

# OPERATING INSTRUCTIONS AND SYSTEM DESCRIPTION FOR THE

## **ELC-03XS**

### **UNIVERSAL AMPLIFIER** with switchable x10 / x100 headstage for **EXTRA & INTRACELLULAR RECORDING,** **SINGLE CELL STIMULATION and** **ELECTROPORATION**



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## 1. Safety Regulations

**VERY IMPORTANT:** Instruments and components supplied by npi electronic are NOT intended for clinical use or medical purposes (e.g. for diagnosis or treatment of humans), or for any other life-supporting system. npi electronic disclaims any warranties for such purpose. Equipment supplied by npi electronic must be operated only by selected, trained and adequately instructed personnel. For details please consult the **GENERAL TERMS OF DELIVERY AND CONDITIONS OF BUSINESS** of npi electronic, D-71732 Tamm, Germany.

**GENERAL:** This system is designed for use in scientific laboratories and must be operated only by trained staff. General safety regulations for operating electrical devices should be followed.

**AC MAINS CONNECTION:** While working with npi systems, always adhere to the appropriate safety measures for handling electronic devices. Before using any device please read manuals and instructions carefully.

The device is to be operated only at 115/230 Volt 60/50 Hz AC. Please check for appropriate line voltage before connecting any system to mains.

Always use a three-wire line cord and a mains power-plug with a protection contact connected to ground (protective earth).

Before opening the cabinet, unplug the instrument.

Unplug the instrument when replacing the fuse or changing line voltage. Replace fuse only with an appropriate specified type.

**STATIC ELECTRICITY:** Electronic equipment is sensitive to static discharges. Some devices such as sensor inputs are equipped with very sensitive FET amplifiers, which can be damaged by electrostatic charge and must therefore be handled with care. Electrostatic discharge can be avoided by touching a grounded metal surface when changing or adjusting sensors. Always turn power off when adding or removing modules, connecting or disconnecting sensors, headstages or other components from the instrument or 19" cabinet.

**TEMPERATURE DRIFT / WARM-UP TIME:** All analog electronic systems are sensitive to temperature changes. Therefore, all electronic instruments containing analog circuits should be used only in a warmed-up condition (i.e. after internal temperature has reached steady-state values). In most cases a warm-up period of 20-30 minutes is sufficient.

**HANDLING:** Please protect the device from moisture, heat, radiation and corrosive chemicals.

## 2. Introduction

“Loose patch” recordings (or “loose seal” recordings [Roberts & Almers, 1992]) are used to record from single excitable cells without damage, i.e. without a direct access to the cell interior. The first recordings were made around 1960 from muscle cells by Alfred Strickholm long time before “tight seal” recording was invented by Erwin Neher and Bert Sakmann twenty years later: “*A method has been developed permitting measurement of membrane impedance and current, as a function of transmembrane potential, at small, electrically isolated regions of the muscle cell surface without microelectrode impalement.*”[Strickholm 1961].

The loose seal has a resistance of a few ten to a few hundred M $\Omega$ , and it creates an electrically isolated access to a single neuron. This isolated area can be used for precise recording, stimulation or drug and dye application on the single cell level without damaging the cell [Babour & Isope, 2000]. In contrast to tight seal recordings the same electrode can be reused for recording from several cells, which is a great advantage.

Since its beginnings several attempts have been made to make such precise extracellular methods accessible to various preparations. A nice overview can be found in the chapter by Roberts & Almers [Roberts & Almers, 1992]. Over the years the method was extended to cultured neurons and brain slice preparations, and also for *in vivo* recordings [Bureau et al, 2004]. The method is particularly well suited for long term recording with little damage to the recorded neuron [Nunemaker et al, 2003]. It can be used both for somatic and axonal recording [Khaliq & Raman 2005]. Even subcellular structures such as synaptic boutons are accessible to loose patch recordings [Auger & Marty, 2000].

Another valuable application of this method is single cell stimulation. The high resistance loose patch makes possible the application of 1-2 V stimuli to one cell only [Babour & Isope, 2000].

In the nineties of the last century the method of juxtacellular dye application (juxtosomal filling) became popular [Pinault, 1996]. This staining method is based on repetitive current pulse trains applied in the close vicinity of cell somata or dendrites and is meanwhile well established in the field of slice and *in vivo* preparations [Klausberger, 2004]. Juxtacellular filling together with extracellular measurements are today often summarized under the term “juxtacellular recording”.

In parallel attempts were made towards transfection of single cells by electroporation using patch pipettes. DNA or other large molecules were successfully inserted through a patch pipette into living cells by using an optimized protocol (application of 10 V / 1 ms pulse trains) [Rathenberg et al, 2003].

Far in excess of classical *in vivo* recording methods [Lalley et al, 1999] several new approaches are used for monitoring neuronal activity under natural conditions, using new techniques, e.g. the combination of two photon excitation and patch clamp *in vivo* [Helmchen et al, 2001; Stosiek et al, 2003; Brecht et al, 2002]. Assays have been developed that allow to monitor and manipulate single cells under *in vivo* conditions [Brecht et al, 2004]. Besides sophisticated optics these techniques always require precise recording and stimulation amplifiers, mostly based on the use of patch electrodes.

Today three methods are used to record electrical *in vivo* or *in vitro*:

- ❑ Recordings using patch (suction) electrodes from single neurons
  - Whole cell patch clamp technique (tight seal recording, intracellular)
  - Loose patch technique (loose seal recording, extracellular)
- ❑ Intracellular recordings with sharp microelectrodes
- ❑ Extracellular recordings with glass or metal electrodes

The amplifiers used for such recordings are specialized on the recording of the potentials or currents generated by the neurons under investigation. If these recording methods are combined with dye injection, electroporation, stimulation protocols etc. through the recording electrode, serious constraints occur and several additional devices have to be added to the experimental set-up.

The ELC series of amplifiers fills this gap. It allows intracellular, extracellular, voltage – clamp or current clamp recordings both with sharp or patch electrodes as well as additional protocols like electroporation or juxtosomal recordings. Even iontophoresis and voltammetry/amperometry can be performed.

The ELC amplifier is the “Swiss Army Knife” of modern electrophysiology. It is easy to use, versatile, and makes possible a lot of sophisticated experiments with only one instrument.

### 3. ELC-03XS amplifier

#### 3.1. ELC-03XS Components

The following items are shipped with the system:

- ✓ ELC-03XS amplifier
- ✓ GND (2.4 mm banana jack) connector and (optional) REF. connector for headstage
- ✓ Headstage
- ✓ User manual

#### 3.1. Optional Accessories

- Differential headstage
- Miniature headstage
- Cell model
- Pipette holder

#### 3.2. System Description

The ELC-03XS was designed for intra- and extracellular recording, precise (single cell) electrical stimulation as well as juxtosomal filling with patch electrodes. The system consists of an amplifier in a rackmount cabinet and a small headstage with dovetail, mounting plate or a holding bar. It can be used in slices or in *in vivo* preparations using the optional headstage with a differential input. It has separate capacity compensation controls for VC and CC mode, all controls (Bridge balance, CC cap comp, offset, holding controls) are calibrated 10-turn potentiometers.

The ELC-03XS is capable to record extracellular DC or AC coupled, to stimulate with current or voltage and to perform non-invasive juxtosomal filling of cells with dyes or DNA. The amplifier can also be used to record and stimulate intracellularly in current clamp (CC) with sharp or patch electrodes, or voltage (patch) clamp (VC) with patch electrodes. It is also suitable for amperometric or voltammetric investigations with carbon fiber electrodes.

#### *Operation modes of the amplifier*

The operation modes of the amplifier are selected by a rotary switch with six positions: The selected mode is indicated by LEDs above the MODE OF OPERATION switch.

- EXT: All MODEs OF OPERATION can be selected by TTL signals connected to the rear panel (see chapter 4).
- CC: CURRENT CLAMP MODE: used to inject predefined current signals
- CCx10: CURRENT CLAMP MODE : for stimulation and electroporation
- OFF: CC Mode with all input signals turned off
- VC: VOLTAGE CLAMP mode: potential commands are applied to the electrode
- VCx10: High range VC mode, for stimulation and electroporation

In addition, using a toggle switch a bridge balance circuit can be activated, to compensate for the electrode artifact (BRIDGE mode, only in CC mode). An ELECTRODE RESISTANCE

test mode can be activated with a push button. The electrode resistance is measured directly in  $M\Omega$  and displayed on the POTENTIAL display.

#### *Input configuration:*

The amplifier has two inputs, both for VC and CC mode. The signal applied to the analog input BNCs is converted either into a voltage command signal (x1 or x10 scaling) for the VC or VCx10 voltage clamp modes, or to a current in the CC, CCx10 and BRIDGE mode. Besides this, a signal generated from the 10-turn HOLD potentiometer can be transferred into a pulse using the GATE TTL input BNC. This control can be also used as HOLDING potentiometer if the switch in the GATE BNC is turned off.

#### *Computer control of the mode of operation*

In the EXT position of the MODE SELECT switch all MODEs OF OPERATION can be selected by TTL signals connected to the rear panel (see chapter 4).

#### *Output configuration*

The ELC-03XS amplifier has two output BNCs for POTENTIAL and one output BNC for the CURRENT signal. The POTENTIAL OUTPUT x1 is a pure DC output that monitors the electrode potential directly from the headstage. The signal at the POTENTIAL OUTPUT can be high and low-pass filtered and amplified.

The current output signal is amplified and can be filtered by a low-pass filter.

#### *Digital displays*

All ELC amplifiers are equipped with two digital displays, one for CURRENT (nA) and one for POTENTIAL (mV) or ELECTRODE RESISTANCE ( $M\Omega$ ). The mode of operation is indicated by LEDs located close to the digital displays.

#### *Oscillation shut-off unit*

All ELC amplifiers are equipped with an oscillation shut-off unit to protect the preparation. If it is active all outputs of the amplifier were disconnected if the system begins to oscillate.

#### *Penetration unit*

All ELC amplifiers are also equipped with a penetration unit (BUZZ) to facilitate the penetration of the cell membrane or to clean clogged electrodes.

3.3. Front Panel View of the ELC-03XS Amplifier

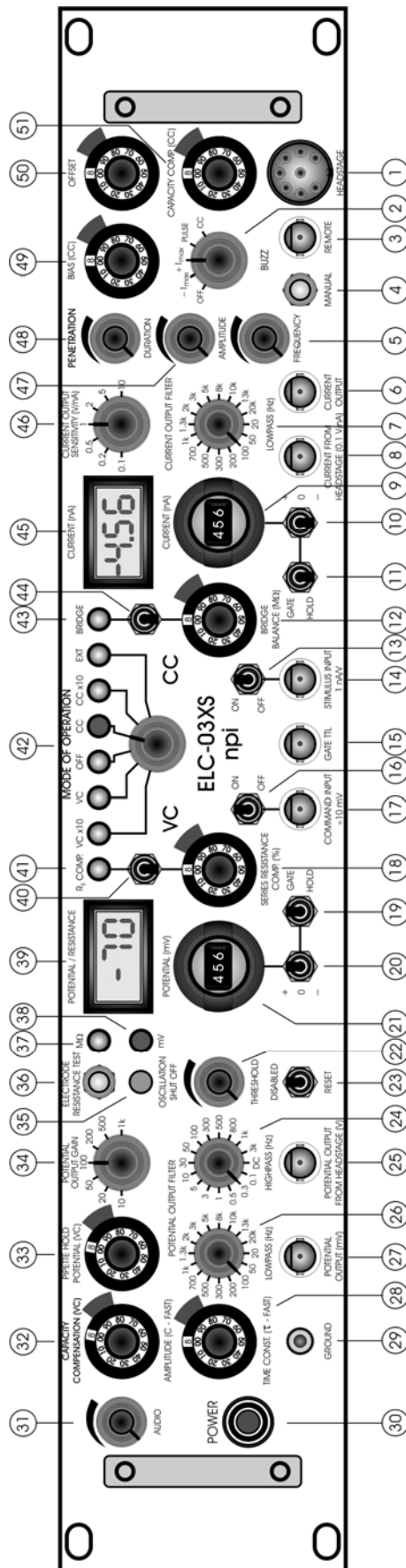


Figure 1: ELC-03XS front panel view



### 3.4. Description of the Front Panel

In the following description of the front panel elements each element has a number (in bold) that is related to that in Figure 1. The number is followed by the name (in uppercase letters) written on the front panel and the type of the element (in lowercase letters). Then, a short description of the element is given. Some elements are grouped in functional units (e.g. PENETRATION / BUZZ unit) and are described as units regardless of the order of numbers.

#### (1) HEADSTAGE connector



Connector for the headstage with optional differential input. REF of the headstage must be connected to ground (single-ended measurement) or to a reference electrode in the bath (differential measurement), see also Figure 10.

#### PENETRATION / BUZZ unit



The PENETRATION / BUZZ unit consist of (2) mode switch, (3) REMOTE connector, (4) MANUAL push button, (5) FREQUENCY potentiometer, (47) AMPLITUDE potentiometer and (48) DURATION potentiometer.

#### (2) mode switch



5-position switch to set the mode of PENETRATION

- CC:** The BUZZ circuit is activated (duration set by **48**). To facilitate the penetration of the cell membrane the BUZZ circuit is provided which is based on oscillations caused by overcompensating the capacitance compensation system. The overcompensation of capacitance compensation yields to very powerful high-frequency (approx. 2 kHz) oscillations (see Figure 2).
- PULSE:** A pulse train is applied to the electrode. The pulses are positive and rectangular. The AMPLITUDE is set by **47**, the DURATION of the train is set by **48** and the FREQUENCY of pulses within the train is set by **5**.
- +Imax:** The maximum positive current is applied to the electrode. The DURATION is set by **48**.
- Imax:** The maximum negative current is applied to the electrode. The DURATION is set by **48**.
- +Imax or -Imax** can also be used to clean the tip of the electrode by passing large amounts of positive or negative current. The maximum current is dependent on the setting of the current range, see also (42).
- OFF:** The PENETRATION / BUZZ unit is disabled. When in OFF position the preparation is protected from unintentional use of the unit.

(3) REMOTE connector



BNC connector to attach a remote switch in active-low configuration for activating the PENETRATION / BUZZ unit.

(4) MANUAL push button



Push button for activating the PENETRATION / BUZZ unit manually. The DURATION is set by 48 and the mode by 2.

**Note:** The duration is dependent on the setting of the DURATION potentiometer, but independent from how long the button is pushed or a REMOTE switch is pressed.

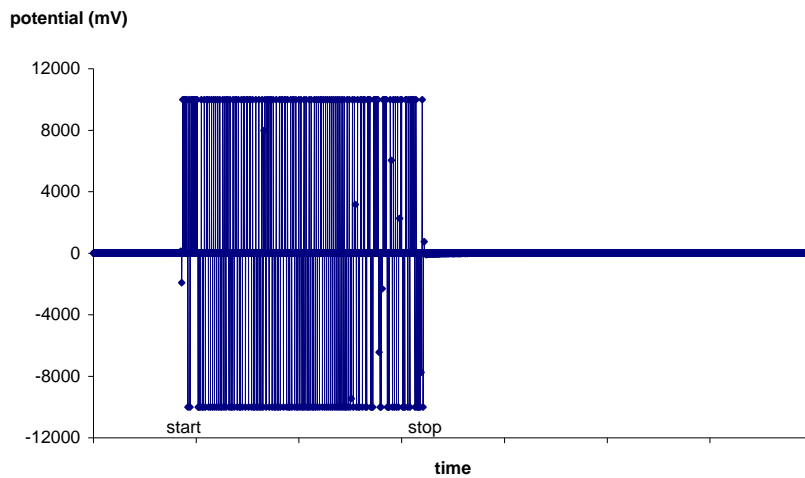


Figure 2: BUZZ function of the ELC-03XS (CC Mode)

(5) FREQUENCY potentiometer



Potentiometer to set the FREQUENCY of pulses within a train in PULSE mode (see also 2). Range: 100 Hz to 2 kHz.

(47) AMPLITUDE potentiometer



Potentiometer for setting the AMPLITUDE of pulses in PULSE mode (see also 2).

(48) DURATION potentiometer



Control to set the duration of the BUZZ (potentiometer, clockwise, range: ~1 ms to ~100 ms). It is effective in all modes; REMOTE controlled and when pushing the MANUAL button.

**(6) CURRENT OUTPUT connector**



BNC connector providing the current output signal; scaling is set by CURRENT OUTPUT SENSITIVITY (V/nA) switch **46**, filter is set by CURRENT OUTPUT FILTER LOWPASS (Hz) switch **7**.

**(7) CURRENT OUTPUT FILTER LOWPASS (Hz) switch**



16-position rotary switch to select the corner frequency of the LOWPASS FILTER (range: 20 Hz to 20 kHz) for the CURRENT OUTPUT connector **(6)**.

**(8) CURRENT FROM HEADSTAGE (0.1V/nA) connector**



BNC connector providing a voltage proportional to the current at the electrode with a fixed scaling of 100 mV / nA. This current signal is not filtered.

**CURRENT set unit**



The CURRENT set unit consist of **(9)** CURRENT nA potentiometer, **(10)** +/- switch and **(11)** GATE / HOLD switch.

**(9) CURRENT nA potentiometer**



Ten-turn potentiometer for generating a holding current (switch **11** in HOLD position) or a gated stimulus (switch **11** in GATE position) in CC / BRIDGE or CCx10 mode, range:  $\pm 10$  nA (100 = 1 nA) or  $\pm 100$  nA (100 = 10 nA), respectively. Polarity is set by the switch **10**.

**(10) +/- switch**



Switch for setting the polarity of the holding current or gated current stimulus, respectively (see also **9**). In zero position the holding current or gated current stimulus is disabled.

**(11) GATE / HOLD switch**



Switch that determines the function of the CURRENT nA potentiometer **9** (see also **9**).

**(12) BRIDGE BALANCE  $M\Omega$  potentiometer**

Potentiometer for balancing the BRIDGE circuit; 10  $M\Omega$  / turn, range: 0 to 100  $M\Omega$ . The BRIDGE must be balanced correctly before using the series resistance compensation in VC mode, because this measure is taken as value for the series resistance compensation (see also **43** and **44**).

**(13) STIMULUS INPUT switch**

Switch for grounding BNC connector **14**. If **14** is not in use, please switch the INPUT off by setting this switch to OFF. This prevents noise pick-up through an open input.

**(14) STIMULUS INPUT 1 nA/V connector**

BNC connector for the current stimulus in CC / BR or in CCx10 mode; scaling 1 nA / V or 10 nA / V, respectively.

**(15) GATE TTL connector**

BNC connector for gating the potential step in VC / VCx10 mode, or the current step in CC / CCx10 mode.

**VC modes:** As long as the voltage linked to this BNC is HIGH, i.e. +5 V, a voltage stimulus with the amplitude set by the POTENTIAL (mV) potentiometer **21** is generated by the amplifier. Switch **19** has to be in GATE position.

**CC modes:** As long as the voltage linked to this BNC is HIGH, i.e. +5 V, a current stimulus with the amplitude set by the CURRENT (nA) potentiometer **9** is generated by the amplifier. Switch **11** has to be in GATE position.

**(16) COMMAND INPUT switch**

Switch for grounding BNC connector **17**. If **17** is not in use, please switch the INPUT off by setting this switch to OFF. This prevents noise pick-up through an open input.

**(17) COMMAND INPUT  $\div 10$  connector**

BNC connector for the command potential in VC mode or VCx10 mode; scaling:  $\div 10$  mV or  $\div 1$  mV, respectively.

**(18) SERIES RESISTANCE COMP. (%) potentiometer**



Potentiometer for setting the compensation for the SERIES RESISTANCE (VC mode) in percent of the resistance value that is set at the BRIDGE BALANCE potentiometer **12**. Therefore, it is important to set the BRIDGE BALANCE correctly before using the series resistance compensation (see also **40** and **41**).

**Very Important:** When using the SERIES RESISTANCE compensation **do not use** the potential values recorded from the POTENTIAL OUTPUT (mV) BNC connector (**27**) or the value shown at the POTENTIAL display (**39**) as a measure for the membrane potential, since these values include also the additional voltage generated by SERIES RESISTANCE COMP. for compensation of the voltage drop at the SERIES RESISTANCE.

For instance, when for calculating I/V curves **do not use** the potential values recorded from the POTENTIAL OUTPUT (mV) BNC connector (**27**). Use the COMMAND potential instead.

**Note:** In VCx10 mode series resistance compensation is automatically disabled.

**COMMAND set unit**



The COMMAND set unit consist of (**19**) GATE / HOLD switch, (**20**) +/- switch and (**21**) POTENTIAL (mV) potentiometer

**(19) GATE / HOLD switch**



Switch that determines the function of the POTENTIAL (mV) potentiometer **21** (see also **21**).

**(20) +/- switch**



Switch for setting the polarity of the holding potential or gated potential stimulus in VC mode, respectively (see also **21**). In zero position the holding potential or gated potential stimulus is disabled.

**(21) POTENTIAL (mV) potentiometer**



Ten-turn potentiometer for generating a holding potential (switch **19** in HOLD position) or a gated stimulus (switch **19** in GATE position) in VC or VCx10 mode; range:  $\pm 1$  V (100 = 100 mV) or  $\pm 10$  V (100 = 1 V), respectively. Polarity is set by the switch **20**.

**OSCILLATION SHUT OFF unit**



The OSCILLATION SHUT OFF unit consist of (**22**) THRESHOLD potentiometer, (**23**) DISABLED / RESET switch and of (**35**) OSCILLATION SHUT OFF LED.

**(22) THRESHOLD potentiometer**



Control to set the activation **THRESHOLD** of the **OSCILLATION SHUT-OFF** circuit (potentiometer, linear clockwise, range: 0-1200 mV).

**(23) DISABLED/RESET switch**



Switch to **DISABLE** the **OSCILLATION SHUT-OFF** unit or **RESET** the circuit. **RESET** is done if one wants to reset the circuit after previous activation. After resetting the **OSCILLATION SHUT-OFF** unit is active again.

**(35) OSCILLATION SHUT-OFF LED**



Indicates whether the **OSCILLATION SHUT-OFF** circuit is active (LED: red) or not (LED: green).

**Note:** If the **OSCILLATION SHUT-OFF** unit is active and the **SHUT-OFF** was triggered, the output of the amplifier to the headstage is disabled. The amplifier is automatically switched to **CC-OFF** mode and only potential measurement works.

**POTENTIAL OUTPUT FILTER**



The **POTENTIAL OUTPUT FILTER** consist of **(24) HIGHPASS** (Hz) and of **(26) LOWPASS** (Hz), and is used mainly for filtering extracellular signals.

**(24) HIGHPASS (Hz) switch**



16-position rotary switch to select the corner frequency of the **HIGHPASS** filter (range: DC to 3 kHz) for the **POTENTIAL OUTPUT** (mV) connector **27**. In DC position the **HIGHPASS** is disabled.

**(26) LOWPASS (Hz) switch**



16-position rotary switch to select the corner frequency of the **LOWPASS** filter (range: 20 Hz to 20 kHz) for the **POTENTIAL OUTPUT** (mV) connector **27**.

**(25) POTENTIAL OUTPUT FROM HEADSTAGE (V) connector**



BNC connector providing the **POTENTIAL** at the electrode from the headstage. This **POTENTIAL OUTPUT** is not filtered and fixed scaled (x1 V).

**(27) POTENTIAL OUTPUT (mV) connector**



BNC connector providing the **POTENTIAL** at the electrode. The scaling is set by **POTENTIAL OUTPUT GAIN** switch **34**. This **POTENTIAL OUTPUT** is filtered by **(26) LOWPASS** and **(24) HIGHPASS** FILTER.

## CAPACITY COMPENSATION (VC) unit



The CAPACITY COMPENSATION (VC) unit consist of **(28)** TIME CONSTANT ( $\tau$ -FAST) potentiometer and of **(32)** AMPLITUDE (C-FAST) potentiometer. It compensates for the electrode capacity and functions only in VC mode. Tuning is done by application of voltage pulses to the electrode and trying to get the resulting current pulses as fast as possible using potentiometers **28** and **32**.

**(28)** TIME CONSTANT ( $\tau$ -FAST) potentiometer



Potentiometer for setting  $\tau$ -FAST in the electrode capacity compensation circuit.

**(32)** AMPLITUDE (C-FAST) potentiometer



Potentiometer for setting C-FAST in the electrode capacity compensation circuit.

**Note:** CAPACITY COMPENSATION works only for the electrode capacity, not for the capacity of the cell membrane. Therefore, capacitive transients are always present when square shaped pulses are applied in VC mode.

**(29)** GROUND connector



Banana jack providing system GROUND. System GROUND is not connected to the chassis or to protective earth, respectively.

**(30)** POWER switch



Push button to switch the amplifier ON (pushed) or OFF (released).

**(31)** AUDIO volume potentiometer



Potentiometer for setting the volume of the AUDIO monitor for POTENTIAL. Turning the potentiometer clockwise increases the volume.

**(33)** PIPETTE HOLD POTENTIAL (VC)



Ten-turn potentiometer setting the current through the pipette to zero when approaching a cell in VC mode, i.e. the pipette is clamped to its OFFSET potential resulting in zero current flow through the pipette. In VCx10 mode the pipette hold potential does not change. Range is  $\pm 100$  mV and zero position is 5, i.e. the same as the OFFSET potentiometer **50**. This is very convenient since this adjustment can be done starting with the same value as in **50**.



**(34) POTENTIAL OUTPUT GAIN switch**



7-position switch for selecting the amplification for the signal at **(27) POTENTIAL OUTPUT (mV)**; range: x10 to x1000.

**(36) ELECTRODE RESISTANCE TEST push button**



Push button for activating the ELECTRODE RESISTANCE TEST. The ELECTRODE RESISTANCE is determined by application of  $\pm 1$  nA square pulses to the electrode and is shown in  $M\Omega$  at display **39**. Function is indicated by the  $M\Omega$  LED **37**. The ELECTRODE RESISTANCE TEST works in OFF mode as well.

**Hints:** This function can also be used to adjust **(51) CAPACITY COMP. (CC)**. The POTENTIAL OUTPUT FILTER LOWPASS **(26)** should be set to 20 kHz and the HIGHPASS **(24)** to DC.

With an amplifier with switchable headstage (ELC-SWI) the electrode resistance test works also in enhanced (x100) current mode.

**Note:** If the ELC-03XS is equipped with the optional SEAL TEST circuit, the push button is replaced by a three position switch. In the upper position the ELECTRODE RESISTANCE TEST is carried out and in the lower position the SEAL resistance test is accomplished. The value of the SEAL resistance is shown at the CURRENT display **(45)**.

**(37)  $M\Omega$  LED**



LED indicating that the unit of the POTENTIAL/RESISTANCE display **(39)** is  $M\Omega$ .

**(38) mV LED**



LED indicating that the unit of the POTENTIAL/RESISTANCE display **(39)** is mV.

**(39) POTENTIAL / RESISTANCE display**



Display for the potential at the electrode in  $\pm XXXXX$  mV ( $\pm 1999$  mV max.) or the electrode resistance in  $XXX M\Omega$  ( $999 M\Omega$  max.). The unit is indicated by **37** or **38**.

**SERIES RESISTANCE COMPENSATION unit**



The SERIES RESISTANCE COMPENSATION unit consist of **(18) SERIES RESISTANCE COMP. (%) potentiometer**, **(40)  $R_s$  COMP. switch** and **(41)  $R_s$  COMP. LED**



(18) SERIES RESISTANCE COMP. (%) potentiometer



Potentiometer for setting the compensation for the SERIES RESISTANCE in percent of the value that is set at the BRIDGE BALANCE potentiometer 12. Therefore, it is important to set the BRIDGE BALANCE correctly before using the series resistance compensation to avoid oscillations (see also 40 and 41).

(40) R<sub>s</sub> COMP. switch



Switch for activating the SERIES RESISTANCE COMPENSATION circuit. The active circuit is indicated by 41.

(41) R<sub>s</sub> COMP. LED

LED indicating that the SERIES RESISTANCE COMPENSATION circuit is active. The amount of SERIES RESISTANCE COMPENSATION is set by 18.

**Very Important:** When using the SERIES RESISTANCE compensation **do not use** the potential values recorded from the POTENTIAL OUTPUT (mV) BNC connector (27) or the value shown at the POTENTIAL display (39) as a measure for the membrane potential, since these values include also the additional voltage generated by SERIES RESISTANCE COMP. for compensation of the voltage drop at the SERIES RESISTANCE.

For instance, when for calculating I/V curves **do not use** the potential values recorded from the POTENTIAL OUTPUT (mV) BNC connector (27). Use the COMMAND potential instead.

**Note:** In VCx10 mode series resistance compensation is automatically disabled.

(42) MODE OF OPERATION switch and LEDs



Switch for selecting the MODE OF OPERATION

VC x10: the amplifier operates in Voltage (Patch) Clamp mode. The COMMAND POTENTIAL is enhanced by a factor of ten.

**Note:** In VCx10 mode series resistance compensation is automatically disabled.

VC: the amplifier operates in Voltage (Patch) Clamp mode

**Important:** VC modes do not function properly with sharp microelectrodes, i.e. electrodes with more than 10 MΩ resistance.

OFF: all inputs of the amplifier are switched OFF, and the amplifier is set to CC mode. REL test and potential measurement work

CC: the amplifier operates in Current Clamp mode

CC x10: the amplifier operates in Current Clamp mode. The STIMULUS amplitude is enhanced by a factor of ten.

EXT: the amplifier is set to CC mode. R<sub>s</sub>, R<sub>EL</sub> test, BR, x10 or VC / CC modes can be selected by application of a TTL HIGH (+5 V) signal to the respective BNC at the rear panel (see also chapter 4).

The MODE OF OPERATION that is currently activated, is indicated by the respective LED above the switch.

**BRIDGE unit**



The BRIDGE unit consist of (12) BRIDGE BALANCE MΩ potentiometer, (43) BRIDGE MODE LED and (44) BRIDGE MODE switch.

**(12) BRIDGE BALANCE MΩ potentiometer**



Potentiometer for balancing the BRIDGE circuit; 10 MΩ / turn, range: 0 to 100 MΩ. The BRIDGE must be balanced correctly before using the series resistance compensation in VC mode, because this measure is taken as value for the series resistance compensation (see also 43 and 44).

**(43) BRIDGE MODE LED**



LED indicating that the BRIDGE circuit is active.

**(44) BRIDGE MODE switch**

Switch for activating the BRIDGE circuit. The active circuit is indicated by 43.

**(45) CURRENT (nA) display**



Display for the current at the electrode in ±XX.XX nA, i.e. 10.00 is 10 nA (19.99 nA max.)

**Note:** If the ELC-03XS is equipped with the optional SEAL TEST circuit, the value of the SEAL resistance is also shown at the CURRENT display in XX.XX GΩ (19.99 GΩ max.).

**(46) CURRENT OUTPUT SENSITIVITY (V/nA) switch**



7-position switch for selecting the amplification of the current output signal in V/nA; range 0.1 V/nA to 10 V/nA.

**(49) BIAS (CC) potentiometer**



Ten-turn potentiometer for BIAS current cancellation of the headstage; (ten-turn potentiometer, symmetrical, i.e. 0 pA = 5 on the dial), range: ±100 pA.

**(50) OFFSET potentiometer**



Ten-turn potentiometer for OFFSET cancellation of the electrode; (ten-turn potentiometer, symmetrical, i.e. 0 mV = 5 on the dial), range: ±100 mV.

(51) CAPACITY COMP. (CC) potentiometer



Ten-turn potentiometer for the capacity compensation of the electrode in CC mode (ten turn potentiometer, clockwise, range: 0-30 pF).

4. Description of the Rear Panel

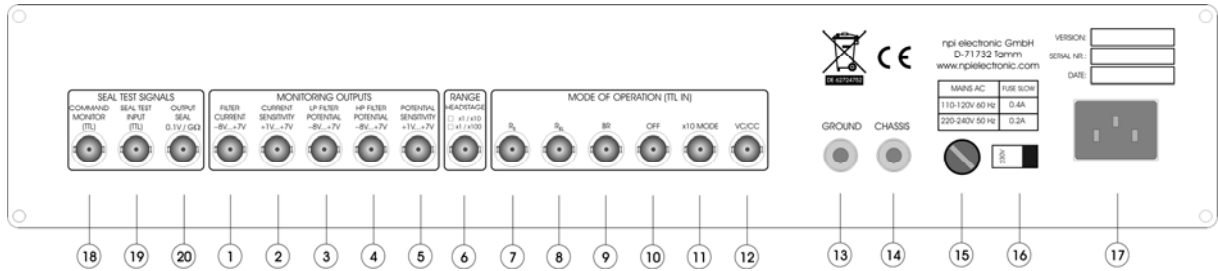


Figure 3: ELC-03XS rear panel view

**MONITORING OUTPUT** connectors

(1) FILTER CURRENT connector

BNC connector providing a voltage monitoring the position of the CURRENT FILTER switch (-7 V to +8 V, 1V/STEP).

(2) CURRENT SENSITIVITY connector

BNC connector providing a voltage monitoring the position of the CURRENT OUTPUT SENSITIVITY switch (+1 V to +7 V, 1V/STEP).

(3) LP FILTER POTENTIAL connector

BNC connector providing a voltage monitoring the position of the POTENTIAL LOWPASS FILTER switch (-8 V to +7 V, 1V/STEP).

(4) HP FILTER POTENTIAL connector

BNC connector providing a voltage monitoring the position of the POTENTIAL HIGHPASS FILTER switch (-8 V to +7 V, 1V/STEP).

(5) POTENTIAL SENSITIVITY connector

BNC connector providing a voltage monitoring the position of the POTENTIAL OUTPUT GAIN switch (+1 V to +7 V, 1V/STEP).

**RANGE** connector

(6) HEADSTAGE connector

BNC connector for remote control of a switchable headstage (ELC-SWI, optional). A TTL HI (+5 V) signal switches the feedback resistance in the switchable headstage from 1 MΩ to 10 MΩ or from 1 MΩ to 100 MΩ. To which resistor is indicated by a box to the left.

**MODE SELECT** connectors (see also chapter 3.4)

All MODEs OF OPERATION can be selected by TTL signals connected to the rear panel (see below), if the MODE OF OPERATION switch (**42**, Figure 1) is in EXT position. This is very convenient when switching often between electroporation and recording, because this can be done automatically by the data acquisition system using TTL signals.

**(7) R<sub>S</sub>** connector

BNC connector for remote control of the series resistance compensation circuit. A TTL HI (+5 V) signal is connected here to activate the series resistance compensation circuit remotely.

**(8) R<sub>EL</sub>** connector

BNC connector for remote control of the electrode resistance test. A TTL HI (+5 V) signal can be connected here to select the electrode resistance test remotely.

**(9) BR** connector

BNC connector for remote control of the bridge mode. A TTL HI (+5 V) signal can be connected here to select the bridge mode remotely.

**(10) OFF** connector

BNC connector to switch the ELC-03XS in OFF mode remotely with a TTL HI (+5 V) signal.

**(11) x10 MODE** connector

BNC connector to switch the ELC-03XS to x10 mode of operation remotely (TTL HI signal). Dependent on the signal level at **12**, this is CCx10 (signal at **12** = LOW) or VCx10 (signal at **12** = HI).

**(12) VC / CC** connector

BNC connector for remote control of the VC / CC mode of operation. A TTL signal can be connected here to select the mode of operation remotely (HI = VC, LO = CC).

**(13) GROUND** connector

Banana plug providing internal ground (see below).

**(14) CHASSIS** connector

Banana plug providing mains ground (see below).

**(15) FUSE** holder

Holder for the line fuse. For changing the fuse rotate the holder counter-clockwise using a screw driver.

**(16) LINE SELECT** switch

Switch for selecting the line voltage. Switch to the right for 230 V AC, to the left for 115 V AC. The selected voltage is indicated on the switch.

**Caution:** Before turning on the instrument, make sure that the correct line voltage is selected.

**(17) Mains connector**

Plug socket for the mains power-plug.

***Important:*** Check line voltage before connecting the ELC amplifier to power. Always use a three-wire line cord and a mains power-plug with a protection contact connected to ground. Disconnect mains power-plug when replacing the fuse or changing line voltage. Replace fuse only by appropriate specified type (one spare fuse is supplied). Before opening the cabinet unplug the instrument.

**SEAL TEST SIGNALS** connectors (optional)

The SEAL resistance is determined similar to the  $R_{EL}$  test.

$\pm 10$  mV square pulses with 15 Hz are applied to the pipette and the resulting current is measured. The resistance is calculated according to Ohm's law and is indicated on the CURRENT DISPLAY. The maximum SEAL resistance that can be displayed is 19.99 G $\Omega$ . The value of the SEAL resistance is also monitored at **20** (see below).

**(18) COMMAND MONITOR (TTL) connector**

BNC connector providing a TTL (+5V) signal synchronous to the  $\pm 10$  mV test pulses.

**(19) SEAL TEST INPUT (TTL) connector**

Starts seal test remotely (see also **36**, Figure 1).

**(20) OUTPUT SEAL 0.1V / G $\Omega$  connector**

BNC connector monitoring the value of the SEAL resistance; scaling 100 mV / G $\Omega$ .

*Grounding*

ELC instruments have two ground systems:

1. the internal ground (called internal GROUND) represents the zero level for the recording electronics and is connected to the recording chamber and the BNC input/output sockets
2. mains ground (CHASSIS) is connected to the 19" cabinet and through the power cable to the protection contact of the power outlet.

Both grounds are provided at the rear panel:

GROUND (black socket): internal system ground

CHASSIS (green/yellow socket): mains ground, 19" cabinet

All ELC systems have a high quality toroid transformer in order to minimize stray fields. In spite of this, noise problems could occur if other mains-operated instruments are used in the same setup. The internal system ground (GROUND sockets) should be connected to only one point on the measuring ground. Multiple grounding should be avoided, i.e. all ground points should originate from a central point to avoid ground loops.

## 5. Setting up the ELC-03XS

The following steps should help you set up the ELC-03XS correctly. Always adhere to the appropriate safety measures (see chapter 1).

After unpacking, the ELC-03XS is attached to the setup by assembling the electrical connections. It is assumed that first a cell model will be attached.

### ① Electrical connections

- Turn POWER off.
- Plug the power cord of the instrument into a grounded outlet.
- Connect the headstage to the HEADSTAGE connector (#1, Figure 1) at the ELC-03XS.
- Connect a cell model (see chapter 6). Connect a digital/analog timing unit or a stimulation device to STIMULUS INPUT or to GATE TTL if you intend to use a gated stimulus.
- Connect a store oscilloscope or a data acquisition system to the POTENTIAL OUTPUT and to the CURRENT OUTPUT triggered from the stimulation device. Set the desired gain at the POTENTIAL OUTPUT GAIN switch (#34, Figure 1) and the CURRENT OUTPUT SENSITIVITY switch (#46, Figure 1).

Before using the ELC-03XS always make the basic settings to avoid oscillations.

### ② Basic settings

- Turn all controls to low values (less than 1) and the OFFSET and BIAS controls in the range of 5 (zero position, see chapter 3.4).
- Set the MODE OF OPERATION switch (#42, Figure 1) to CC.
- Turn POWER switch on.

Now the ELC-03XS is ready for an initial check with the cell model.

## 6. Passive Cell Model

The ELC-03XS can be ordered with a passive cell model as an optional accessory. An active cell model is also available on request (for ref. see Draguhn et al. (1997)).

The passive cell model is designed for use with single electrode amplifiers (BA series, ELC series) to check the function of the instrument in the following circumstances:

1. just after unpacking to see whether the instrument has been damaged during transport or
2. to train personnel using the instrument or
3. in case of trouble to check which part of the setup does not work correctly, e.g. to find out whether the amplifier or headstage is damaged, or something is wrong with the electrodes or holders etc.

The passive cell model consists only of passive elements, i.e. resistors that simulate the resistance of the cell membrane and the electrodes, and capacitances that simulate the capacitance of the cell membrane. A switch allows simulation of two different cell types: a cell with 50 M $\Omega$  and 22 pF (CELL 1, represents an astrocyte like cell) or a “small” cell with 200 M $\Omega$  membrane resistance and 100 pF membrane capacitance (CELL 2, represents a neuron like cell). Electrode immersed into the bath or SEAL formation can be mimicked as well. The headstage of the amplifier can be connected to one of two different types of electrodes (see below).

### 6.1. Cell Model Description

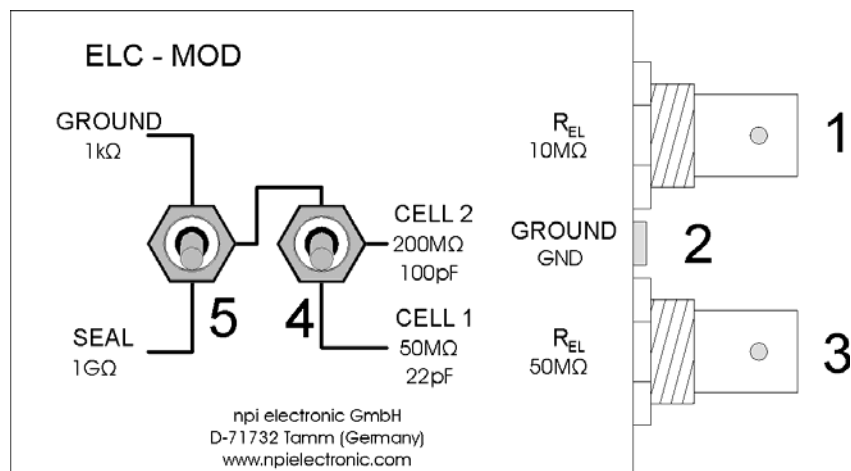


Figure 4: passive cell model

- 1, 3:** connectors for the headstage, **1:** electrode resistance: 50 MΩ, **3:** electrode resistance: 10 MΩ
- 2:** GND ground connector, to be connected to GND jack of the headstage
- 4:** CELL: switch for cell membrane representing a membrane of either 50 MΩ and 22 pF (CELL 1) or 200 MΩ and 100 pF (CELL 2).
- 5:** In GROUND (upper) position the electrodes are connected to ground via a 1 kΩ resistor. In SEAL (lower) position are connected to a 1 GΩ resistor simulating the formation of a GIGASEAL with a patch electrode.

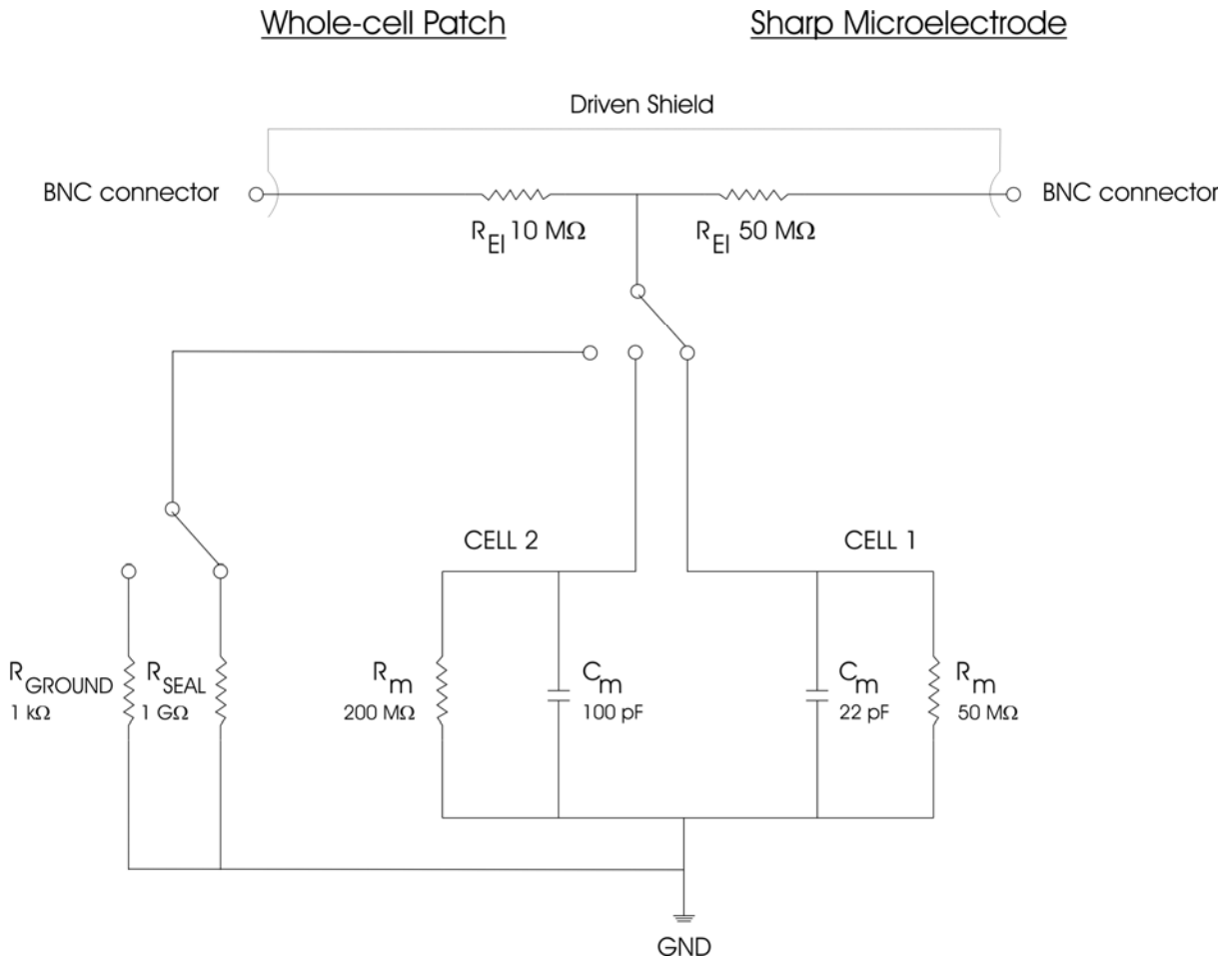


Figure 5: Schematic diagram of the passive cell model

## 6.2. Connections and Operation

It is assumed that all connections are built as described in chapter 5.

### Checking the configuration

- Turn POWER switch of the amplifier off.
- a) For simulation of an experiment using a suction electrode



- Connect the BNC jack labeled 10M $\Omega$  of the cell model to the BNC connector P<sub>EL</sub> of the headstage.
- b) For simulation of an experiment using a sharp electrode
- Connect the BNC jack labeled 50M $\Omega$  of the cell model to the BNC connector P<sub>EL</sub> at the headstage. For headstages with SMB connector use the supplied SMB to BNC adapter.

For a) and b)

- Connect GND of the cell model to GND of the headstage.

***Important:*** When using the **differential headstage (optional)** the **REF connector** must not be left open. It must be connected to ground.

#### Simulation of electrode in the bath

- Set switch #4, Figure 4 to the upper position.
- Set switch #5, Figure 4 to GROUND position. The 1 k $\Omega$  resistor simulates the resistance of the bath solution. This can be used to train cancellation of offsets, using the bridge balance and using the capacity compensation.

#### Simulation of SEAL formation

- Set switch #4, Figure 4 to the upper position.
- Set switch #5, Figure 4 to SEAL position. The 1 G $\Omega$  resistor simulates the SEAL resistance when forming a GIGASEAL in patch clamp experiments.

#### Simulation of intracellular recording

Intracellular recordings can be mimicked with one of two cells with different properties. Use the 50 M $\Omega$  electrode connector (#3, Figure 4) for an experiment with sharp electrodes or the 10 M $\Omega$  electrode connector (#1, Figure 4) for simulating an experiment with patch electrodes.

- Switch the CELL membrane switch (see #4, Figure 4) to the desired position (CELL 1 or CELL 2).
- Turn all controls at the amplifier to low values (less than 1) and the OFFSET in the range of 5 (zero position) and the OSCILLATION SHUTOFF in the DISABLED position.
- Turn POWER switch of the amplifier on.

Now you can adjust the amplifier (see below) and apply test pulses to the cell model. The lower position of the CELL membrane switch (CELL 1) simulates a cell with a resistance of 50 M $\Omega$  and a capacitance of 22 pF. In the middle position (CELL 2) a cell membrane with 200 M $\Omega$  and 100 pF is simulated.

## 7. Headstage

The ELC-03XS comes with a headstage for connecting suction electrodes for loose-patch clamp or whole cell recordings and / or stimulation or electroporation, respectively or sharp electrodes for intracellular or extracellular recordings. The use of metal electrodes is possible as well.

A differential (miniature) headstage (see **Optional accessories** in chapter 3.1) for measurements *in vivo* is also available (see also chapter 8.1 and contact npi for details).

### 7.1. Headstage Elements

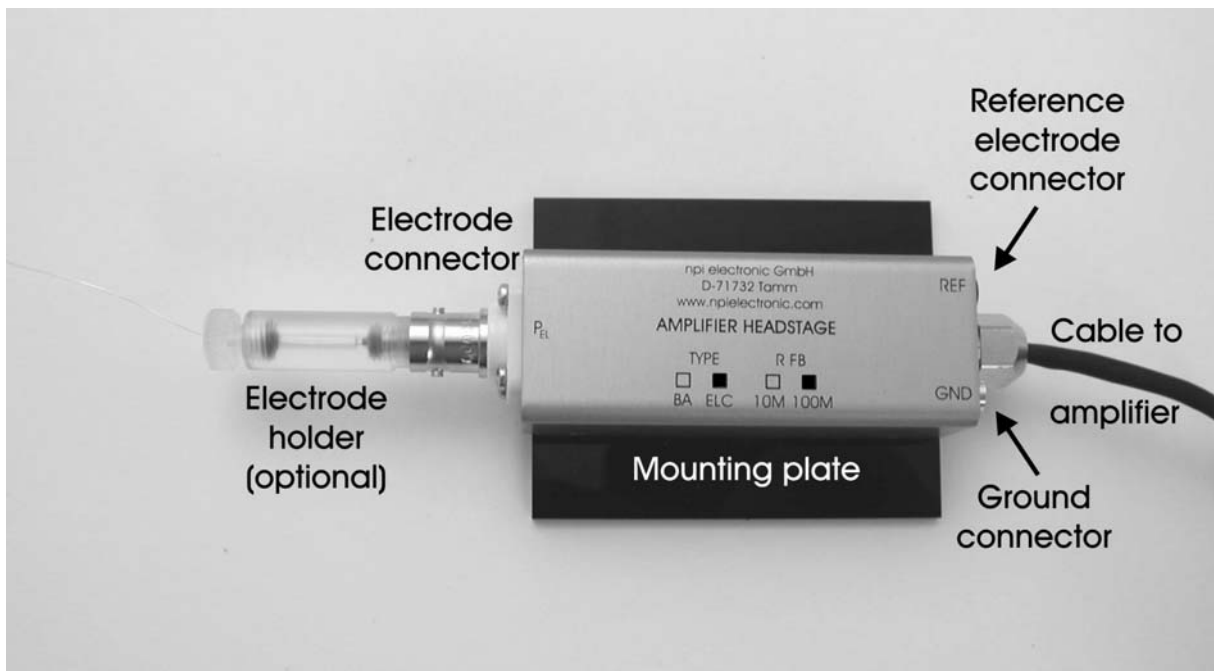


Figure 6: ELC-03XS headstage

- P<sub>EL</sub> BNC connector for the electrode holder
- REF Connector for the reference electrode (differential headstage only)
- GND Ground connector
- TYPE Type of amplifier, BA → Bridge amplifier, ELC → ELC amplifier
- R FB Value of feedback resistor, 10M: 10 MΩ, 100 M: 100 MΩ
- Headstage cable to amplifier
- Mounting plate

The electrode filled with electrolyte is inserted into an electrode holder (optional) that fits into the BNC connector of the headstage or into an electrode holder adapter. The electrical connection between the electrolyte and the headstage is established using a carefully chlorinated silver wire. Chlorinating of the silver wire is very important since contact of silver to the electrolyte leads to electrochemical potentials causing varying offset potentials at the electrode, deterioration of the voltage measurement etc. (for details see Kettenmann and Grantyn (1992)). For optimal chlorinating of silver wires an automated chlorinating apparatus (ACI-01) is available (contact npi for details).

GND provides system ground and is linked to the bath via an agar-bridge or a Ag-AgCl pellet. The headstage is attached to the amplifier with the headstage cable and an 8-pole connector. The headstage can be mounted directly to a micromanipulator using the mounting plate, a dove tail or a holding bar.

***Important:*** The shield of the BNC connector is linked to the driven shield output and must not be connected to ground. The headstage enclosure is grounded.

***Caution:*** Please always adhere to the appropriate safety precautions (see chapter 1). Please turn power off when connecting or disconnecting the headstage from the HEADSTAGE connector!

## 7.2. Headstage Bias Current Adjustment

***Caution:*** It is important that this tuning procedure is performed ONLY after a warm-up period of at least 30 minutes!

The ELC-03XS is equipped with a voltage-to-current converter with a very high output impedance which is connected to the recording electrode. The zero current of this unit is tuned with the BIAS current potentiometer (#49, Figure 1).

The tuning procedure should be performed regularly (at least once a month) since the BIAS current changes over time.

The tuning procedure is performed using high-value resistors and/or a cell model. It cannot be performed with an electrode, since there are always unknown potentials involved (tip potential, junction potentials).

- Disconnected **all** input signals (except the headstage). Put the CURRENT (nA) switch (10, Figure 1) to position 0. Set the operation mode to CC.
- Set the BIAS control potentiometer to 5 (zero position).
- Connect the PEL connector of the headstage to ground.

***Note:*** This cannot be done with the cell model. Please use a wire to connect the input of the BNC connector on the headstage to GND of the headstage. Do not use the shield of the BNC connector since it is connected to driven shield.

- Tune the OFFSET to zero using the OFFSET control.

***Remember:*** The zero position of the OFFSET control is at 5!!

- Remove the wire and attach the cell model or a resistor with a value of about 5 M $\Omega$  across the same connection.
- The value displayed at the POTENTIAL DISPLAY is related to the BIAS current of the headstage according to Ohm's Law. Cancel this voltage by tuning the headstage BIAS current potentiometer until the POTENTIAL DISPLAY shows 000.

### 7.3. Headstage with x10 / x100 current range (option)

Optionally, the ELC-03XS amplifier has an extended current range, selectable by switch or TTL. This is realized by reduction or increase of the headstage feedback resistance. It enables the amplifier to generate several current ranges and voltages with low resistance electrodes, e.g. for electroporation, both in current clamp and in voltage clamp mode.



Figure 7: ELC-XS with switchable headstage, front panel view

The x10 / x100 current range can be set at the front panel of the ELC-03XS with an additional switch labeled RANGE x10 or with a TTL signal applied to the x10 INPUT (TTL) BNC connector at the rear panel of the amplifier (see also chapter 4). If the switch is set to x100 position the current range is  $\pm 12 \mu\text{A}$ . If the switch is set to x10 position the current range is reduced to  $\pm 1.2 \mu\text{A}$ . This mode is indicated at the headstage by an additional LED (x10 ON, see Figure 9).

**Note:** In VCx10 mode series resistance compensation is automatically disabled.

**Note:** All basic operations and settings (e.g. bridge balance or BIAS adjustment) should be performed in standard x100 mode, i.e. RANGE x10 switch set to x100!

**Exception:** The electrode resistance test works well in x100 and x10 mode!!

If the ELC-03XS operates in x100 high current mode, all current related signals have to be corrected by the factor of hundred:

- CURRENT OUTPUT SENSITIVITY (V / nA): 0.001, 0.002, 0.005, 0.01, 0.02, 0.05, 0.1
- STIMULUS INPUT: 100 nA / V (CC x1) or  $1 \mu\text{A} / \text{V}$  (CC x10)
- Holding CURRENT / gated CURRENT: 0 to 999 nA (CC x1) or  $9.99 \mu\text{A}$  (CC x10)
- CURRENT FROM HEADSTAGE: 0.001 V / nA
- CURRENT DISPLAY: shows always nA, scaling is adapted automatically.

If the ELC-03XS operates in x10 current mode, all current related signals have to be corrected by the factor of ten:

- CURRENT OUTPUT SENSITIVITY (V / nA): 0.01, 0.02, 0.05, 0.1, 0.2, 0.5, 1
- STIMULUS INPUT: 10 nA / V (CC x1) or  $100 \text{nA} / \text{V}$  (CC x10)
- Holding CURRENT / gated CURRENT: 0 to 99.9 nA (CC x1) or 999 nA (CC x10)
- CURRENT FROM HEADSTAGE: 0.01 V / nA
- CURRENT DISPLAY: shows always nA, scaling is adapted automatically.



Figure 8: Current range switch



Figure 9: ELC x10 / x100 switchable headstage

**Important:** Although the ELC-03XS is modified for the x10 / x100 switchable headstage it can also be used as a standard amplifier with a non-switchable headstage. In this case the amplifier will recognize that such a headstage is connected, and the additional current switch at the front panel and the TTL input at the rear panel are automatically disabled.

## 8. Introduction into Experiments

The ELC-03XS is capable to perform several types of experiments that are briefly introduced in the following with special focus on loose-patch stimulation and recording. It is assumed that the capacity of the electrode is compensated, the offset of the electrode is cancelled and, for intracellular recordings in BRIDGE mode, electrode artifact is eliminated using the bridge balance circuit. Particularly, when working with charged substances and approaching the cell in VC mode, the current offset is cancelled using the PIPETTE HOLD POTENTIAL potentiometer (33, Figure 1).

### 8.1. Recordings with the Differential Headstage (optional)

Extracellular measurements are mostly done in slices or *in vivo*, in noisy environments, where distortions of the recorded signal caused by other instruments and the animal itself are very common. Additionally, extracellular signals are very small and have to be amplified enormously. The drawback is that noise is amplified as well. Therefore, the headstage of the ELC 03X can be equipped with a differential input that minimizes noise pick-up. Differential means, that the signal for the amplifier is the difference between the positive (+) (P<sub>EL</sub>) and negative (-) (REF.) input of the headstage. This results in canceling of all common mode signals (i.e. which both electrodes record, e.g. noise). P<sub>EL</sub> is connected to the measuring electrode and REF. to the reference electrode. The experimental chamber is grounded by an Ag-AgCl pellet (or an AGAR bridge) connected to GND of the headstage (see Figure 10).

**Important:** For reliable measurements the experimental chamber or animal, respectively must be grounded.

**Also Very Important:** When operating in VC mode, the reference electrode must not have a resistance of more than 1 kΩ!! CC mode is not affected.

If differential measurement is not required (single-ended measurement configuration, see Figure 10), the REF input must be connected to ground (GND). The amplifier is in an undefined state, if the REF is left open, and can go into saturation making reliable measurements impossible (for more details see Lalley et al., 1999).

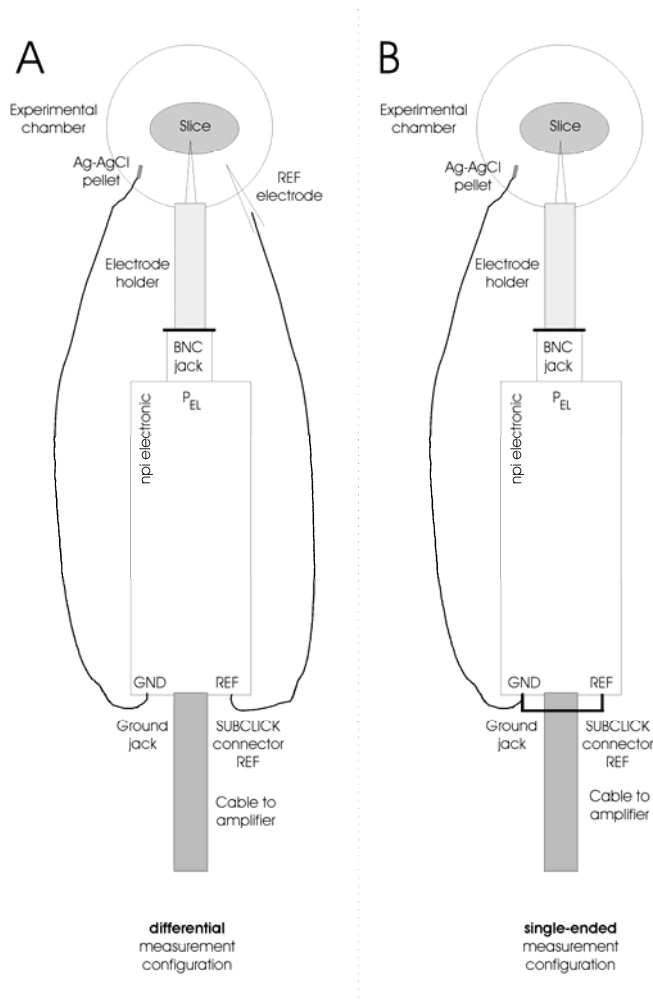


Figure 10: headstage connections, **A**: differential measurement, **B**: single-ended measurement

## 8.2. Extracellular Voltage Measurement

Extracellular measurements are usually done in the loose-patch configuration or with special metal microelectrodes. Recordings with extracellular metal electrodes is simple. The electrode is advanced into the region where the recordings will be made using a micromanipulator and the signals are filtered and amplified (see chapter 5 in Lalley et al., 1999 for details) as required. For loose patch recording the basic procedure is the following (Barbour & Isope, 2000, Nunemaker et al, 2003):

- Approach the cell in VC mode and apply square voltage pulses to the electrode.
- Contact the cell and establish the loose-patch.
- Set the MODE OF OPERATION switch to OFF.
- Set the required amplification of the POTENTIAL OUTPUT.
- Set the HIGHPASS FILTER to the desired corner frequency, e.g. 0.3 Hz.
- Set the LOWPASS FILTER to the desired corner frequency, e.g. 3 kHz.

### **8.3. Extracellular Stimulation and Electroporation**

Cells can be stimulated using current or voltage signals.

#### *Stimulation with Current*

- Approach the cell in VC mode and apply square voltage pulses to the electrode.
- Contact the cell, establish the loose-patch and disconnect the voltage signal from the COMMAND INPUT :10 mV connector.
- Set the MODE OF OPERATION switch to CC or CCx10.
- Set the HOLDING CURRENT to zero.
- For stimulation:

Apply the stimulus signal to the STIMULUS INPUT 1 nA/V connector.

or

Adjust the stimulus amplitude with the HOLDING CURRENT potentiometer and set the stimulus polarity using the switch aside. Gate the preset stimulus with a TTL signal linked to the GATE TTL BNC connector.

#### *Electroporation with Current*

Electroporation can be done using the stimulation procedure, but usually the applied current is much higher and the stimulus duration is shorter. Therefore, most electroporation experiments are done in CCx10 mode.

#### *Stimulation with Voltage*

- Approach the cell in VC mode and apply square voltage pulses to the electrode.
- Contact the cell and establish the loose-patch.
- For stimulation apply a voltage signal of the required amplitude and duration to the COMMAND INPUT :10 mV connector.

or

Adjust the stimulus amplitude with the HOLDING POTENTIAL potentiometer and set the stimulus polarity using the switch aside. Gate the preset stimulus with a TTL signal linked to the GATE TTL BNC connector.

#### *Electroporation with Voltage*

Electroporation can be done using the stimulation procedure, but usually the applied voltage is much higher and the stimulus duration is shorter. For stimuli with large amplitudes set the MODE OF OPERATION switch to VCx10 enabling electroporation with up to 12 V.

## 8.4. Intracellular Recording

Intracellular recordings can be performed in the whole-cell patch configuration and with sharp microelectrodes.

**Note:** VC mode does not function properly with sharp microelectrodes, i.e. electrodes with more than 10 M $\Omega$  resistance.

### *Current Clamp Recording*

The ELC-03XS can be used like a standard bridge amplifier.

- Set the MODE OF OPERATION switch to CC and the BRIDGE MODE switch to the upper position. The BRIDGE MODE LED lights up.
- Compensate the electrode artifact using the BRIDGE BALANCE potentiometer.
- After impaling the cell readjust the bridge.
- If needed set an appropriate holding current using the HOLDING CURRENT potentiometer and the HOLDING CURRENT polarity switch.
- Apply stimuli to the cell using the STIMULUS INPUT 1 nA/V BNC connector.

### *Voltage Clamp Recording*

The ELC-03XS can also be used like a whole-cell patch-clamp amplifier.

- Approach the cell in VC mode and apply square voltage pulses to the electrode.
- Contact the cell, set a holding potential using the HOLDING POTENTIAL potentiometer and establish the whole-cell patch clamp configuration.
- Set the amplifier to BR mode and adjust the BRIDGE. Only if the BRIDGE is adjusted correctly the SERIES RESISTANCE COMPENSATION works with the correct value.
- Switch to VC mode.
- If necessary, adjust the shape of the voltage signal using the CAPACITY COMPENSATION (VC) potentiometer (#22 and #27, Figure 1) and compensate for the SERIES RESISTANCE.
- Apply stimuli to the cell using the COMMAND INPUT :10 mV BNC connector.

**Note:** CAPACITY COMPENSATION works only for the electrode capacity, not for the capacity of the cell membrane. Therefore, capacitive transients are always present when square shaped pulses are applied in VC mode.



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## 10. Technical Data

### Headstage:

Input voltage range:	±12 V
Operating voltage:	±15 V
Enclosure:	Size: 23 x 70 x 26 mm, grounded
Mounting plate:	Size: 70 mm x 50 mm
on request	
Holding bar:	length 150 mm, Ø 8 mm
Electrode connector:	BNC with driven shield
Ground connector:	2.4 mm connector
Input resistance (CC):	>10 <sup>13</sup> Ω (internally adjustable)
Current range:	±1.2 μA max. (10 MΩ feedback, x10 range) ±12 μA max(1 MΩ feedback, x100 range)

### Electrode parameter controls:

OFFSET:	range ±100 mV, ten-turn control
PIPETTE HOLD POTENTIAL (in VC):	range ±100 mV, ten-turn control
CAPACITY COMPENSATION:	range 0 – 30 pF, ten-turn control
BIAS:	range ±100 pA, ten-turn control

### Bridge balance:

0-100 MΩ	adjustable with ten-turn control
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### Electrode resistance test:

Sensitivity 1 mV / MΩ	application of square current pulses ±1 nA
Display:	3 ½ digit, XXX MΩ, activated by key switch (same as POTENTIAL display)

### Bandwidth and speed response (CC mode, optimal capacity compensation):

Full power bandwidth (R <sub>EL</sub> = 0 MΩ):	>30 kHz, rise time (10% - 90%) <10 μs (R <sub>EL</sub> = 100 MΩ) <5 μs (R <sub>EL</sub> = 10 MΩ)
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### Outputs:

Output impedance:	50 Ω
Max. voltage:	±12 V
Current output:	BNC connector, sensitivity 0.1...10 V/nA,
Current output sensitivity:	Rotary switch, 0.1, 0.2, 0.5, 1, 2, 5, 10 V/nA
Current display:	3 ½ digits, XX.XX nA, resolution 10 pA
Current LP filter:	4-pole BESSEL filter (other options available)
attenuation:	-24 dB/octave,
corner frequencies (Hz):	20, 50, 100, 200, 300, 500, 700, 1k, 1,3k, 2k, 3k, 5k, 8k, 10k, 13k, 20k
Potential output x1:	BNC connector, sensitivity 1 V/V
Potential output:	BNC connector, sensitivity 10...1k V/V
Potential output gain:	Rotary switch, 10, 20, 50, 100, 200, 500, 1k
Potential output resolution in AC:	50 μV

Potential LP filter:	4-pole BESSEL filter (other options available)
attenuation:	-24 dB/octave,
corner frequencies (Hz):	20, 50, 100, 200, 300, 500, 700, 1k, 1,3k, 2k, 3k, 5k, 8k, 10k, 13k, 20k
Potential HP filter:	1-pole filter, (other options available)
attenuation:	-6 dB/octave
corner frequencies (Hz):	DC, 0.1, 0.3, 0.5, 1, 3, 5, 10, 30, 50, 100, 300, 500, 800, 1k, 3k
Telegraph potential LP filter	-8...+7 V, 1V/step
Telegraph potential HP filter	-8...+7 V, 1V/step
Telegraph current filter	-8...+7 V, 1V/step
Telegraph potential output sensitivity	+1...+7 V, 1 V/ step
Telegraph current output sensitivity	+1...+7 V, 1 V/ step

Digital displays:

Display mV/MΩ	3 ½ digits, XXXX mV or XXX MΩ
Display current	3 ½ digits, XXX.X nA (range x10) or XXXX nA (range x100)

Inputs:

Input impedance analog	100 kΩ
Input range	±12 V
Input impedance digital (TTL)	10 kΩ
Input range TTL	0-5 V
Current stimulus input CC	via BNC connectors, sensitivity 1 nA / V
Current stimulus input CCx10	via BNC connectors, sensitivity 10 nA / V
Step gate input	via BNC connector (TTL)
Gated stimulus CC	with ten-turn control of holding current resolution: 10 pA, range: ±10 nA
Gated stimulus CCx10	with ten-turn control of holding current resolution: 100 pA, range: ±100 nA
Polarity	selectable with toggle switch
Voltage command input VC	via BNC connectors, sensitivity: ÷10 mV
Voltage command input VCx10	via BNC connectors, sensitivity: ÷1 mV
Step gate input	via BNC connector (TTL)
Gated stimulus VC	with ten-turn control of holding potential resolution: 1 mV, range: ±1 V
Gated stimulus VCx10	with ten-turn control of holding potential resolution: 10 mV, range: ±10 V
Polarity	selectable with toggle switch

Dimensions:

19" rackmount cabinet  
19" (483 mm), 10" (250 mm), 3.5" (88 mm)

Power requirements:

115/230 V AC, 60/50 Hz, fuse 0.4/0.2 A, slow, 25 W

Weight: 4.5 kg

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