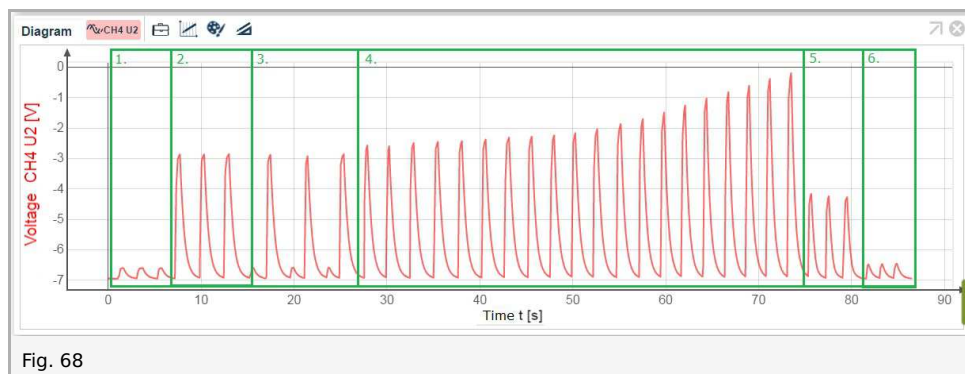
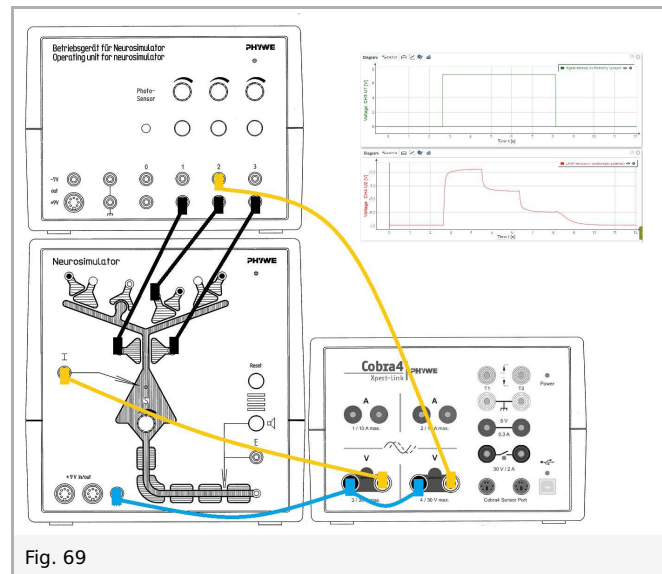


Fig. 68

1. Activation of the Hebbian synapse.
2. Activation of the excitatory synapse.
3. Consecutive activation of the Hebbian and excitatory synapses (no learning process).
4. Simultaneous activation of the Hebbian and excitatory synapses (learning process).
5. Activation of the Hebbian synapse after learning process.
6. Activation of the Hebbian synapse after synaptic forgetting.

4. Inhibitory synapse

Introduction



Afferent signals that reach the cell across inhibitory synapses hyperpolarize the neuron, i.e. the intracellular potential changes in the negative direction. As the resting potential is already negative, the negative polarity is increased: hyperpolarization. This is generally the result of an outflow of potassium cations from the cell and an inflow of chloride anions. The inhibitory effect of hyperpolarization is based on the removal of the membrane potential further from the firing threshold. A frequent inhibitory transmitter substance is gamma amino-butyric acid (GABA).

In the anatomical sample the location of the inhibitory synapse is in the shaft of the dendrite of a nerve cell.

The following experiments can be performed:

1. Hyperpolarization
2. Spatial inhibitory-excitatory summation

Set-up and procedure

Connect the Xpert-Link to the PC.

Experiment 1: Hyperpolarization

The experiment is set up as per fig. 70. Two BNC-adapters (plug/socket 4 mm) are needed for voltage measurement. Now we measure the stimulus intensity of the inhibitory synapse (the inhibitory postsynaptic potential = IPSP).

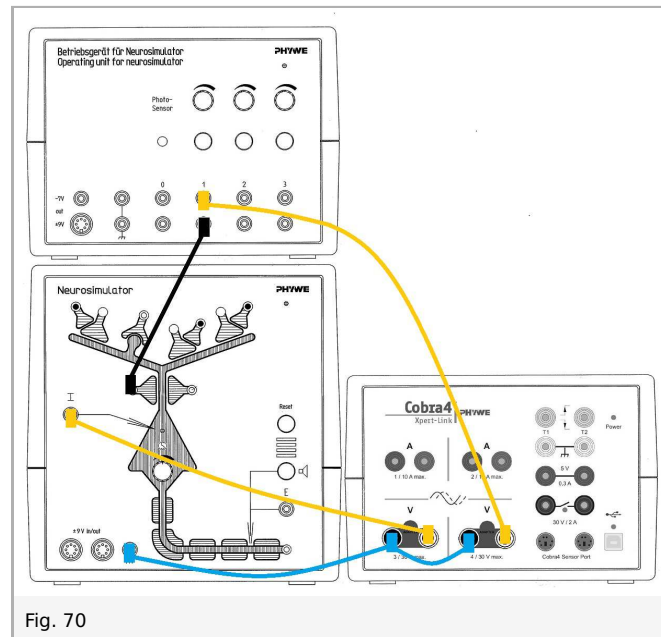
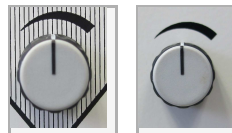


Fig. 70

Neurostimulator, knob threshold: 50%
Operating unit, knob stimulation intensity 1: 50%



- Start measurement in the measurement window.
- Press the stimulation button 1 for 1 to 2 seconds.
- Finish the measurement as soon as the voltage of IPSP (now CH3) has reached the initial value.
- Save and evaluate the results.

Experiment 2: Spatial inhibitory-excitatory summation

For measurement of the excitatory postsynaptic potential (EPSP), the experiment is set up as per fig. 71. Now we measure stimulus intensity of the excitatory synapse and EPSP as a result of spatial inhibitory-excitatory summation.

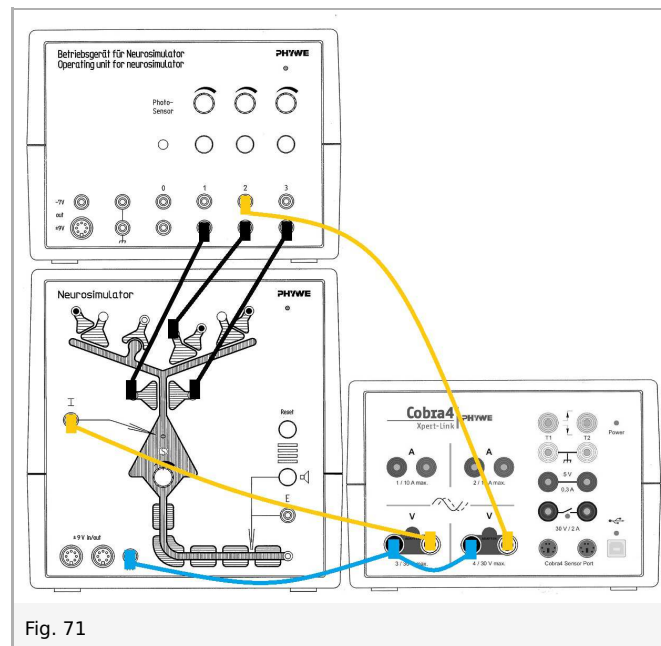
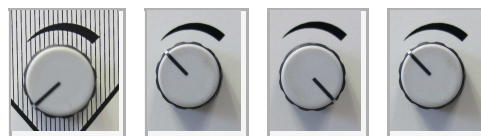


Fig. 71

Neurostimulator, knob threshold: 0%
 Operating unit, knob stimulation intensity 1: 33%
 Operating unit, knob stimulation intensity 2: 100%
 Operating unit, knob stimulation intensity 3: 33%



- Start measurement in the measurement window.
- Press the stimulation button 2. Keep this button pressed and wait for approximately 2 seconds.
- In addition press the stimulation button 1. Keep both buttons pressed and wait for approximately 2 seconds.
- In addition press the stimulation button 3. Keep all buttons pressed and wait for approximately 2 seconds.
- Finish the measurement as soon as the voltage of EPSP (now CH3) has reached the initial value.
- Save and evaluate the results.

Results and evaluation

Experiment 1: Hyperpolarization

Inhibitory synapses exert an inhibitory effect. The effect is similar to excitatory synapses, however, its effect is inverse: the negative polarisation of the intracellular potential increases further (> 70 mV) = hyperpolarization = reduced excitation.

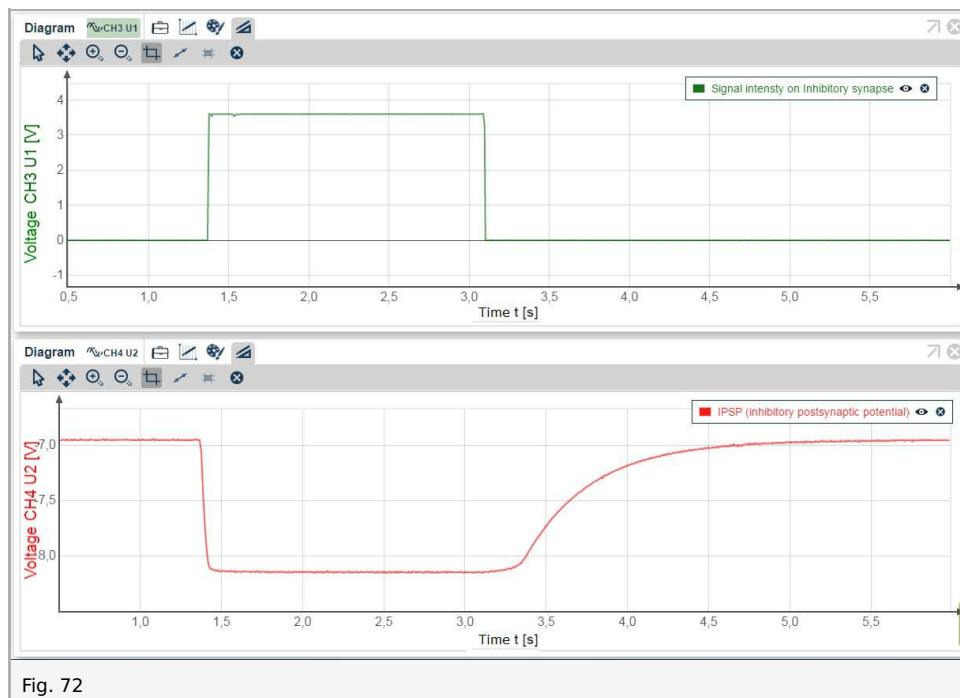


Fig. 72

Experiment 2: Spatial inhibitory-excitatory summation

You will receive the following diagram:

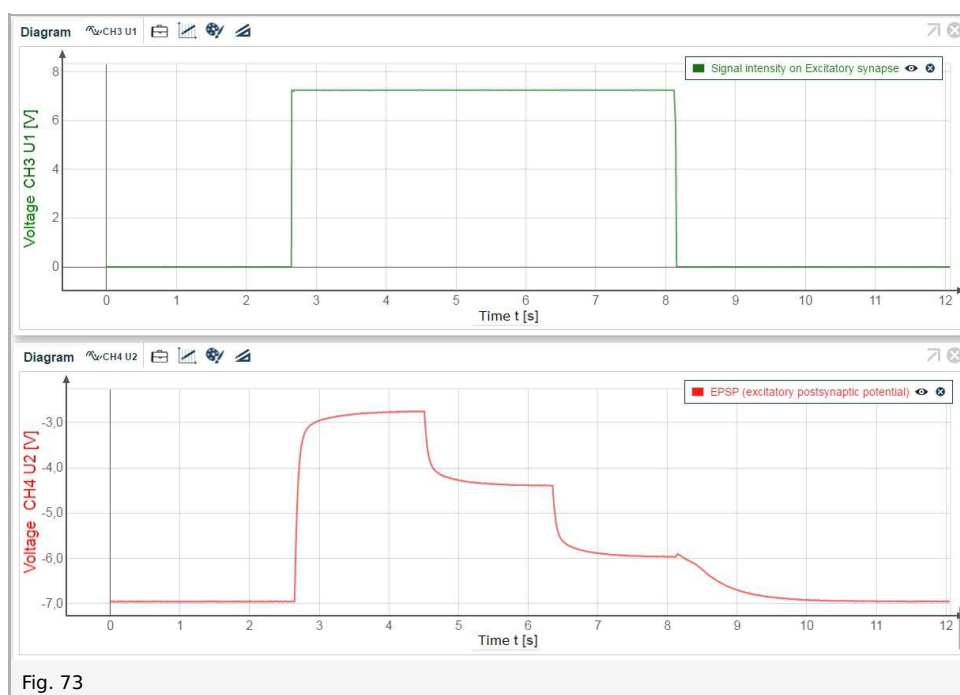


Fig. 73

5. Veto synapse

Introduction

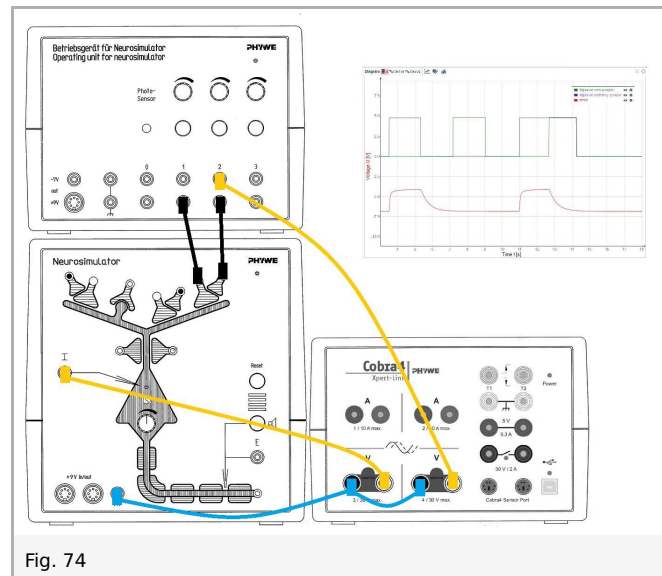


Fig. 74

Synapses with veto properties are mostly presynaptic to excitatory synapses. They prevent the excitement of postsynaptic cells by selectively stopping the signal transmission of an excitatory synapse. This can be effected by blockage of the transmitter distribution in the corresponding excitatory synapses. As a result, the postsynaptic cell shows, according to the intensity of the signal at the veto synapse, only a low or no depolarization with simultaneous activation of the excitatory and veto inhibiting afferents. A signal at the veto synapses alone has no influence on the potential of the postsynaptic cell and contrary to the inhibitory synapses, no hyperpolarization takes place. Signals of other synapses are also not influenced. This type of inhibition is called "still inhibition", as it cannot be derived.

The following experiment can be performed:

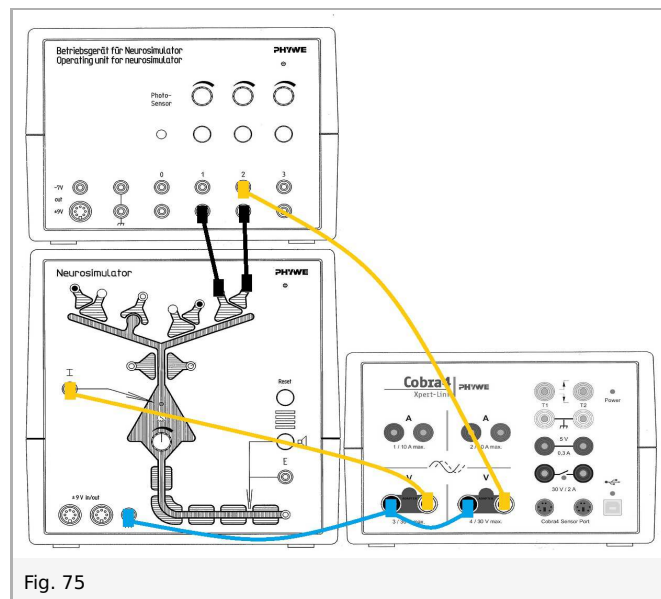
- Effect of the Veto synapse

Additional information: The value „stimulation intensity 1“ can be measured additionally by using a Cobra4 Sensor Port. To use the sensor port, a Cobra4 Sensor-Unit Electricity (12644-00) and a Cobra4 Xpert-Connect (12625-01) are needed additionally. (For settings please see: Task and equipment, Instructions for measureLAB, Additional channel with additional material.)

Set-up and procedure

Notes: Connect the Xpert-Link to the PC.

The experiment is set up as per fig. 75. Two BNC-adapters (plug/socket 4 mm) are needed for voltage measurement.



- Neurosimulator, knob threshold: 50%
- Operating unit, knob stimulation intensity 1: 66%
- Operating unit, knob stimulation intensity 2: 66%



- Start measurement in the measurement window.
- Press the stimulation button 1 for 1 to 2 seconds. Wait until the voltage of EPSP (now CH3) has reached the initial value.
- Press the stimulation button 2 for 1 to 2 second. Wait until the of EPSP voltage has reached the initial value.
- Press the stimulation button 1. Keep this button pressed and wait for approximately 2 seconds. In addition press the stimulation button 2. Keep both buttons pressed for 1 to 2 seconds.
- Finish the measurement as soon as the voltage of EPSP has reached the initial value.
- Save and evaluate the results.

Results and evaluation

The veto synapse does not bring about a change of the intracellular potential (so-called silent inhibition). Its effect is only on the excitatory synapse to which it is attached (so-called presynaptic inhibition or shunting inhibition). There is no spatial summation (see fig. 76).

You will receive the following diagram:

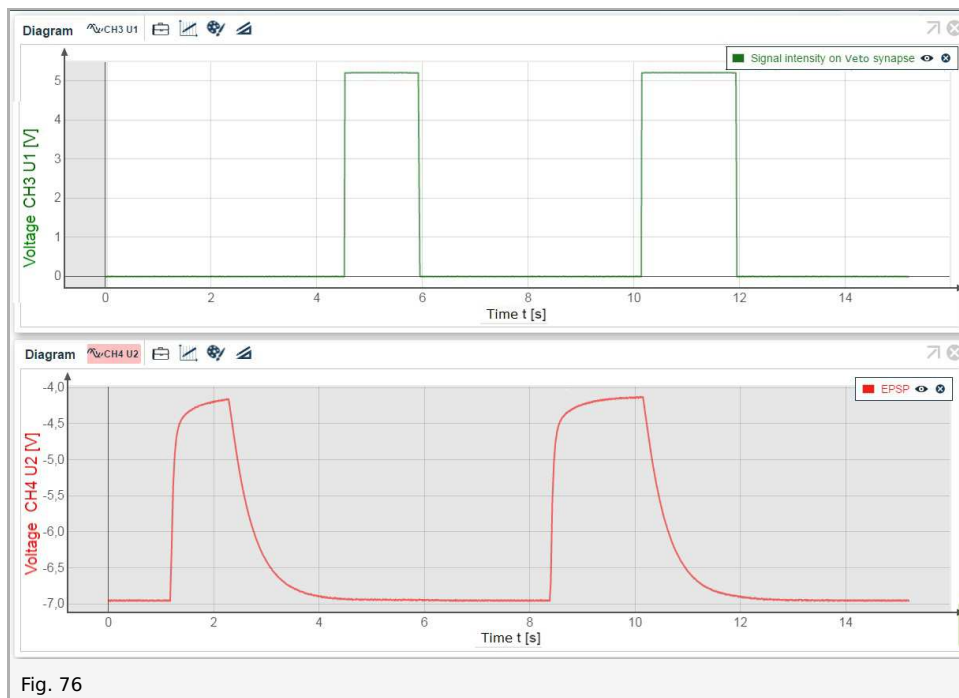


Fig. 76

Additional information: As shown in fig. 77, the stimulation by an excitatory synapse can be measured on the stimulation output on the operating unit. For this measurement, you need a third voltage channel using one Cobra4 Sensor Port together with an Cobra4 Xpert-Connect adapter and the Cobra4 Sensor-Unit Electricity. Please note the different settings for Xpert-Link.

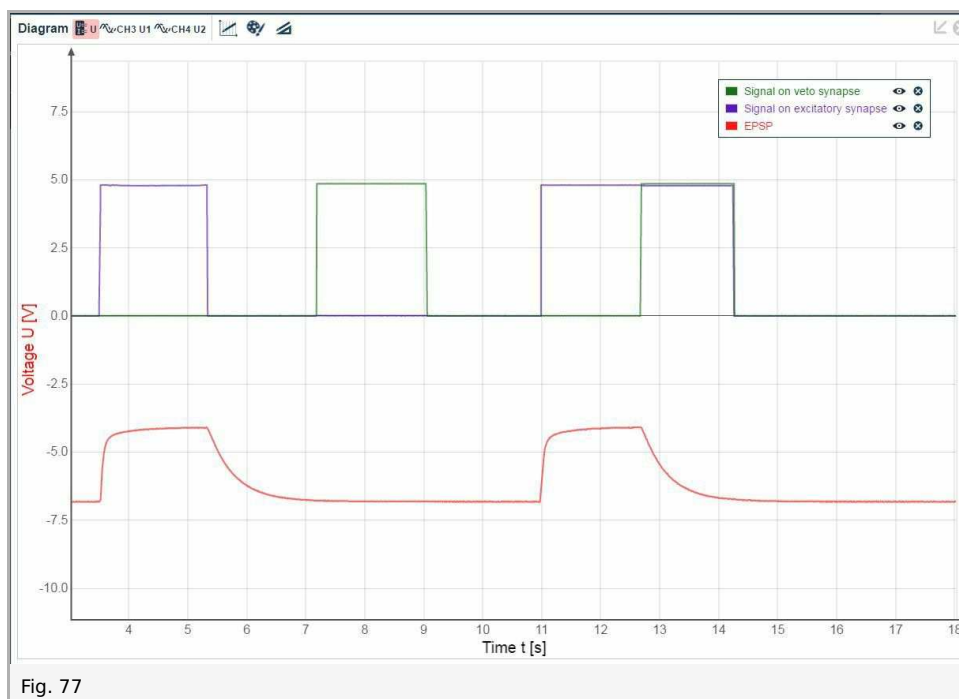


Fig. 77