

## K-7400S Semi-Micro Osmometer

### ► Freezing Point Osmometer User Manual

V3700



**Note:** For your own safety, **read** the manual and **always** observe the warnings and safety information on the device and in the manual

**Manuel en français:** Si jamais vous préférez un manuel en français pour ce produit, veuillez vous contacter le support technique (Technical Support) par email ou par fax avec le no. de série. Merci beaucoup.

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For latest version of the manual, check our website:  
<http://www.knauer.net/knowledge/user-manuals.html>



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# Product Information

**Note:** Only use the device for applications that fall within the range of the intended use. Otherwise, the protective and safety equipment of the device could fail.

## Views

### Front View

#### Legend

- ① Measuring head
- ② LCD display
- ③ Keypad



Fig. 1 Front view

### Keypad

Buttons	Function
<b>START/STOP</b>	Starts a measurement or calibration run. If this button has no specific function on the screen, it causes to return on the main screen. <b>Note:</b> With this button, every running measurement can be interrupted.
<b>VIB</b>	Activates the stirring wire. During RUN and CALIBRATE, the button is without any function.
<b>Arrows</b>	Cursor navigation buttons for parameter or value selection

## Rear View

### Legend

- ① RS-232 connector
- ② Measuring head connector
- ③ Ventilator
- ④ Power switch
- ⑤ Power connector

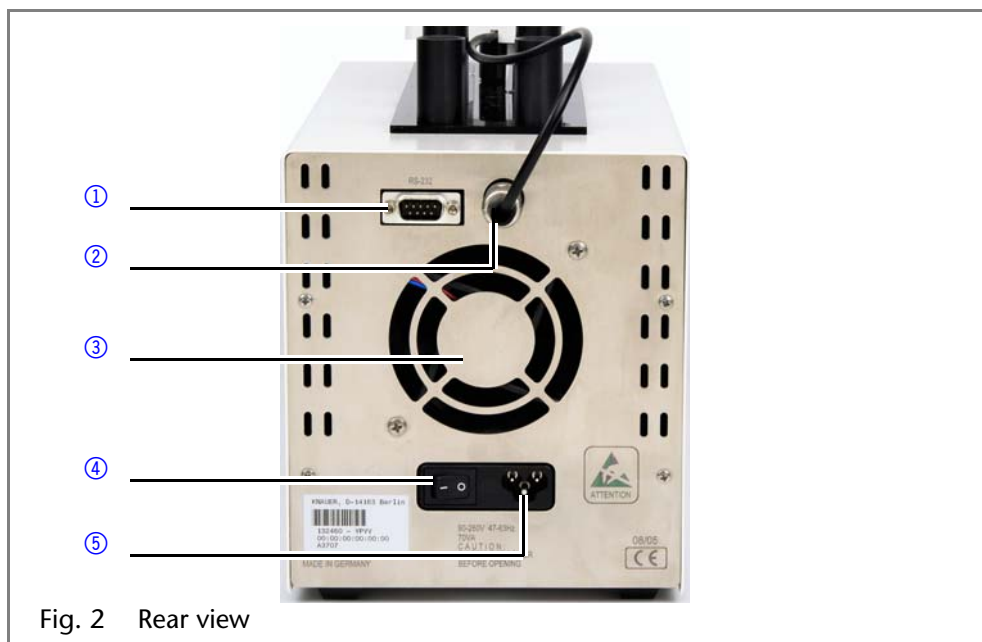


Fig. 2 Rear view

## Scope of Delivery

Please check whether the accessories are complete in accordance with the scope of delivery mentioned below. If a part is missing despite our comprehensive exit controls, please contact our customer support.

**Note:** Only use original parts and accessories made by KNAUER or a company authorized by KNAUER.

- Freezing point osmometer K-7400S
- Power cable
- User manual (German/English)
- Installation Qualification document ("IQ", English)
- RS-232 Connection cable
- Measuring head
- Pack of 1000 sample vials
- 400 mOsmol/kg calibration solution (10 vials)
- 850 mOsmol/kg calibration solution (12 vials)
- Pack of cotton swabs

## Safety for Users

*Professional Group* The user manual addresses persons who are qualified as chemical laboratory technicians or have completed comparable vocational training.

The following knowledge is required:

- Knowledge regarding the health risks of chemicals
- Participation during an installation of a device or a training by the company KNAUER or an authorized company.

*Safety Equipment* When working with the device, take measures according to lab regulations and wear protective clothing:

- Safety glasses with side protection
- Protective gloves
- Lab coat
- All safety instructions in the user manual

*What must be taken into account?*

- The environmental, installation, and connection specifications in the user manual
- National and international regulations pertaining to laboratory work
- Original spare parts and tools made or recommended by KNAUER
- Good Laboratory Practice (GLP)
- Accident prevention regulations published by the accident insurance companies for laboratory work
- Power cable: Defective power cables are not to be used to connect the device and the power supply system.
- Power strip: If several devices are connected to one power strip, always consider the maximum power consumption of each device.
- Power supply: Only connect devices to voltage sources, whose voltage equals the device's voltage.

*Where is use of the device prohibited?*

Never use the system in potentially explosive atmospheres without appropriate protective equipment. For further information, contact the Technical Support of KNAUER.

*Secure decommissioning*

At any time, take the device completely out of operation by either switching off the power switch or by pulling the power plug.

*Opening the Device*

The device may be opened by the KNAUER Technical Support or any company authorized by KNAUER only.

## Signal Words

Possible dangers related to the device are divided into personal and material damage in this user manual.



Lethal injuries will occur.



Serious or moderate injuries can occur.



Minor injuries can occur.



Device defects can occur.

## Decontamination

Contamination of devices with toxic, infectious or radioactive substances poses a hazard for all persons during operation, repair, sale, and disposal of a device.



### Life-threatening injuries

Health danger if getting in contact with toxic, infectious or radio-active substances.

→ Before disposing of the device or sending it away for repair, you are required to decontaminate the device in a technically correct manner.





All contaminated devices must be properly decontaminated by a specialist company or the operating company before they can be recommissioned, repaired, sold, or disposed of. All materials or fluids used for decontamination must be collected separately and disposed of properly.

*Decontamination Report*

Devices without a completed Decontamination Report will not be repaired. If you would like to return a device to KNAUER, make sure to enclose a completed **Decontamination Report** with the device: <http://www.knauer.net/en/downloads/service.html>

# Symbols and Signs

The following symbols and signs can be found on the device or in the user manual:

	Symbol	Meaning
<i>Warning Signs</i>		Electric shock hazard
		Electrostatic discharge hazard, damages to system, device, or components can occur.
		Biohazard
<i>CE Mark</i>		A device or system marked with CE fulfills the product specific requirements of European directives. This is confirmed in a Declaration of Conformity.

## Installation

This chapter describes all preparatory steps prior to start-up.

### Preparations

**Note:** To ensure structural stability, the device is not intended to be stacked on or below other devices.

### Work Location

The intended use be ensured only if the requirements for ambient conditions of the operating environment are met. You will find the ambient conditions in the chapter "Technical Data" (see page 28).

*General Purpose*

- Position the device on a level surface.
- Protect the device against direct exposure to sunlight.
- Set up the device at a location not exposed to air drafts such as air conditioning systems.
- Do not set up the device near to other machines that cause floor vibrations.
- Keep the the devices away from high frequency sources. High frequencies may compromise measurement values.

*Space requirements*

- At least 5 cm, if there is another device on one side.
- At least 10 cm, if there are devices set up on both sides.
- At least 15 cm to the cooler fan on the rear.

*Ambient temperature*

The device should not be exposed to major temperature fluctuations during measurement.



**NOTICE****Device defect**

The device overheats at exposure to sunlight and insufficient air circulation. Device failures are very likely.

- Set up the device in such a way that it is protected against exposure to direct sunlight.
- Keep at least 15 cm clear at the rear and 5–10 cm at each side for air circulation.

**Power Supply**

Use only the enclosed power cable to connect the device to the mains to make sure that the specifications stated in Technical Data are met. But check beforehand to use power cables admitted for use in your country. Replace defective power cables only with accessories from KNAUER. Detachable power cables must not be replaced with different cable types.

The maximum power input is 70 watts.

**NOTICE****Electronic defect**

Electronic hazard when using an identically constructed power adapter from another manufacturer.

- Contact the Technical Support of KNAUER.
- Do not connect the device to the power connection.

**Note:** The nominal capacity of the connected devices must be maximum 50 % of the power supply to account for larger inrush currents when switching on the modules.

*Prerequisites*

- The electrical power supply at the installation site must be connected directly to the nearest main power line.
- The power must be free from ripple, residual current, voltage peaks and electromagnetic interference.
- The connectors for the mains voltage are grounded accordingly.
- The device receives sufficient power with reserve capacity.

*Power plug*

- The device is intended for use with AC power networks of 100 – 240 V.
- Make sure that the power plug on the rear of the device is always accessible, so that the device can be disconnected from the power supply.
- To disconnect the device from the main power, disconnect the power plug.
- The osmometer needs a three-core power plug.

**⚠ DANGER****Electric shock**

Make sure that the power cable is not exposed to high stresses.

- The power cable should not be bent or clamped.
- Make sure that the power cable does not come in contact with liquids and heat sources.

**Unpacking the Device**

At the factory, all KNAUER devices are carefully packed for safe transport. After unpacking, check all device parts and accessories for signs of damage during transport and if necessary assert claims for damages immediately to the transport company.

The complete listing of consumable and spare parts can be found in section "Accessories and spare parts" on page 29.

*Prerequisite*

Check packaging for damage caused during transportation. If necessary, put forward any claim for damages to the carrier.

*Tools*

Utility knife

**CAUTION****Bruising danger**

Damage to the device by carrying or lifting it on protruding housing parts. The device may fall and thus cause injuries.

→ Lift the device only centrally on the side of the housing.

*Process*

1. Set up the package in such a way that you can read the label. Using the utility knife, cut the adhesive tape and open the packaging.
2. Remove the foam insert. Take out the accessory kit and the manual.
3. Open the accessory kit and check the scope of delivery. In case any parts are missing, contact the Technical Support.
4. Clasp the device from below, lift it out of the packaging and place it on its feet.
5. Check the device for signs of damage that occurred during transport. In case you notice any damage, contact the Technical Support.
6. Place the device in its site of operation and remove protective foil.

*Next steps*

Store packaging and keep the included packing list for repeat orders.

**Start-up**

**Note:** The device is not suitable for constant operation because it would reduce the lifespan of the Peltier elements.

**WARNING****Risk of Infection**

Risk of infection by injuries caused by a contaminated stirring wire.

→ Wear safety gloves when handling the measuring head.

**CAUTION****Skin Injury**

The cooling compartment reaches temperatures up to -30 °C during the running measurement. Contact with unprotected hands may lead to skin injuries.

→ Avoid direct contact with the cooling compartment.

→ For cleaning or drying, use cotton swabs or a lint-free tissue.

For information and safety instructions for power supply, see page 9.

**DANGER****Electric shock**

High voltages build up inside the device, the housing serves as a protective cover.

→ Never put the device into operation when the housing is open.

→ First close the housing, then plug in the power plug and switch the device on.

**NOTICE****Electronic defect**

Damage to the electronics if the device is turned on while connecting or interrupting the power connection.

→ Switch off the device beforehand.

*Prerequisites*

The device is switched off. The power cable is detached from the power supply.

*Procedure*

1. Attach the power cable to the power connector on the rear of the device.
2. Switch the device on by using the "ON/OFF" switch.

*Next Steps*

Bring the device into operation. To put the device out of operation, switch off the device and detach the power plug from the power supply.

# Operation

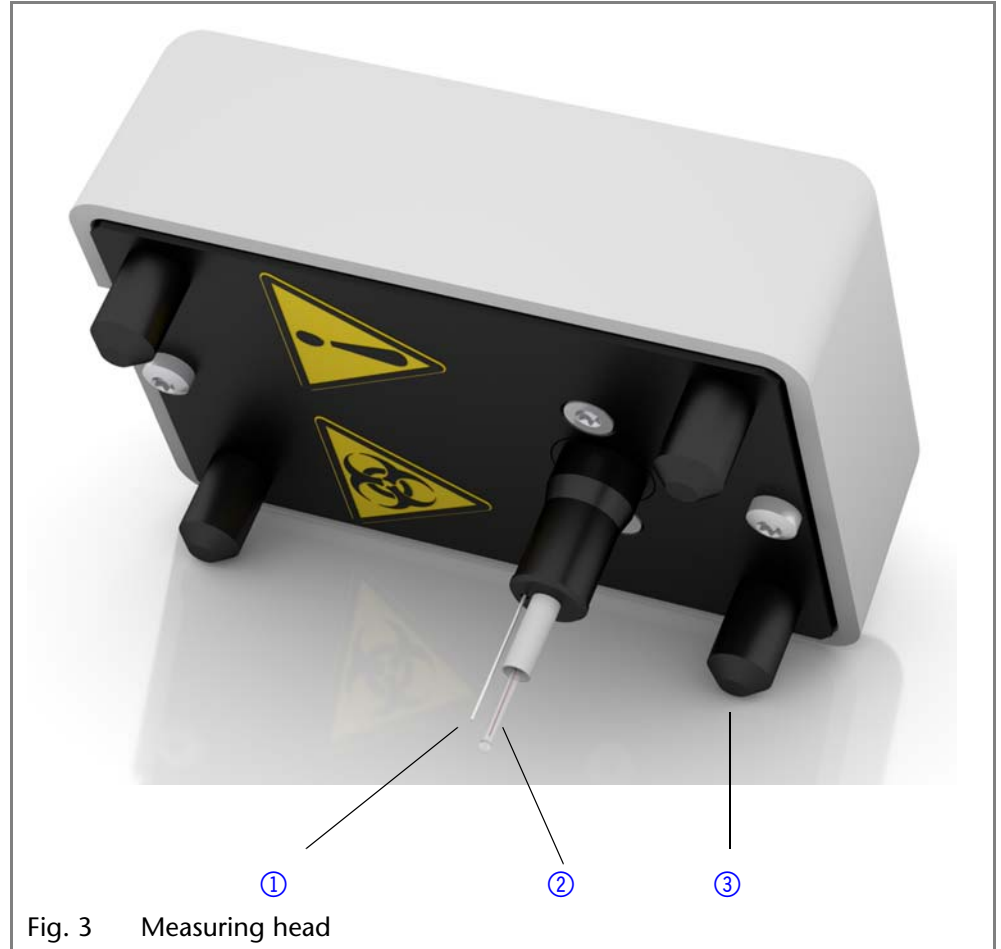
## Connecting the Measuring Head

Parts of the measuring head are an inbuilt motor with a stirring wire and the thermistor as temperature sensor. The measuring head cable is to be plugged into the connector on the back of the device.

The sample vial, filled with the exact volume of calibration or sample solution (50 – 150 µl) must be mounted onto the adapter.

### Legende

- ① Stirring wire
- ② Thermistor
- ③ Foot



**Note:** Any change of the measuring head causes the need for a precalibration of the device to adapt the device to the measuring head (see “Pre-calibration” on page 16).

The measuring head is placed on the housing plate with the sample vial rising into the cooling cavity. The device is now ready for measurement.

**Note:** The sample volume can be reduced down to a volume of 50  $\mu\text{l}$ . It is important that all calibrations must be performed with exactly the same volume as the later sample.

## Stand-by Position of Measuring Head

**Note:** Generally the measuring head should be stored with attached sample vial.

While working with the osmometer, use the four columns on top of the device for temporarily set down the measuring head without sample vial. In the measurement position, it is not necessary that the feet of the measuring head rest on the device.

To avoid sample spilling into the measuring head, it must be held upright.

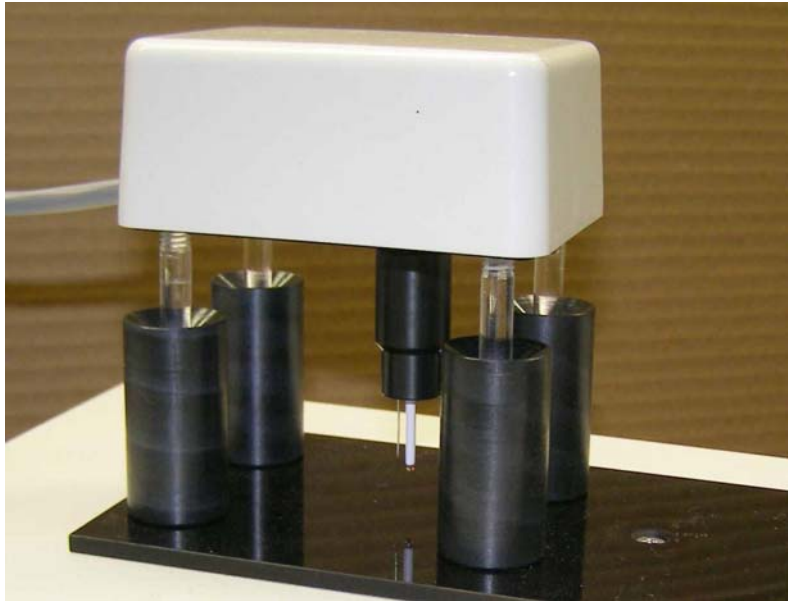


Fig. 5 Set-down of the measuring head without sample vial

## Measurement Principle and Process

The osmotic pressure in a solution is proportional to the freezing point depression of that solution. An aqueous solution, with an osmotic pressure corresponding to an ideal 1-molal solution, freezes at  $-1.858\text{ }^{\circ}\text{C}$ . An aqueous solution with this freezing point has a concentration of 1 Osmol/kg.

To perform a measurement, the sample solution is cooled down first without stirring. Even pure water can be super-cooled down to about  $-5$  through  $-8\text{ }^{\circ}\text{C}$  without freezing. The freezing is initiated by an automatically start of the stirring wire at a set temperature. The temperature reaches the freezing point,  $0\text{ }^{\circ}\text{C}$  in case of pure water.

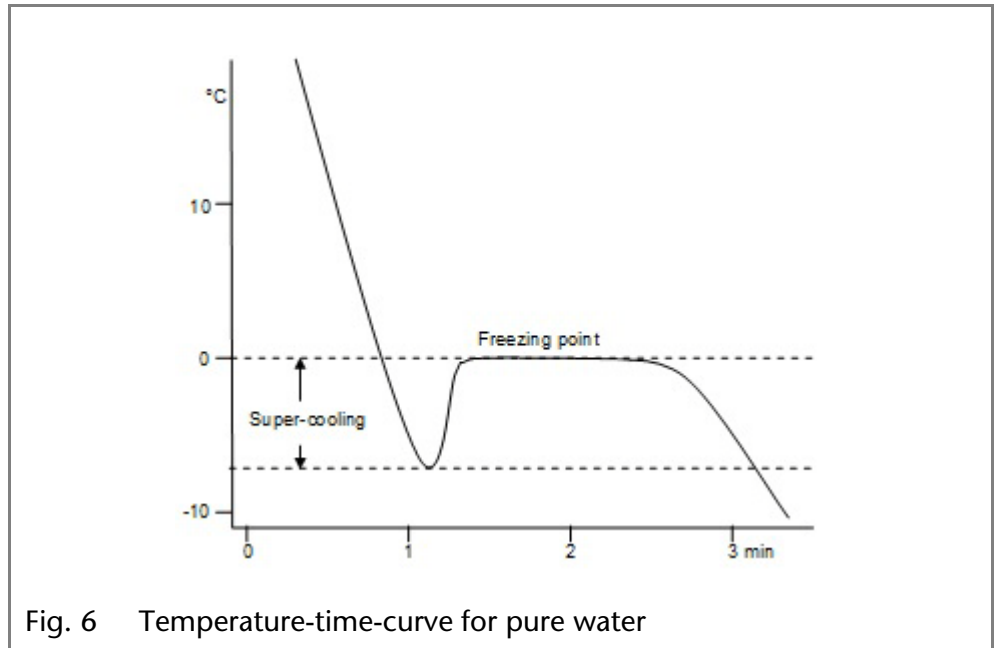


Fig. 6 Temperature-time-curve for pure water

The freezing point of a solution is below 0 °C. The freezing point depression  $\Delta T$  is a measure for the osmolality of a solution. It can be read directly from the display given in mOsmol/kg.

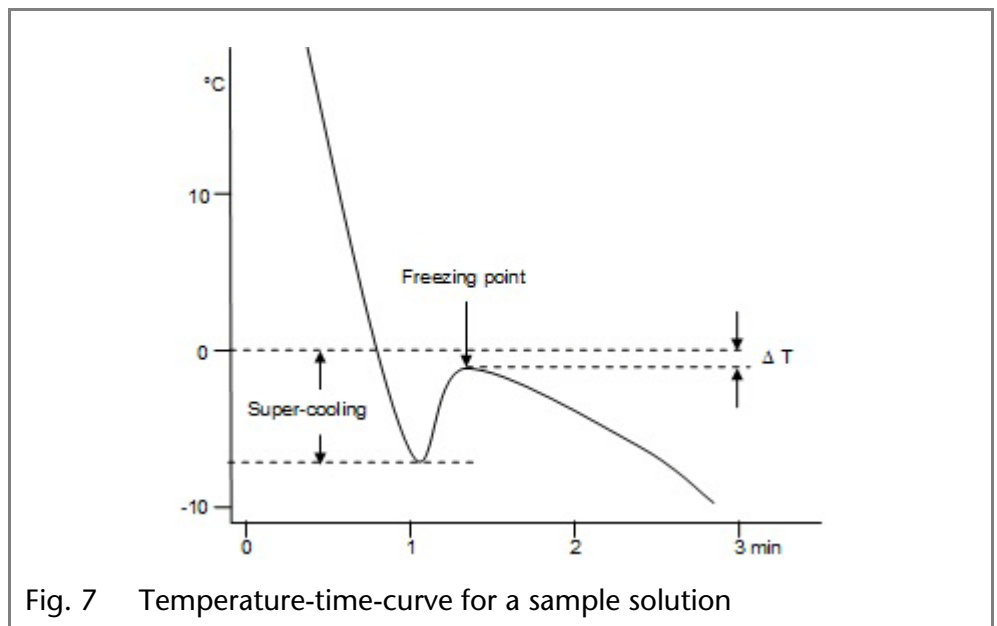


Fig. 7 Temperature-time-curve for a sample solution

During a measurement, the actual temperature is displayed. The device detects the maximum temperature reached, stores it, and if crystallization occurred satisfactory, displays the result.

After finishing the measurement, the sample vial is warmed up to 20°C (stand-by temperature of the device). The measuring head can remain there before and after measurement as long as desired.

## Switch-on

While switching on, the device performs an automatic self-test. During this time, the display presents the start screen with the current firmware version for a short time.

KNAUER	V01.XX
Freezing Point Osmometer	

After completion of the self-test, the display shows the measurement screen (see Fig. 8 screen 1). The cursor is placed on the rhombus field. As shown in Fig. 8, it will take you to the relevant menu points.

## Menu Guide

Starting with the cursor on the rhombus field, further menus become accessible in an endless loop by pushing the vertical arrow buttons ▲ ▼. By pushing the horizontal arrow buttons ► ◀, the cursor moves from field to field. Only those fields are accessible which are allowed to change the setting. Other fields, like the actual measured temperature, will be skipped.

On any changeable field the possible settings can be scrolled using the vertical arrow buttons ▲ ▼. The displayed values will be accepted and stored when leaving the field with one of the horizontal arrow buttons ► ◀. In this way, you can e. g. choose between printer and PC for data output (screen 2) or display the measurement data in mOsmol or °C (screen 3) or display the measurement data in mOsmol or °C (screen 1).

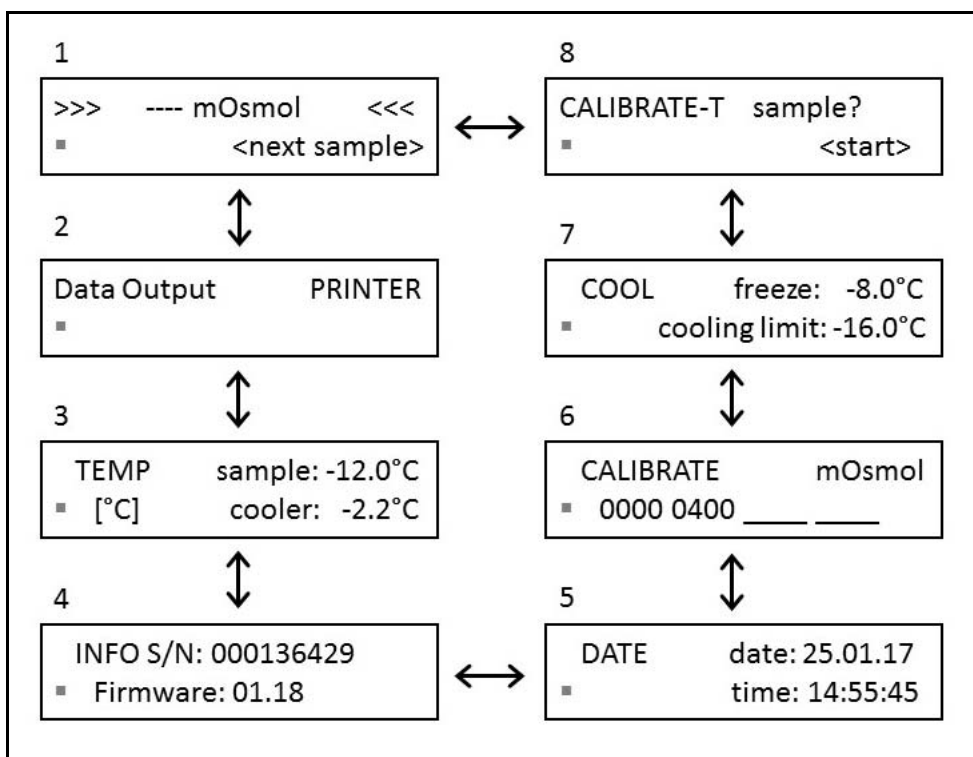


Fig. 8 Menu path

Screen	Explanation
<b>1</b>	Main menu
<b>2 Data Output</b>	Controls the connection of the Osmometer with a printer or PC.
<b>3 TEMP</b>	Displays the current temperatures of the sample and the cooling cavity. Additionally, units can be changed from [°C] to [°F] with the ▲ ▼ buttons. If the sample temperature is displayed with "----", the measuring head is not connected.
<b>4 INFO</b>	Shows the serial number and the current firmware version.
<b>5 DATE</b>	Setting for the date and time. The setting remains saved when switching off the device.

Screen	Explanation
<b>6 CALIBRATE</b>	Displays the calibration points which are currently in use. There are 3 possible calibration points. The first field is fixed at "0000". At least 2 calibration points are necessary for regular measurements. From this screen, the calibrations will be started (see "Performing the Calibration" on page 17).
<b>7 COOL</b>	Sets following parameters, in 0,5°C steps: <ul style="list-style-type: none"> <li>▪ Initiation point "freeze" within the range from -13,0 to -4,0°C.</li> <li>▪ Minimum temperature "cooling limit" within the range from -18.0 to -12.0 °C.</li> </ul>
<b>8 CALIBRATE-T</b>	Enables the pre-calibration to adapt the osmometer to the measuring head, see "Pre-calibration" on page 16.

**Note:** Calibration and measurement must be performed at the same settings (freeze & cooling limit). Additionally, an identical sample volume must be used for measuring and calibration to ensure exact and reproducible data. As a result, every change of the temperature settings or the sample volume demands a new calibration.

## Preparing a Measurement

**Note:** The device should be switched on at least 5 minutes before use. To check the stirring wire, press the <VIB> button.



### Stab Injury

Risk of injury from vibration wire during function test performance.  
 → Perform the function test with attached measuring vial.

**Note:** If you want to print out the measurement results, you may connect a printer via RS-232 connector.

The result of each measurement will be printed automatically in the following format:

Measured 0400 mOsmol  
23.03.15 08:24:35

After a calibration run, you get the following print:

Calibrated 1234 mOsmol  
23.03.15 07:36:17

If a measurement is cancelled due to an error, the screen displays the corresponding error message. If a printer is connected, it prints the error message, without the specification about the type of error.

(NO) FREEZE  
23.03.15 09:06:04

## Measurement Preparation Process

*Prerequisites*

The device is switched on and calibrated.

*Process*

1. Pipet 50 – 150 µl sample or calibration solution into a clean, dry sample vial.

**Note:** The sample volume can be freely chosen in the mentioned range. It is important that you use exactly the same amount for calibration and measurement.

**Practical Tip:** Please consider, the higher the volume, the better the precision of the measurement.

2. Put the filled sample vial on the thermistor, so that the thermistor and the stirring wire dip in the solution. The meniscus of the liquid must be aligned horizontally.
3. Place the measuring head on top of the device, so that the sample vial is inserted into the cooling cavity. The feet of the measuring head do not have to rest on the device.

*Next Steps* The device is now ready for measurement.

## Calibration

### Pre-calibration

A pre-calibration is necessary when:

- the device is taken into operation the first time.
- the measuring head has been replaced.

*Prerequisites* Prepare a measurement with water (HPLC quality).

*Process*

1. Select CALIBRATE-T in the osmometer menu (screen 8 in Fig. 8).
2. Push the <START> button. While the pre-calibration is running, the screen displays the sample temperature and the elapsed time. The run can be interrupted at any time by pressing the <STOP> button.

thermistor	> +9.99°C<
00:56	

3. After finishing the calibration, the screen displays the message "successful". The previous calibration values will be deleted.

### NOTICE

#### Device Defect

If the sample vial is removed from the measuring head while the sample or the calibration solution is frozen, the thermistor or the stirring wire can be damaged.

→ When the message "next sample" appears on the display, the sample vial is safe to remove.

**Note:** Any performed pre-calibration deletes the existing calibration values.

4. If you get the message "not successful" on the screen at the end of the pre-calibration, the sample is not frozen. Make sure if
  - the sample is 0 mOsmol/kg
  - a valid measuring head is proper connected to the device.
5. Repeat the pre-calibration if necessary.

### Measurement Calibration

Normally a Two-Point Calibration is sufficient.

**Note:** Calibrations must be performed very carefully. A confusion of the calibration solutions will affect all later measurements and leads to incorrect results.

Up to 3 calibration points are possible. The desired calibration concentration can be selected on any of the fields. After that, the calibration run is started by pressing the <START> button.

The first field is fixed on "0000". On the other fields the choices are ----, %%%%, 50, 100, 150, up to 1900, 1950, 2000 in steps of 50 mOsmol/kg. Selecting "----", the corresponding calibration point will be deactivated.



The selection of "%%%%" deactivates all other calibration points (except "0000"). Simultaneously the displayed measurement results will be shown in % of this calibration point instead of "mOsmol/kg". The measurement results are now displayed in % of this calibration value.

**Note:** At least two calibration points must be active, the "0000" and one of the others.

The calibration point remains active until a new valid value for the selected field is created on the CALIBRATE screen. If a calibration run is not successfully finished the last calibration value becomes reactivated.

At least 2 calibration points are necessary for regular measurements. To determine the measurement results, it will be interpolated between the closest calibration points, or extrapolated from there.

**Note:** It is recommended to choose the calibration solutions so that the expected measurement values are situated between the calibration points.

### Performing the Calibration

*Prerequisites* Prepare a measurement with water (HPLC quality, see "Measurement Preparation Process" on page 15).

- Process*
1. Activate the CALIBRATE screen and set the first calibration field to "0000".
  2. Push the <START> button. The run can be interrupted at any time by pressing the <STOP> button. While the calibration is running, the screen displays the sample temperature and the elapsed time.

0000 mOsmol > +5.43 °C  
 1:23

3. After the measurement is successfully completed, the measurement value is displayed.

0000 mOsmol > -0.00 °C  
 o.k.? <start>

Push the <START> button for confirmation. An earlier measured calibration will be overwritten. By pushing any other button, the new value will be abandoned and the CALIBRATE menu will be shown again.

4. Prepare a measurement with a calibration solution (see "Measurement Preparation Process" on page 15). Enclosed to the osmometer are fitting solutions for calibrations with an osmolality of 400 or 850 mOsmol/kg.
5. Activate the second calibration field on the CALIBRATE screen and set the osmolality for the calibration solution.
6. Repeat step 2 to 3.

For special applications, especially in the pharmaceutical industry, a Three-Point Calibration is required. If this is the case, proceed according to the process above and repeat the process for the calibration field 3.

### Re-calibration

A re-calibration is necessary after

- replacing the measuring head
- changing the temperature settings (see fig. 8 screen 7)
- changing the sample volume

# Measuring Sample Solutions

## Performing a Sample Measurement

*Prerequisites* Prepare a measurement with a sample solution (see "Measurement Preparation Process" on page 15).

*Process* 1. To perform measurements, choose screen 1 (Fig. 8) with the ▲ ▼ buttons and push the <START> button. While the measurement is running, the screen displays the sample temperature and the elapsed time.

```

RUN                > -1.23°C
                1:23
  
```

The run can be interrupted any time by pressing the <STOP> button.

2. The device starts the crystallisation automatically and detects the freezing point depression. If the crystallisation occurred properly, it displays the corresponding osmolality on the display:

```

>>> 1234 mOsmol <<<
      ■          <next sample>
  
```

The printer output is given in the following format:

```

Measured 1234 mOsmol
19.03.15 16:05:35
  
```

3. Before the next measurement, wait for the sample to unfreeze. This is shown by the message "next sample" on the display.

If no crystallisation occurs after the automatic start of the stirring wire, the measurement will be stopped with the following error message:

```

> ERROR <
NO FREEZE
  
```

If the crystallisation occurs before the start of the stirring wire, the measurement will be stopped with the following error message:

```

> ERROR <
FREEZE
  
```

### NOTICE

#### Device Defect

If the sample vial is removed from the measuring head while the sample or the calibration solution is frozen, the thermistor or the stirring wire can be damaged.

→ When the message "next sample" appears on the display, the sample vial is safe to remove.

**Note:** The sample melting can be quickened by warming up with the fingers. When "next sample" appears below the measurement value, the sample is unfrozen, and it is safe to remove the vial.

## Important Operating Notes

1. The device is only suitable for aqueous solutions.
2. Only use sample vials which were obtained from KNAUER or recommended by KNAUER. Sample vials should not be reused.
3. To ensure a fixed sample volume, always introduce the solutions with a clean and dry pipette tip into the sample vial. Make sure that there are no air bubbles in the liquid or drops on the vial wall above the liquid. The sample solution should stand upright in the sample vial.

4. Handle the thermistor always very carefully. Do not use abrasive materials for cleaning. The thermistor should be cleaned occasionally with a soft tensidic cleaning agent (dilution: 1:10 with water (HPLC quality) about  $40\pm 5$  °C). After immersing, activate the stirring function 3 – 5 times for 5 seconds and rinse with water (HPLC quality). The cleaning frequency depends on the used sample matrix. It is sufficient for pure saline solutions to rinse the thermistor with the following sample solution. A dirty thermistor does not give accurate measurement results.
5. Cleaning and drying the thermistor with alcohol or acetone is not recommended. High-frequency sources may compromise measurement values. Remaining solvent vapours could cause false results.
6. Solutions containing proteins, such as sera, can only be measured once. Freezing causes denaturation of the protein. A repetition of the measurement with the same sample would result in increased values.
7. Please note the difference between ideal and real osmolality. Only real osmolality can be measured. It is not possible to prepare other calibration solutions by dilution of a calibration solution since the activity coefficient of the solution changes with dilution.
8. Water which is stored in polyethylene bottles over a longer period could be difficult to be super-cooled. If such difficulties may occur, we recommend a storage in glass bottles.
9. Make sure that the sample material is homogeneous. If the measurement result has to be reproduced with the same sample, it has to be mixed with the stirring wire before each further measurement. As ice does not contain salines and floats on top while unfreezing, it results in a concentration gradient which has to be removed before measurement.
10. Evaporation can increase the concentration of dissolved particles which leads to increase of osmolality of a solution. Calibration solutions should be used not longer than 30 - 60 min after the vials were opened. We recommend using KNAUER calibration solutions which are mentioned in chapter "Reorders" (see page 29).
11. After finishing all measurements, the osmometer should be switched off, because a permanent operation reduces the lifespan of the device. If the device is switched on but unused for a longer period, it may occur that some water from room air is condensed in the cooling cavity. It should be removed carefully with a dry, lint-free tissue prior to the next measurement.
12. The stirring wire triggers the reproducible freezing and is set optimal in the factory. Make sure that you do not bend the wire while attaching the sample vials and cleaning.

## Functionality Tests

### *Installation Qualification* (IQ)

The customer may request the Installation Qualification, which is free of charge. In case of a request, the Technical Support of KNAUER or from a provider authorized by KNAUER performs this functionality test during the installation.

The Installation Qualification is a standardized document that comes as part of the delivery and includes the following:

- confirmation of flawless condition at delivery
- check if the delivery is complete
- certification on the functionality of the device

### *Operation Qualification* (OQ)

The Operation Qualification includes an extensive functionality test according to KNAUER standard OQ documents. The Operation Qualification is a standardized document and free of charge. It is not part of the delivery, please contact the Technical Support in case of request.

The Operation Qualification includes the following:

- definition of customer requirements and acceptance terms
- documentation on device specifications
- device functionality check at installation site

*Test Intervals* To make sure that the device operates within the specified range, you should test the device regularly. The test intervals are dependent on the usage of the device.

*Execution* The test can be carried out either by the Technical Support of KNAUER or from a provider authorized by KNAUER (for a fee).

# Troubleshooting

## System Messages

Any error message will be deleted simply by pressing any button. The main screen will be displayed.

In any case the first row shows > ERROR < which is specified in the second row.

Following messages can appear:

Error message	Cause	Remedy
FREEZE	The crystallization took place or an error occurred before the active triggering of the freezing process.	see "Self-Freezing of the Sample" on page 20
NO FREEZE	No sample crystallization took place after the activation of the stirring wire.	see "Non-Freezing of the Sample" on page 21
OUT OF RANGE	The measured value is outside the acceptable range.	Usually a malfunction of the thermistor. Contact the Technical Support.
NOT CALIBRATED	No calibration points are available.	See section "Measurement Calibration" on page 16
COOLER TIMEOUT	Timeout due to too slow cooling: >5 minutes from START	Contact the Technical Support.
MEASUREMENT TIME-OUT	Timeout due to too long measurement: >5 minutes from START	Contact the Technical Support.

## Self-Freezing of the Sample

If the sample does not super-cool but freezes spontaneously without initiation, there are three possible causes:

If the following solutions do not solve the problem, the super-cooling temperatures are too low. The freezing temperatures in the device must be set higher. The cooling temperature can be changed in screen 7 COOL (Fig. 8 "Menu path" on page 14).

Cause	Remedy
Too little sample solution is in the sample vial. A very cold zone, which causes self-freezing of the sample, develops above the solution on the sample vial wall.	Use exact the required sample volume: 50 – 150 µl

Cause	Remedy
The sample vial or the thermistor is scratched or contaminated. Both could act as crystallization nuclei causing premature freezing.	<ul style="list-style-type: none"> <li>▪ Use a new sample vial and carefully clean the thermistor.</li> <li>▪ Only use sample vials which were obtained from KNAUER or recommended by KNAUER.</li> </ul>
Solutions saturated with air or gases are sometimes difficult to super-cool.	Degas the sample solution by ultrasound.

**Note:** Calibration and measurement have to be performed with the same temperature settings and equal volume.

## Non-Freezing of the Sample

If the sample does not freeze, one of the following causes may be responsible:

Cause	Remedy
The cooling rate is not sufficient.	Ensure that the air inlets and outlets at the device are free.
Too much sample solution is in the sample vial. This leads to an uncooled zone in the upper part of the solution. The stirring causes a mixing of warm and cold sample layer.	Use exact the required sample volume: 50 – 150 µl

If these solutions are not effective, the freezing temperature is too high and has to be lowered. The freezing temperature can be changed in screen 7 COOL (Fig. 8 "Menu path" on page 14).

**Note:** Calibration and measurement have to be performed with the same temperature settings and equal volume.

# Using the EuroOsmo® 7400 Software

This chapter will explain the basic knowledge needed for working with K-7400S and the EuroOsmo® 7400 software.

**Note:** The License Agreement should be read before using the EuroOsmo®7400 software (see page 30). Using the software, the user indicates that he has read the License Agreement and agrees with the regulations.

## Installation

The hardware requirements (processor, working memory) are very low for the installation of EuroOsmo® 7400.

The program runs under Windows 7, 8 and 10, but also with older operating systems, back to Windows XP.

- Process*
1. Insert the EuroOsmo CD into the CD ROM drive.
  2. Wait for 30 seconds. The set-up starts automatically. You will be guided through the installation via self-explanatory windows.
  3. If your CD ROM drive does not have AutoPlay:
    - Double-click on <My Computer>
    - Right-mouse click on your CD ROM drive.
    - Select <AutoPlay> from the right-click menu.

- If there is no AutoPlay menu, select Explore from the right-click menu, go to EuroOsmo® 7400/Disk1 and double-click on <Setup.exe>.

## Hardware Installation

The device has to be connected with a defined port of the computer (e.g. COM 1) prior to starting the program.

## Main Screen

After starting the program you will see the main screen of the software.

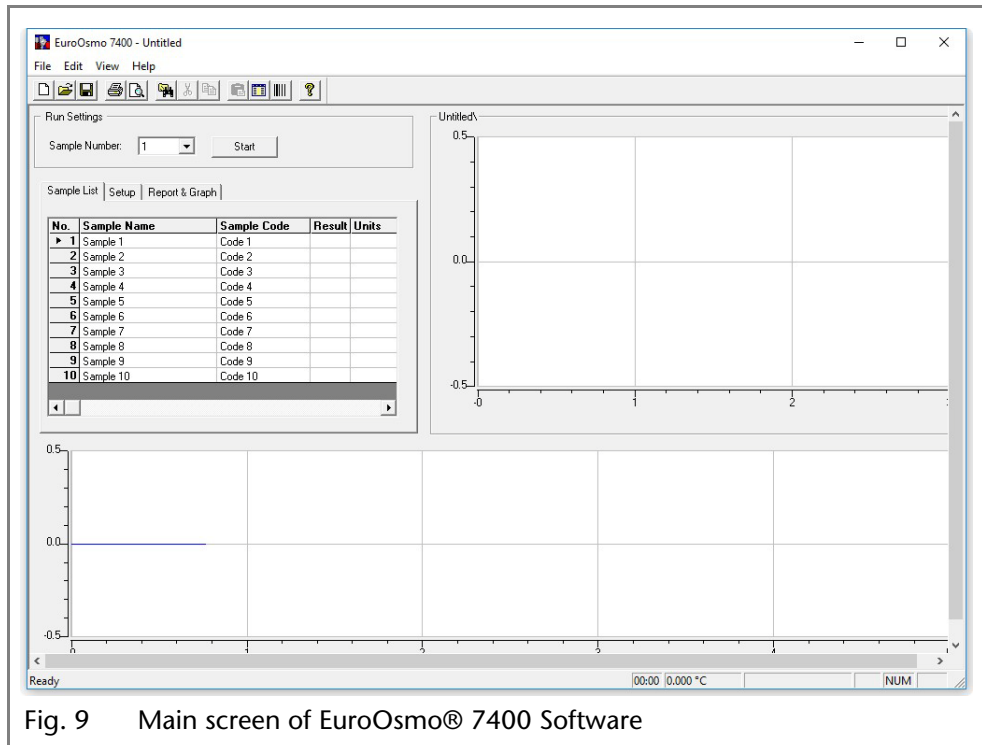


Fig. 9 Main screen of EuroOsmo® 7400 Software

If the osmometer is not connected or switched off, you will get the following message.

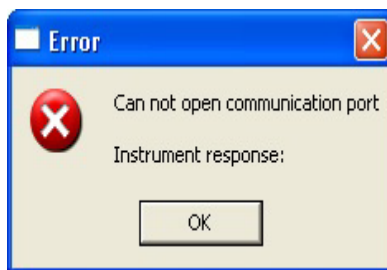







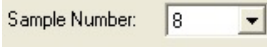
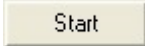
Fig. 10 Error message for an unconnected osmometer

## Icons

Command	Explanation
	Prints the result list.
	Displays a preview of the actual report.
	Starts the search function.

Command	Explanation
	Pastes the data of a selected field into following lines of the chosen column.
	After each bar code scan, the cursor switches to the next sample line.

## Run Settings

Command	Explanation
	Select the number of the sample to be measured from the sample list.
	Start button for any measurement or calibration run. While starting the run it changes to a stop button to enable a manually aborting of the run.

If the sample has already been measured, the following warning will be displayed.

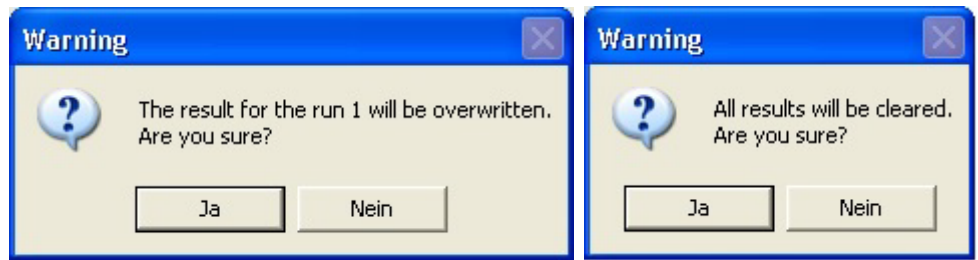


Fig. 11 Warning prior to overwriting existing results

## Sample List

This list is used to name the samples and codes. The measurement results will be filled automatically and can be saved. Using the scroll bar at the bottom, a <Comments> column becomes accessible for detailed information. The samples will be measured in the order of the list and the results are filled in and saved.

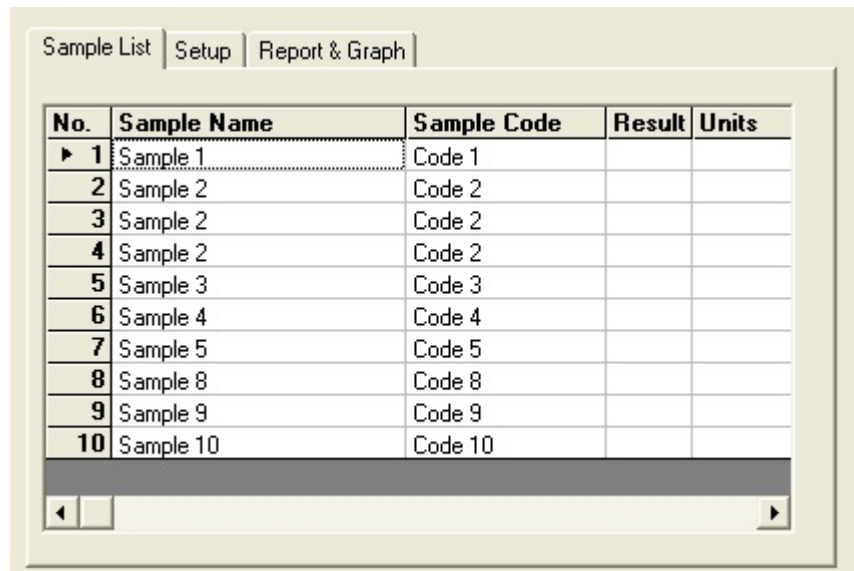


Fig. 12 New sample list

Double clicking on <No.> of an already measured sample displays the corresponding temperature sequence. The path of the already stored sample list is shown on top of the graph.

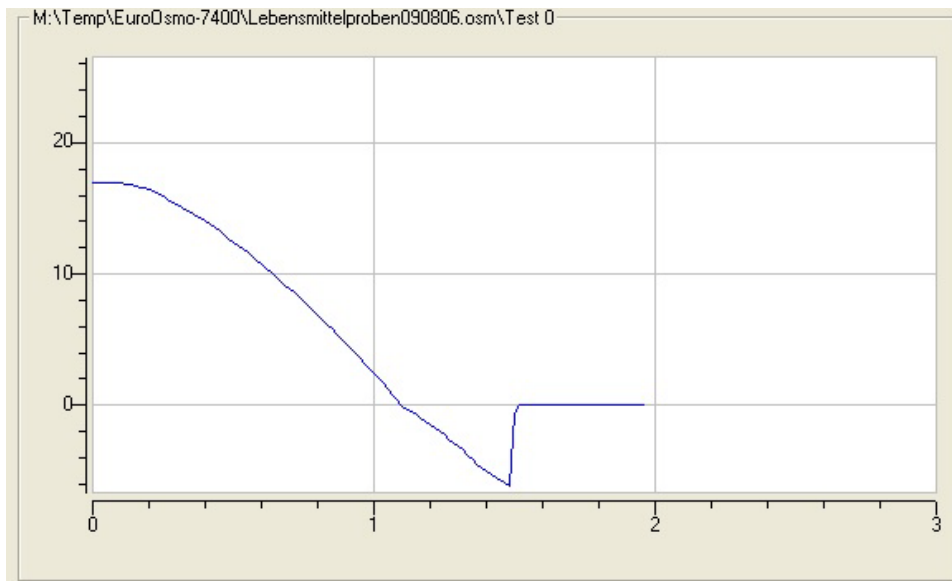


Fig. 13 Measurement window with the temperature sequence of a run

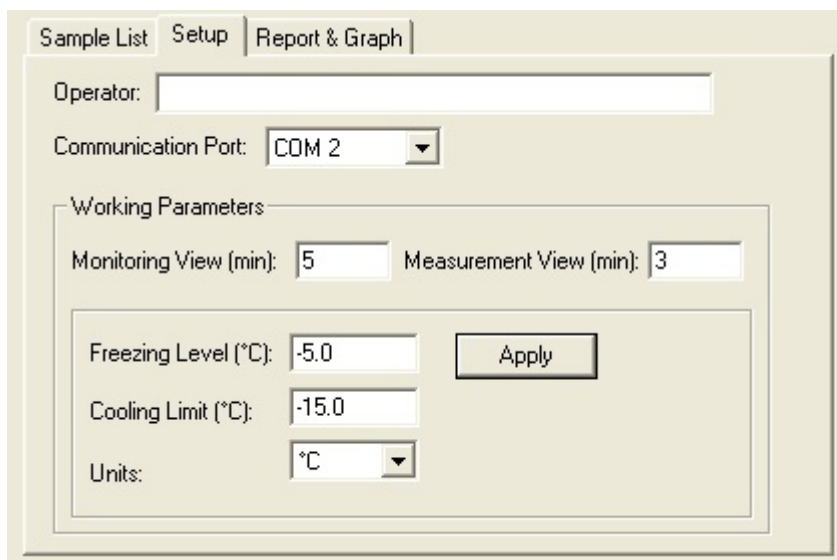


Fig. 14 Setup area of the main window

## Setup

Area for editing general settings for measurement sequences and corresponding reports.

Command	Explanation
Operator	Enter the name of the user.
Communication Port	Select the COM port to which your osmometer is connected.



## Working Parameters


Command	Explanation
Monitoring view	The entered value defines the scale of the monitor by its final value in minutes. The temperature pattern is shown continuously for the defined period.
Measurement view	The entered value defines the scale of the measurement by its final value in minutes.
Freezing Level	The initiation point FREEZE can be set within the range from -13.0 to -4.0 °C. Any decimal value will be rounded in steps of 0.5°C.
Cooling Limit	The minimum temperature can be set within the range from -18.0 to -12.0 °C. Any decimal value will be rounded in steps of 0.5°C.
Units	Select the units valid for the measurement sequence. The choice is mOsmol, °C, °F, and %. If °F is selected, the values of the freezing level and cooling limit are automatically changed to °F.
	By clicking on the <Apply> button, the entered values are sent to the osmometer. If any value is out of the allowed range a corresponding message will be displayed. The setting of the osmometer remains unchanged.



Fig. 15 Error message in case of not valid parameters

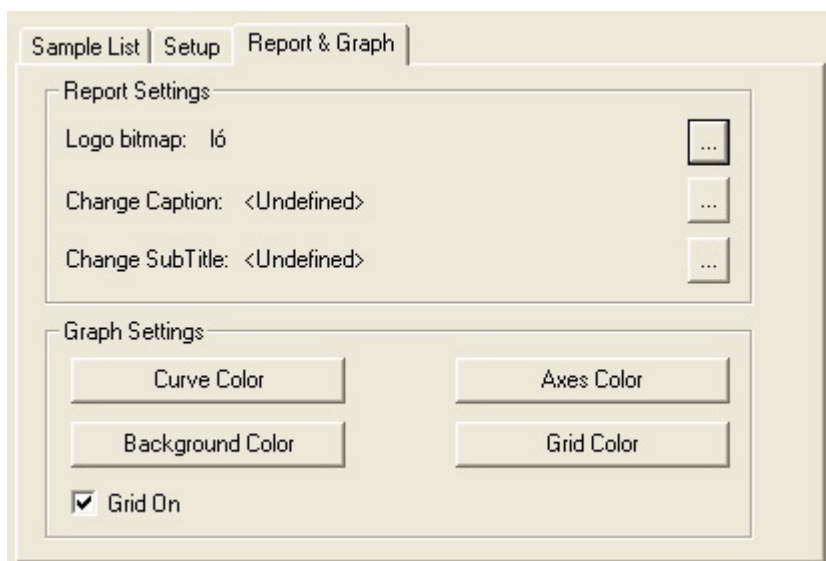


Fig. 16 Report design area

## Report & Graph

Command	Explanation
Report Settings	Report design area.
Logo Bitmap	Clicking on the <...> button, an explorer window appears to select the logo.
Change Caption	Clicking on the <...> button, an explorer window appears to edit the caption. Clicking on the <Font...> button to format the caption.
Change SubTitle	Clicking on the <...> button, an explorer window appears to edit the subtitles. Additionally, you can format the subtitles here.
Graph Settings	Area for settings of the graph design. A click on one of the four buttons will open the colour selection window where the colours of the curve, axes, grid lines, and the back ground can be defined separately.
GridOn	This option field is to show or to hide the grid lines of the graphs.

A click on the <print> button opens a printer setup window prior to printing the report. A preview of the actual report will be displayed when clicking on the <preview> button.

## Search Function

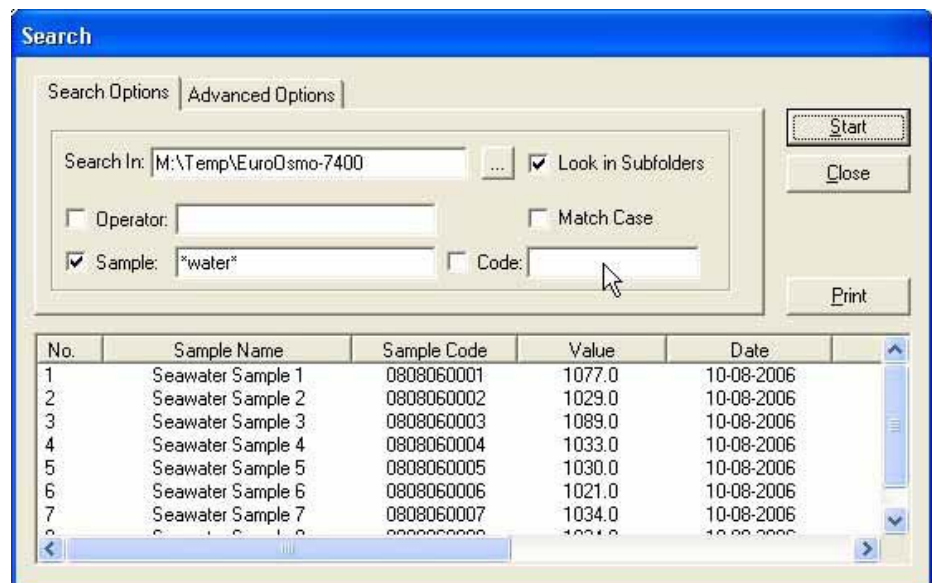


Fig. 17 Search function

Clicking on the <...> button, an explorer window appears to enter the search path. Additionally you have the option to search also in subfolders. Search subjects are the operator, the sample name, and/or the sample code. As wildcard characters can be used ? for single characters and \* for undefined areas before and/or after to the defined search characters.

If necessary the search results can be reduced by advanced options. These are the measurement values and/or the date of the measurement.

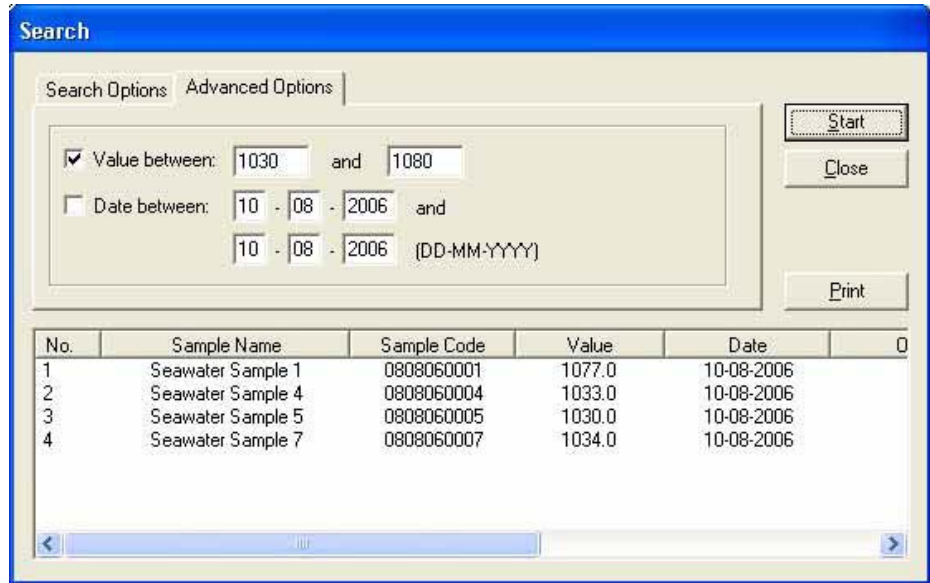


Fig. 18 Search function window, advanced options

## Maintenance and Care

### Cleaning the Stirring Wire and the Thermistor

**Note:** Stirring wire and thermistor should not be touched.



#### Risk of Infection

Risk of infection by injuries caused by a contaminated stirring wire.

→ Wear safety gloves when handling the measuring head.



#### Stab Injury

Risk of injury from stirring wire when detaching it by hand.

→ Detach the stirring wire only with attached measuring vial.

The measuring head parts which are wetted from the sample can be cleaned easily by the user by attaching a sample vial with a suitable cleaning solution to the measuring head (see "Important Operating Notes" on page 18).

**Note:** The supply and the appropriate use of these cleaning solutions is in the responsibility of the user.

### Function Checks of Cooling Capacity

The cooling capacity can be checked by measuring the temperature in the cooling cavity (-13 to -16 °C during cooling down and ~ 20 °C in standby position).

#### Prerequisites

The device must be calibrated.

#### Process

1. Connect the cable of the measuring head with the device and place the head on top of it (see page 12).
2. Press the <START/STOP> button to start the measurement and wait 5 minutes to get the error message <MEASUREMENT TIMEOUT> on the display.
3. Press quickly twice the ▼ button to read out the cooling cavity value on the COOL menu (see page 14).
4. The value must be in the range of -13 to -16 °C.
5. If the value is not reached, please check that the cooling limit is set to -16 °C.

6. Retry the test and press quickly twice the  $\nabla$  button after 5 minutes without displaying the error message.
7. If you change the cooling limit, you must perform a new calibration after this test.

### Reference Thermometer

Additionally, you can check the device with a calibrated reference thermometer. This thermometer should display temperature changes quickly.

- Process*
1. Fill the cooling cavity close to the top with water (HPLC quality) and insert the thermometer.
  2. Perform the test as described above and observe the temperature progression.
  3. After the test, it is necessary to dry completely the cooling cavity with a tissue.

The cooling capacity is usually sufficient for a sample volume of 150  $\mu$ l in the sample vial if the freezing turbulence in the solution is initialized by a start of the stirring wire within 150 seconds.

If the temperature of -13  $^{\circ}$ C cannot be obtained, the device must be returned for repair.

The thermistor should always be placed in the sample centre 0.5 to 1 mm above the bottom of the sample vial.

**Note:** The shape of the stirring wire is essential for a precise and reproducible measurement. Therefore, take care to not bend it. The lower end of the wire should contact the vial wall.

## Technical Data

### Main Features

Sample volume	50 – 150 $\mu$ l
Solvent	Water (HPLC quality)
Measurement time	ca. 2 min
Measurement range	0 – 2000 mOsmol/kg
Precision	SD $\leq$ 4 mOsmol/kg [0 – 400 mOsmol/kg] RSD $\leq$ 1.0 % [400 – 2000 mOsmol/kg]
Linearity	$\pm$ 1 % [0 – 1500 mOsmol/kg] $\pm$ 1.5 % [0 – 2000 mOsmol/kg]
Calibration	Two-Point Calibration (0 and one free selectable osmolality) optional: Three-Point Calibration (0 and 2 free selectable osmolalities)
Measuring procedure	Completely automatic after sample insertion

### Communication

Port	RS-232
Control	Keypad (LED screen, 2 rows with 24 characters), software

### General

Power supply	100 – 240 V, 50 – 60 Hz, 70 W
Dimensions	160 x 182 x 340 mm

Weight	4.5 kg (device + measuring head)
Ambient conditions	Temperature range: 10 – 35 °C Rel. humidity: 20 – 80 % (non-condensing)

## Repeat Orders

This list for repeat orders is valid for the time the document has been published. Deviations afterwards are possible.

**Note:** For repeat orders of spare parts use the enclosed packing list. Contact the Technical Support in case there are any questions on spare parts or accessories.

### Further Information

Further information on spare parts and accessories can be found online: [www.knauer.net](http://www.knauer.net)

Name	Order number
Freezing point osmometer K-7400S	A0006AC
Power cable 230V	M1479
RS-232 Connection cable	M1702
Plain printer	A3711
User manual	V3700
Measuring head for plastic sample vials	A0840-2
Pack of 500 plastic sample vials	A0272
Pack of 1000 plastic sample vials	A0720
300 mOsmol/kg calibration solution (12 vials)	A01240
400 mOsmol/kg calibration solution (10 vials)	A01241
850 mOsmol/kg calibration solution (12 vials)	A01250
2000 mOsmol/kg calibration solution (12 vials)	A01248
Pack of cotton swabs	A0469
Paper roll for plain printer	V7013

## Legal Information

### Transport Damage

The packaging of our devices provides the best possible protection against transport damage. Check the devices for signs of transport damage. In case you notice damages, contact the Technical Support and the forwarder company within three workdays.

### Warranty Conditions

The factory warranty for the device is stipulated by contract. During the warranty period, any components with material or design-related defects will be replaced or repaired by the manufacturer free of charge. Please connect to our website for further information on terms and conditions.

All warranty claims shall expire in the event that any unauthorized changes are made to the device. This warranty also excludes the following:

- accidental or willful damage
- damage or errors caused by third parties that are not contractually related to the manufacturer at the time the damage occurs
- wear parts, fuses, glass parts, columns, light sources, cuvettes and other optical components
- damage caused by negligence or improper operation of the device and damage caused by clogged capillary
- packaging and transport damage

In the event of device malfunctions, directly contact the manufacturer.

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Internet: [www.knauer.net](http://www.knauer.net)

## Declaration of Conformity

The Declaration of Conformity is part of the delivery and accompanies the product as a separate document.

## Disposal

Hand in old devices or disassembled old components at a certified waste facility, where they will be disposed of properly.

*AVV Marking in Germany*

According to the German "Abfallverzeichnisverordnung" (AVV) (January, 2001), old devices manufactured by KNAUER are marked as waste electrical and electronic equipment: 160214.

*WEEE Registration*

KNAUER as a company is registered by the WEEE number DE 34642789 in the German "Elektroaltgeräteregister" (EAR). The number belongs to category 8 and 9, which, among others, comprise laboratory equipment.

All distributors and importers are responsible for the disposal of old devices, as defined by the WEEE directive. End-users can send their old devices manufactured by KNAUER back to the distributor, the importer, or the company free of charge, but would be charged for the disposal.

## KNAUER Software License Agreement

between

KNAUER Wissenschaftliche Geräte GmbH,

Hegauer Weg 38, D-14163 Berlin

(hereinafter Licensor or KNAUER)

and the software user (hereinafter Licensee)

for the software package

EuroOsmo®7400 V1.5 and handbooks.

In return for acquiring a license to use the software ("EuroOsmo®7400 V1.5") and related documentation, Licensee agrees to the following terms and conditions:

### 1. License.

This agreement grants the Licensee a license to

(a) use the EuroOsmo®7400 V1.5 on a single computer system or, in the case of a multi-user or networked system which permits access to the EuroOsmo®7400 V1.5 by more than one user at the same time, at one (1) single working location, and

(b)make one copy of the EuroOsmo®2000 V1.2 in machine readable form solely for back-up purposes provided your reproduce KNAUER's copyright proprietary legends and mark the back-up copy with the express wording "back-up copy Knauer GmbH, Hegauer Weg 38, D-14163 Berlin".

## **2. Restrictions.**

Licensee may not distribute copies of the EuroOsmo®7400 V1.5 to others or electronically transfer the EuroOsmo®7400 V1.5 from one computer to another over a network. Licensee may not use the EuroOsmo®7400 V1.5 from multiple locations of a multi-user or networked system at any time. The EuroOsmo®7400 V1.5 contains trade secrets and in order to protect them the Licensee may not decompile, reverse engineer, disassemble, or otherwise reduce the Software to a human-perceivable form: LICENSEE MAY NOT MODIFY, ADAPT, TRANSLATE, RENT, LEASE, LOAN, RESELL FOR PROFIT, DISTRIBUTE, NETWORK OR ANY DERIVATIVE WORKS BASED UPON THE SOFTWARE OR ANY PART THEREOF. The Licensee must not remove from the program firm names, trademarks, logos, copyright notices and other markings or devices, which serve the protection of the program.

## **3. Ownership of EuroOsmo®7400 V1.5.**

Licensee owns the media upon which the EuroOsmo®7400 V1.5 is recorded or fixed, but KNAUER, Licensor, retains title and ownership of the EuroOsmo®7400 V1.5 recorded on the original media and all subsequent copies of the EuroOsmo®7400 V1.5, regardless of the form or media in which or on which the original and other copies may exist. This license is not a sale of the EuroOsmo®7400 V1.5 or any copy.

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## **5. Term.**

This license is effective until January 1, 2030, unless terminated earlier. Licensee may terminate the license at any time by destroying the Software (including the related documentation) together with all copies or modifications in any form. KNAUER will have the right to terminate the Licensee's license immediately if the Licensee fails to comply with all copies or modifications in any form. The termination does not raise Licensee's claim for refunding of the license fee.

## **6. Limited Warranty.**

KNAUER warrants only that the media upon which the EuroOsmo®7400 V1.5 is furnished will be free from defects in material or workmanship under normal use at the time of delivery. KNAUER DOES NOT AND CANNOT WARRANT THE PERFORMANCE OR RESULTS LICENSEE MAY OBTAIN BY USING THE SOFTWARE OR DOCUMENTATION. THE FOREGOING STATES THE SOLE AND EXCLUSIVE REMEDIES KNAUER WILL PROVIDE FOR BREACH OF WARRANTY. EXCEPT FOR THE FOREGOING LIMITED WARRANTY, KNAUER MAKES NO WARRANTIES; EXPRESSED OR IMPLIED, AS TO NON-INFRINGEMENT OF THIRD PARTY RIGHTS, MERCHANTABILITY OR FITNESS FOR A PARTICULAR PURPOSE. Some countries do not allow the exclusion of implied warranties or limitations on how long an implied warranty may last, so the above limitations may not apply to the Licensee. This warranty gives the Licensee specific legal rights and the Licensee may also have other rights which vary from country to country.

## **7. Limitation of Liability.**

IN NO EVENT WILL KNAUER BE LIABLE TO LICENSEE FOR ANY SPECIAL DAMAGES, INCLUDING ANY LOST PROFITS, LOST SAVINGS, OR OTHER INCIDENTAL OR CONSEQUENTIAL DAMAGES - EVEN IF KNAUER HAS BEEN ADVISED OF THE POSSIBILITY OF SUCH DAMAGES, OR FOR ANY CLAIM BY ANY OTHER PARTY. Some countries do not allow the exclusion of limitation of special, incidental, or consequential damages, so the above limitation, or exclusion may not apply to the Licensee. Any liability is - in any case - limited to the license fee.

#### **8. Limitation of Remedies.**

KNAUER's entire liability and Licensee's exclusive remedy shall be:

(a) the replacement of any media not meeting KNAUER's limited warranty which is returned to KNAUER, or

(b) if KNAUER or its distributor is unable to deliver replacement media which is free of defects in materials or workmanship, the Licensee may terminate this Agreement by returning the EuroOsmo 7400 V1.5 and the Licensee's money will be refunded.

#### **9. This license is not assignable.**

The Licensee does not have the right to assign rights or grant sublicenses.

#### **10. License Fee / Royalty.**

For the license to use the contractual EuroOsmo®7400 V1.5 in the above defined scope and within the restrictions the Licensee pays to the Licensor a fee. The amount becomes due and payable upon delivery of the program discs. The license to use of the program commences with payment of the royalty.

**11. If a provision of this Agreement should be invalid or become invalid,** then the legal effect of the other provisions shall remain unchanged. Instead of an invalid provision, a valid provision shall be deemed to be agreed which comes closest to what the parties intended economically.

This document is the complete Agreement between KNAUER and Licensee; it supersedes all and any prior agreement or offer. All and any modifications of this agreement have to be made in writing.

For all controversies out of this Agreement, the District Court Berlin shall have exclusive jurisdiction. The applicable law shall be that of the Federal Republic of Germany (German Civil Code) excluding the UN-Code on the international sale of goods.



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