

Stage Top Incubator



STX -E series Instruction Manual



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For safe and comfortable use

About symbols and indications

These warnings enable safe and correct usage and prevent dangerous situations. Please follow them at all times.

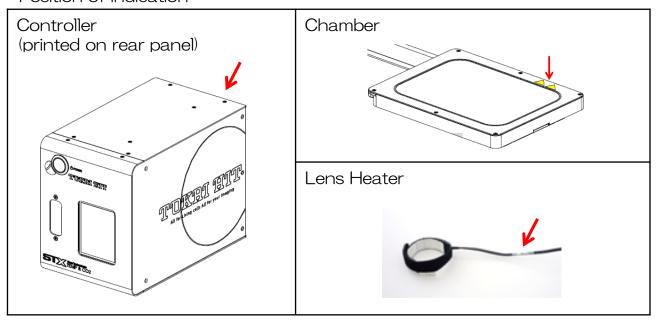
Symbol meaning

⚠ WARNING	Indicates a hazardous which could result serious injury.
∆ CAUTION	Indicates a situation which could result slight injury.
CAUTION	Indicates a situation which could damage the equipment or other apparatus.
NOTE	Point of attention

Indication meaning

\triangle	Injury or damage to equipment is possible
\bigcirc	Must not be done
0	Must be done
	High temperature possible
A	Electric shock possible
•	Information

Position of indication



To all users

- This device is to be used by researchers doing cultivation observations in a laboratory. It cannot be used at home.
- Do not take the equipment apart. This results in damage.

When shipping

- Please treat this precision instrument with care. Impacting it is forbidden.
- O not put any heavy things on the box.
- O Do not turn the box upside down.
- Do not let the unit moisten.

Regarding the environment for usage and storage

1 Do not use this equipment in the below conditions.

Flammable or corrosive gas or oil mist that can damage electrical insulation

Intense vibration or impact

High voltage power lines, inductive interference

Place where dew drops are present or with direct sunlight

Place with much dust

Points to pay attention when installing to a microscope

- Install the chamber in the state that the condenser is high enough. There is a risk that the condenser hits and breaks the top glass.
- Avoid stress on wires and tubes connected to the chamber.

- Connect the cylinder to the controller with supplied tube and fasten with band. Using another tube causes breakage and escape of gas. If the tube is not fastened well, there is a risk that it comes off.
- Do not bend the silicon tubes. If they are plugged, gas cannot be adequately supplied to the chamber.
- Do not use any gas other than specified. Gas leaks, dissolution of tubes and equipment, and malfunction may be occurred.
- Connect gas cylinder to the controller independently. If gas is blanched off and used with other devices, it may be the cause of trouble for both the equipment and other devices.
- Use only the specified electricity and cables. If other cables are used, this might result in electric shocks. Using a non-specified voltage might result in equipment damage.

When using the equipment

- Please use this equipment in a large room. If you use the equipment in a small tightly isolated room, CO2 concentration in air might rise and adversely affect human bodies.
- Do not handle the equipment with wet hands. It might cause damage to electric equipment.
 - In the case of glass breakage, do not touch the glass. It might result in injury and electric shock.
 - When revolving the objectives, be careful not to hit and damage both microscope and lens heater by the wire.
 - Take the following points into consideration in order to avoid focus drift during long-term microscope observations.
 - Keep the room temperature stable at 20°C-28°C.
 - Do not let air flow directly over the chamber.
 - With the exception of bath unit, wipe off media or water quickly. It can be the cause of damage.
 - Do not immerse the gas spouting nozzle into water. It may cause vibration and affect images under observation.



lacktriangle To pull out the plug from the outlet swiftly in an emergency, do not place any things near the outlet.

Storage



Turn off power supply while performing maintenance,

- When opening the chamber, do not inhale air in the chamber directly. It may adversely affect human bodies.
- Clean device with soft cloth using small amount of diluted neutral detergent. Do not use organic solvents.
- Do not wash the equipment with water.
- After use, make sure to remove all water completely. If some water remains, this can result in molt, breeding of bacteria and erosion.
- Do not use volatile materials such as benzene or thinner for cleaning. Use of such materials will discolor and/or damage key device surfaces.
- It is important to read this Instruction Manual before using the equipment. If it is used in manner other than specified in this manual, safety cannot be assured and it may result in damage to the equipment which will not be covered under warranty. Please follow the instructions

Summary

This equipment is an incubation system for research which has the following characteristics.

Long term observations of cell cultures are possible right on the microscope stage through reproduction of temperature, humidity and CO2 needed for cell culture. The temperature regulation is conducted by Top Heater of Chamber Unit, humidity with heating water in Water Bath. In order to achieve clear view during observation lid includes glass heater, even during high humidity no condensation appears.

The Lens Heater prevents heat from escaping to the objective lens under oil/water immersion.

The chamber is small and compact (stage top type) to be used on the microscope stage. It does not take much space around the microscope.

The optimal cell-culture environment is created with the controller equipped with PID control.

1 System Structure

1.1 Components

Name of components	Qty
Controller * STXG or STXF	1
Chamber	1
Lens Heater	1
Dish Attachment	Nlata
Fixing Lid	Note
Temperature Sensor	1
Extension Wire for Temperature Sensor	1
Gas Tube	2
Precision Screwdriver	1
Spare coverglass for LX-D35	1
Power Cable	1
Disposable Syringe for Draining Water	1

Note: Accessories differ depending on the product model.

Economy series include following items:

Included Dish Attachment and Fixing lid

ATX-A for ATX-D (holder)
ATX-W for wellplate (holder)
LX-W for wellplate (Lid)
UNIV2-SG-4 for 1-4pc slide glass holder

-OPTION-

- ① Sample Feedback regulation
- 2 STX-APP: Software
- ③ ATX-CSG for chamber slide, chambered coverglass and slide glass LX-CSG for chamber slide, chambered coverglass and slide glass ATX-D for 35mm dish and 50/60mm dish (dish holder) LX-D35 for 35mm dish (magnetic lid) LX-D56 for 50/60mm dish (magnetic lid)
- ※ Please refer to 7 Options (for STX-E upgrade) for more details.

2 Controller

2.1 Size and weight

Model	Dimension	Weight
STXG	W151xD263xH196 mm	3,8 kg
STXF	W151xD296xH196 mm	3,5 kg

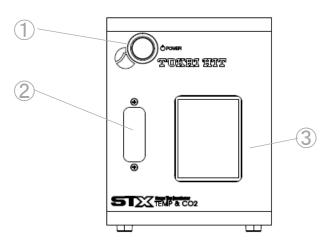
2.2 Name of parts

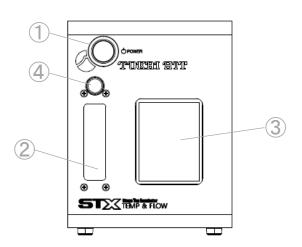
The image is shown for reference only.

Front panel

STXG with built-in gas mixer

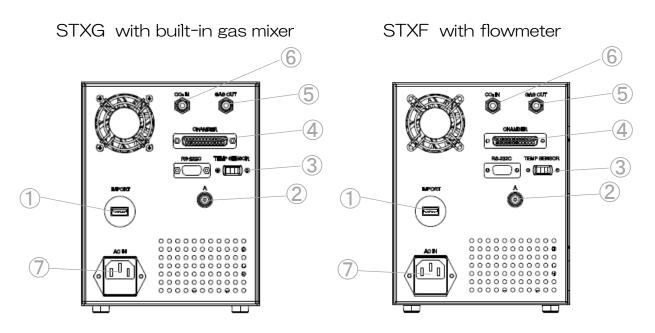
STXF with analog flowmeter





No.	Name	Role	Note
1	Main Switch	Power ON/OFF	
2	Touch Panel	Setting and indication	For the usage, see P.27-30.
3	Flowmeter	Indication of gas flow	
4	Flow Adjustment Knob	Adjustment of gas flow	Equipped to STXF

Rear Panel



No.	Name	Role	Note
1	USB Connection	Connection to PC	For connection to PC, see P.25.
2	Link Connection A	Connection to optional gas mixer	Use only if the system is used with STX optional gas mixer.
3	Sensor Connection	Connection to Extension Wire	
4	Chamber Connection	Connection to Chamber	
5	Mixed Gas Outlet	Gas supply to Chamber	
6	CO2 Inlet	Intake of 100% CO2 (STXG) 5% CO2 premixed gas (STXF)	For connection of tubes, see P.25-26.
7	Power Cable Connection		

2.3 Specifications

			STXG with built-in gas mixer	STXF with analog flowmeter
Temperature control		rol	PID control	
	Increments		0.1℃ step	-
	Setting method		Touch Panel or PC (optional, STX-APP requires)	
\exists		SAMPLE TEMP	30°C - 40°C (optional, STX-FB requires)	
Temperature	Possible temp. setting range	TOP HEATER	10℃ -	- 65°C
Dera	Controllable temp.	BATH HEATER	10℃ -	- 50°C
atur	setting range is from ambient +5°C	STAGE HEATER	10℃ -	- 50℃
O O		LENS HEATER	10°C - 45°C	
	Rise time		10 minutes to reach 50°C	
	Accuracy		Within ±0,3°C at each heater surface (in our terms and conditions)	
	Sensor (for heaters)		Pt100	
Hum dity	Humidification control		Forced humidification by heating Bath Unit	
ţ Ţ	Condensation prev	vention	Top Heater equipped with glass heater	
	CO2 concentration	range	5% - 20%	-
	Control method		PID control	Precision needle valve
Gas	Accuracy		Within ±0.1%	Within ±2% FS
SE	Gas type		100% CO2	5% CO2 premixed gas
	Input gas pressure		0.1Mpa - 0.15MPa	
	Output gas pressure		160 ml/min 75 ml/min - 250 ml/min	
Power source			100 V ~ 240 V AC ±10% 50/60Hz	
Maxin	Maximum power consumption		110W	90W

Power Code

For use in areas with 100V-120V power	Use only Power Cable described below: detachable cord set, 3-conductor grounding at 125V, AC125V7A minimum, listed in UL. In case of using with extension cord, use only Power Cable with PE (Protective Earth) wire.
For use in areas with 220V-240V power	Use only 3-pole Power Cable, with plug and outlet complying with EU/EN standards in EU territory. Class 1 equipment must be connected to PE (Protective Earth) terminal. In case of using with extension cord, use only Power Cable with PE (Protective Earth) wire.

Use Conditions

Location	Indoor	
Temperature	20°C - 28°C	
Relative Humidity	35% - 85%	
Altitude	Up to 2000m maximum	
Environmental Conditions	Installation category of IEC664, Pollution degree 2	

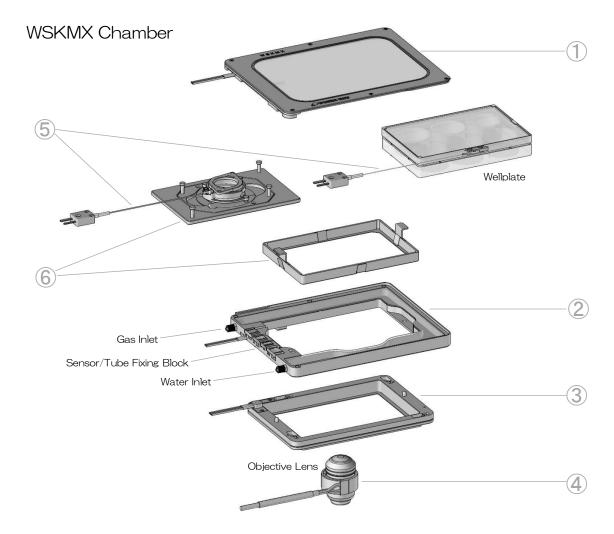
Safety Measures

Burn Prevention Function	When the Top Heater exceeds 65°C, safety function operates and temperature rise stops.
Fuse	250 V AC 2.0A Do not use except the rated fuse.

3 Chamber

3.1 Structure

The image is shown for reference only.



No.	Name	Role	Note
1	Top Heater	Condensation prevention Sample temperature control	
2	Bath Unit	Humidification inside of chamber by steam	Presence/absence of Bath Heater depends on the chamber model.
3	Stage Heater	Heats Dish AttachmentPrevents heat from escaping to the stage.	
4	Lens Heater	Prevents heat from escaping the objective lens.	For use of Lens Heater, see P.22.
5	Temperature Sensor	Measurement of sample temp.	Option: Please refer P.41.
6	Dish Attachment & Fixing Lid	• Fixation of vessel	Dish Attachment and Fixing Lid differ depending on the chamber model.

TIZWX

Applicable vessels















Applicable stage

Nikon Ti2/Ti piezo stage

Optional Stage Adapter requires for Ti2/Ti manual/motorized stage.

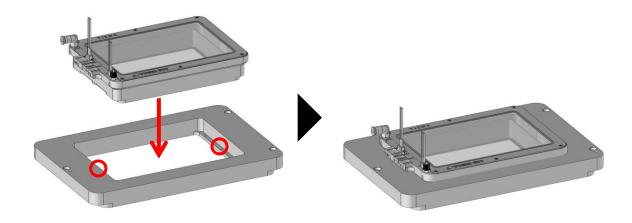
Dimension

Recommended water volume

 $W175.5 \times D118 \times H30 \text{ mm}$

26cc

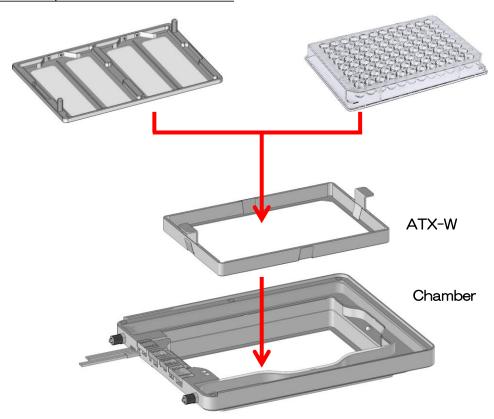
Installation



Remove Top Heater and use two screws to fix the chamber on piezo stage or optional Stage Adapter.

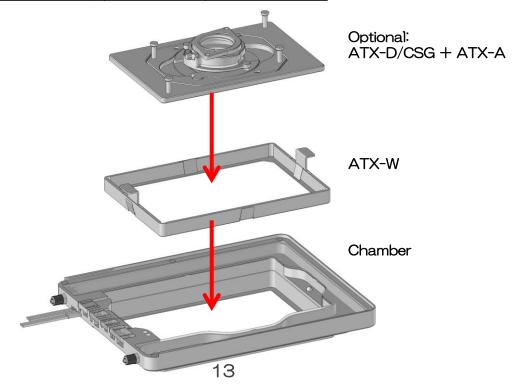
3.3 How to set Dish Attachment to Chamber

For wellplate or slide(s) holder



Simple place wellplate or slide holder to the ATX-W holder and insert them to Chamber Unit.

For small vessel use, please use optional holders.



3.4 How to set vessel to Dish Attachment (optional holder info)

See the figure below and set vessel.

35 mm dish



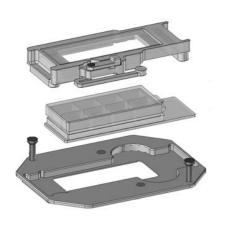
Use Adapter for 35 mm dish and fix by magnets of Fixing Lid LX-D35.

50/60 mm dish



Remove Adapter for 35 mm dish and fix by magnets of Fixing Lid LX-D56.

Chamber slide and Slide glass (Option)

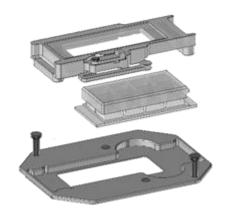


LX-CSG



Place the slide on ATX-CSG and mount Fixing Lid LX-CSG.

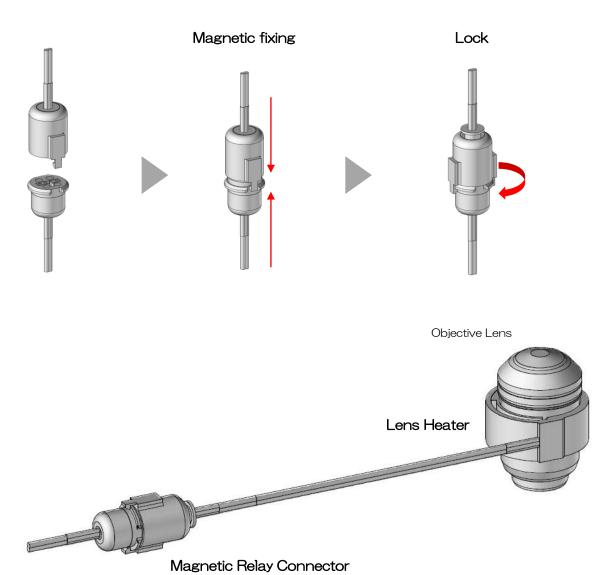
Chambered coverglass (Option)



Place the chambered coverglass and mount Fixing Lid LX-CSG. Please confirm that the opening of LX-CSG is completely covered by chambered coverglass.

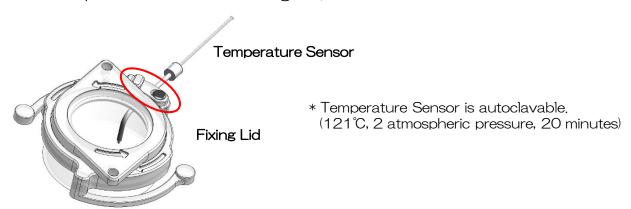
3.5 How to set Lens Heater

Wrap Lens Heater so as to be in close contact with the objective lens. Avoid uneven surface such as correction collar and diaphragm. The cable has magnetic Relay Connector to prevent the cable from getting entangled around the objective revolver.

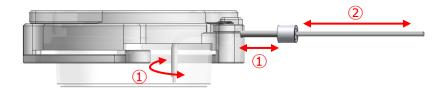


3.6 How to set Temperature Sensor

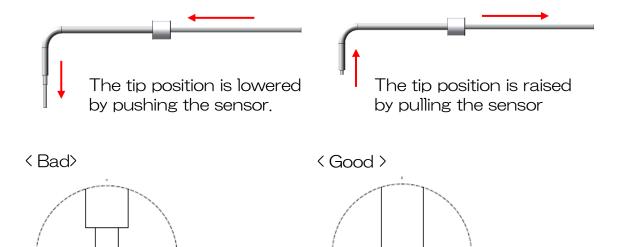
Set Temperature Sensor to Fixing Lid.



Set Temperature Sensor so that the tip is dipped properly into media in the vessel.



- 1) Adjust the position and angle of the pipe.
- 2 Adjust the height of the sensor tip.

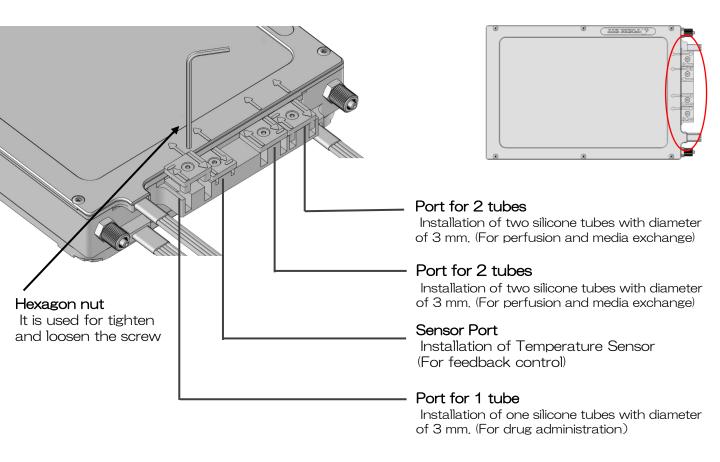


The sensor tip is not dipped into media.

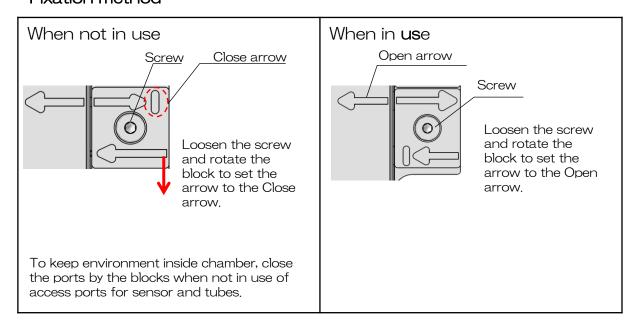
The sensor tip is dipped into media.

3.7 Access Ports

Set Fixing Blocks properly according to use/non-use of Temperature Sensor and tubes.

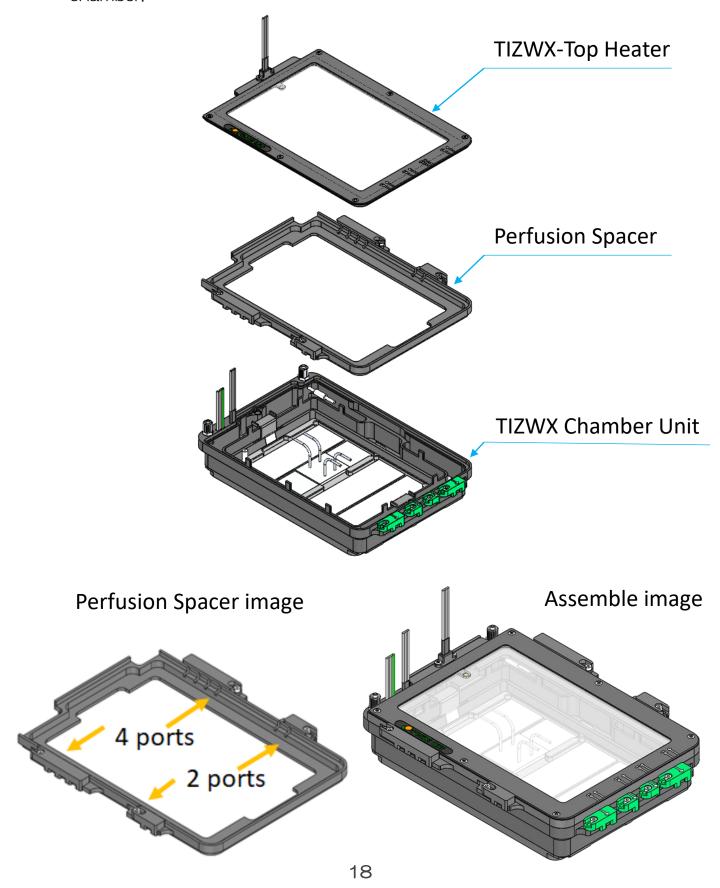


Fixation method



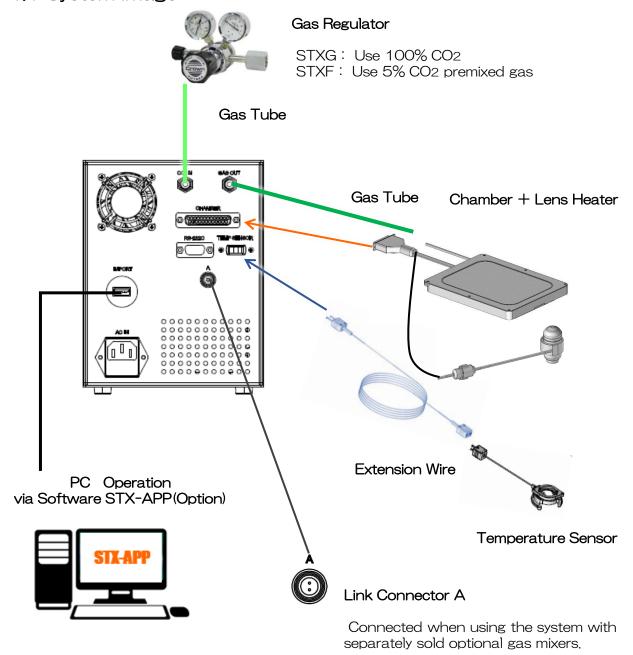
3.8 additional Access Ports (optional)

With optional perfusion spacer, you can have additional 12 ports to the chamber.



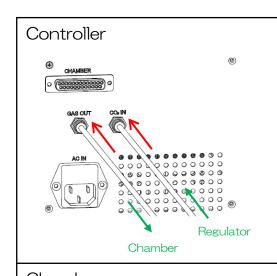
4 Connection

4.1 System Image



- 1 Connect Power Cable between controller and the outlet.
- 2 Set Lens Heater to the objective lens.
- 3 Install the chamber on the microscope stage.
- 4 Plug the connector of chamber cable into the socket at the rear panel of controller.

4.2 Connection of Gas Tubes



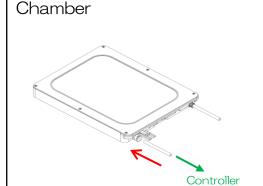
STXG

Connect Gas Tube between the regulator of 100% CO2 and CO2 IN at the rear panel of controller.

STXF

Connect Gas Tube between the regulator of 5% CO2 premixed gas and CO2 IN at the rear panel of controller.

Connect another Gas Tube to GAS OUT.



Connect the opposite side of Gas Tube connected to controller to the gas inlet of chamber.

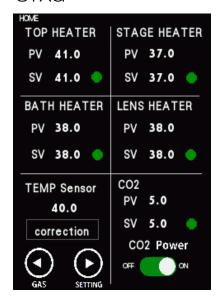
The Gas Tube is connected to the chamber in advance at the time of factory shipping.

5 Operation

5.1 Touch Panel

HOME

STXG



STXF

HOME		
TOP HEATER	STAGE HEATER	
PV 46.0	PV 39.0	
sv 46.0	sv 39.0	
BATH HEATER	LENS HEATER	
PV 41.0	PV 37.0	
sv 41.0	sv 37.0	
TEMP Sensor		
0.0		
correction		
GAS	SETTING	

< TOP HEATER / BATH HEATER / STAGE HEATER / LENS HEATER >

Shows Process Value and Setting Value of Top Heater, Bath Heater, Stage Heater and Lens Heater. When one area is tapped in each heater area, Setting Value can be changed.



The heater is functioning normally.



When using WELSX, GSI2X, TIZWX or DMIWX, the error is displayed in area of Bath Heater since the Bath Heater is not included.

< Correction / TEMP sensor>

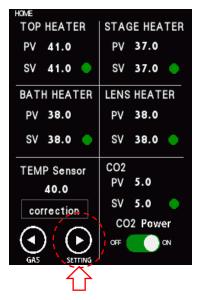
By clicking the button, it can input the correction value of sensor which is included in original package possible (off-set of sensor)

< CO2 / CO2 Power > Only STXG

Shows Process Value and Setting Value of CO2 concentration of mixed gas supplied from controller. When the CO2 Power is turned ON, the mixed gas begins to be supplied.

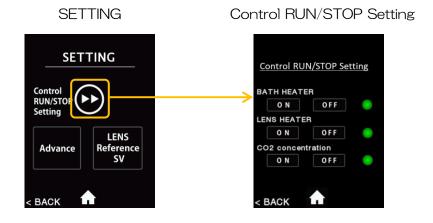
SETTING

Tapping SETTING button, the display changes to the screen for ON/OFF setting of heaters and gas supply, the reference setting value of Lens Heater and parameters.



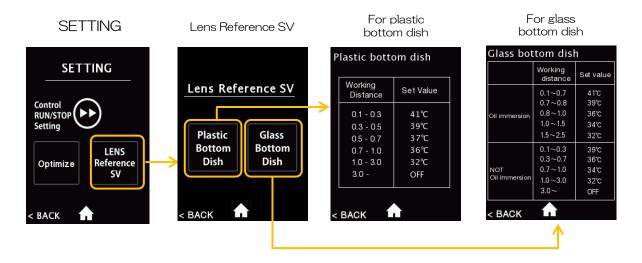
<ON/OFF setting >

You can turn ON/OFF for Bath Heater, Lens Heater and gas supply. When using the chamber which does not have Bath Heater, the Bath Heater is set to OFF in advance.



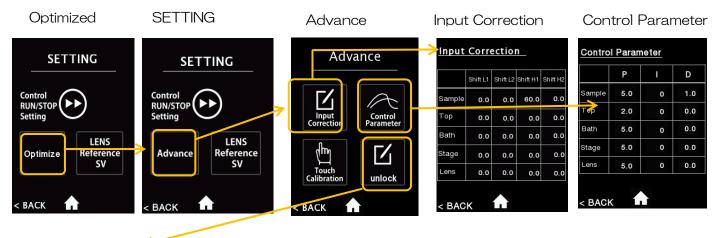
< Reference Setting Value of Lens Heater >

You can check reference Setting Value of Lens Heater. Change the SV depending on vessel type and objective lens in use.



< Advance Setting >

You can change Input Correction values and Control Parameter values. The values calculated by calibration at Tokai Hit are inputted at the time of factory shipping. Do not change the default values for any purpose other than changing the chamber. The default value are printed on the top surface of controller.

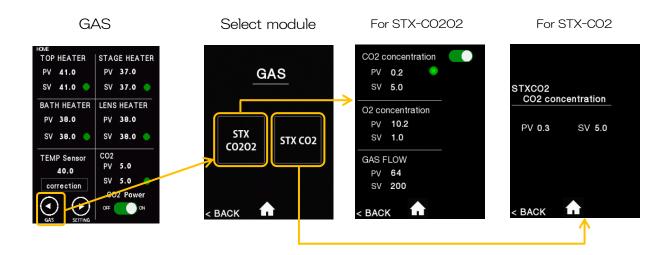


<unlock>

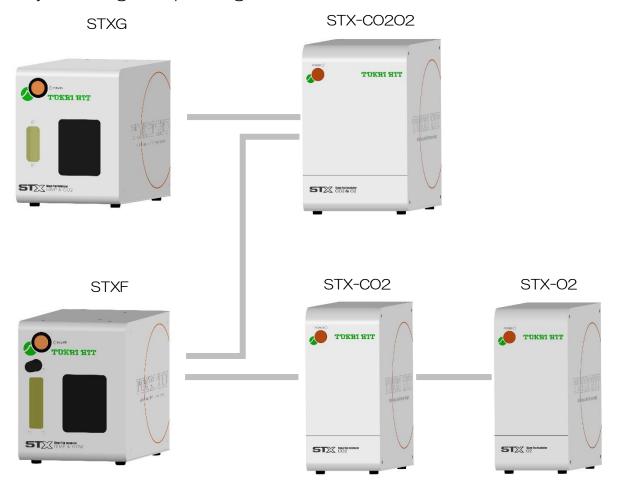
The unlock is only used when adding optional functions of "Sample Feedback" and "software and communication port" only. It requires password and USB/CD to unlock these functions. If you were interested in these function, please contact Tokai Hit at solution@tokaihit.com

< Optional Gas Mixer >

When using the system with optional gas mixers, tap GAS button at HOME screen and set CO₂/O₂ concentration and gas flow.



System Image of optional gas mixers



5.2 Reference Setting Value

Reference Setting Value is described at the seal at front panel of controller. It is provided to heat the dish bottom temperature to 36.5°C-37.0°C under room temperature of 25°C. Temperature inside chamber may vary depending on actual use condition.

example

S/N	35mmDish	Well plate
ТОР	48.0	48.5
BATH	41.0	41.0
STAGE	38.0	37.5
LENS		
SAMPLE	37.0	37.0

Shows Reference Setting Value for 35mm dish and wellplate.

For Reference Setting value of Lens Heater, see P.29.

5.3 Work-flow

Preliminary Operation

Carry out preliminary operation for about 1 hour.



Gas Supply

Start gas supply in 30 minutes before experiment.



Cultivation

A Reference Setting Value

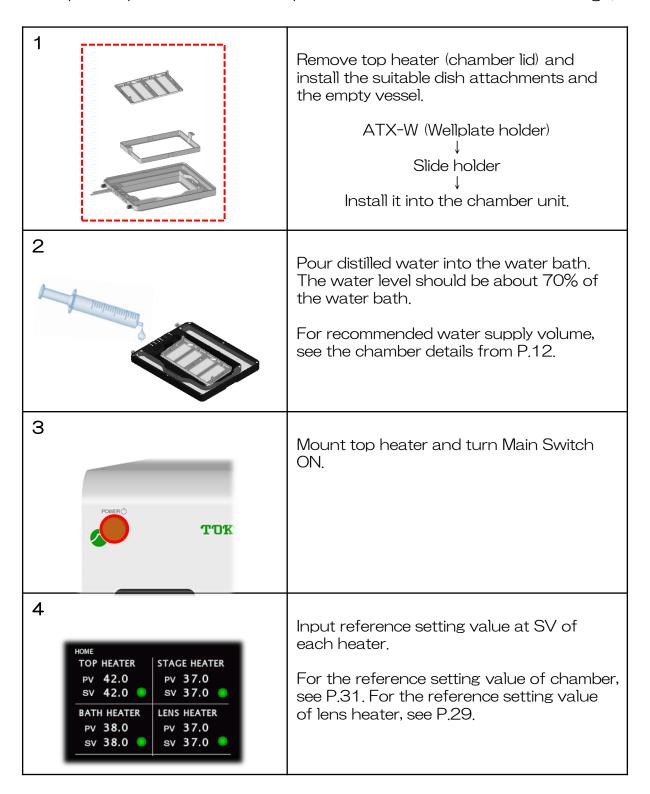
Easily start experiment by reference setting value.

B Feedback (Option)

Actual sample temperature can be checked and adjusted in real time. It is possible to maintain the sample temperature precisely even if room temperature is not stable.

5.4 Preliminary Operation

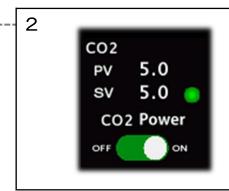
In order to stabilize the environment inside/outside the chamber, carry out preliminary operation for about 1 hour before experiment. Making the temperature of microscope and chamber before observation keeps the sample temperature stable and prevents drift of the observation image.



5.5 Gas Supply

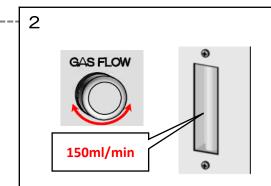
- 1) Confirm regulator valve 2 is closed.
- 2) Confirm pressure adjustment 3 is slack.
- 3) Open cylinder valve 1).
- 4) Open regulator valve 2.
- 5) Turn pressure adjustment ③ toward right to adjust secondary pressure indicated in meter B to be in a range from 0.1MPa to 0.15MPa.





Set CO2 concentration and turn CO2 Power ON at touch panel of controller or software STX-APP.

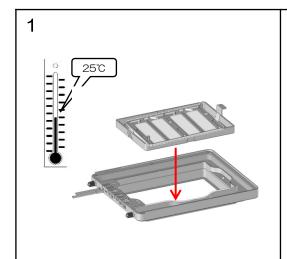
STXF



Turn the adjustment knob and adjust flow rate at flowmeter to 150ml/min.

5.6 Cultivation

A Reference Setting Value



Confirm room temperature is stable at around 25°C.

Remove the empty vessel from the chamber and install the observation vessel.



Input reference setting value at SV of each heater.

For the reference setting value of chamber, please find it at the front right hand bottom corner (see image bellow).

Describing position of reference setting value



example

S/N	35mmDish	Well plate
TOP	48.0	48.5
BATH	41.0	41.0
STAGE	38.0	37.5
LENS		
SAMPLE	37.0	37.0

5.7 Cleaning

1 Shut off regulator valve 2 and then 1 to stop CO₂ supply. 2 Turn Main Switch OFF. Remove top heater carefully and wipe dew water at the inside surface. Remove remaining water at water bath with using syringe. 3 Remove chamber and lens heater from the microscope. 4 Clean the chamber with using cloth with alcohol. If water drops are left inside water bath, it may cause mold.

6 Note and Trouble-shooting

Check List for Gas Supply

Please confirm the following when Process Value of gas concentration is not stable at Setting Value or gas flow rate does not increase.



- A: Primary pressure meter Remaining amount of gas
- B: Secondary pressure meter Delivery gas pressure

- 1 Confirm the secondary pressure indicated in meter B to be in a range from 0.1MPa to 0.15MPa.
- 2 Confirm the remaining gas amount.
- 3 Confirm both cylinder valve 1) and regulator valve 2 are opened.
- 4 Connect gas tubes without buckling it.
- 5 Confirm gas is supplied to the controller from the regulator. Ensure that gas is delivered to the controller-side of tube.
- 6 < STXG > Confirm there is no abnormal noises resulting from gas leakage.

If the trouble cannot be solved, please contact Tokai Hit.

Vessel to be used

35 mm petri dish

The petri dishes of Iwaki, Nunc or Greiner have a cutout at the bottom and there is a possibility that the incubation environment cannot be kept stable or dew condensation is generated on the lens heater due to leakage. In a case of that, please use the exclusive spacer sold separately.



Chamber slide / Chambered coverglass

General slides with 75 x 25 mm can be used on the standard attachment.

The lid of Lab-Tek I has a structure of handle. To use this type of vessel, please set Fixing Lid on the vessel without lid or cut off the handle in advance.

Wellplate for WSKMX, IX3WX, TIZWX or DMIWX chamber

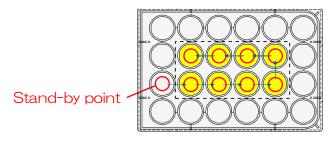
Multi wellplates with 127×85 mm can be used in the chamber. Please avoid the wellplate which has cutouts at the side or bottom.





Note for multi points time-lapse:

When using the objective lens with short working distance, sample heat escapes to the lens. In order to minimize temperature gap between wells, please set the stand-by position to a well without sample.



Trouble-shooting

Status	Guidance
"XXX Heater SENSOR ERROR" on the status information	Confirm if the chamber unit cable are properly connected. If the display has not change then there may be a potential for sensor disconnection. Please contact dealer or Tokai Hit.
PV does not keep stable at SV after preliminary operation.	Confirm the plug coming from chamber is connected to controller properly. The PV reaches to the SV within 30 minutes after power on. Please avoid direct impact from air conditioner to stabilize the heaters.
PV becomes higher than SV and stay it high.	First, please run the system without feedback control. If the trouble cannot be solved, there is a possibility of the heating cable disconnection. Please contact dealer or Tokai Hit.
Media evaporates fast.	Evaporation rate depends on use environment, but approximate rate is 0.5-1.0ml per an hour. If water does not remain in water bath, please refill water through water inlet tube. Possibly the Top Heater is not placed correctly.
Water in water bath evaporates too fast.	Possibly the Top Heater is not placed correctly. Ensure the Top Heater is set on chamber properly.
Dew condensation is generated at the bottom glass of Top Heater.	Decrease the Bath Heater SV by 1.0-2.0 or increase the Top Heater SV by 1.0-2.0 as needed.
Dew condensation is generated at the objective lens.	There may be cut-out at the bottom of dish, which causes leakage of humidity. Please use the dish without cut-out or use the exclusive spacer sold separately.

Status	Guidance	
Glass breakage of Top Heater	Please contact dealer or Tokai Hit with serial number of your device.	
Gas flow rate does not increase enough.		
Mixed gas is not supplied from GAS OUT of controller.		
Floater in flowmeter stays at the bottom.		
	If the trouble cannot be solved, please contact Tokai Hit.	
How to clean chamber and accessories?	Clean the chamber with using cloth with alcohol. The chamber is not autoclavable but Temperature Sensor and Fixing Lid corresponds to autoclave sterilization.	
Cannot incubate well.	Confirm the system status (temp., room temp., gas supply) and sample status (color, cell condition).	
	Temperature: Avoid direct impact from air conditioner. Measure the actual temperature in the vessel and adjust the heaters to maintain the sample at the target temperature.	
	CO2: The concentration cannot be stable if the input pressure to controller is less than 0.1 MPa. If it is more than 0.3 MPa, there is a possibility of malfunction.	
	Humidity: If the Top Heater or Dish Attachment is not installed properly, it may affect to humidifying function and water in bath unit evaporates earlier than usual.	

7 Options (for STX-E upgrade)

Optional STX-FB: Sample Feedback regulation

Sample Temperature Feedback Regulation

allows to keep the optimal sample temperature from

room temperature fluctuation.



- 1. Feedback actual media temp. to Controller
- 2. Controller regulate main heater based on the feedback signal
- 35

 30

 25

 30

 25

 Temp. in dish

 Room Temp. (18°C 30°C)

 Temperature Stability in chamber during room temperature change (25°C±5°C)

*This feature can be unlocked after purchase by optional Pass word and USB memory.

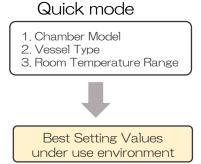
Optional STX-APP-UL: Software features

STX-APP: Software for multiple application

Intuitive Operation via User Graphic Interface



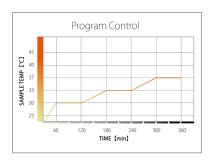




Screen transfer



Timer Setting of Temperature and Gas Concentration



This feature can be unlocked after purchase by optional Pass word and installation CD.

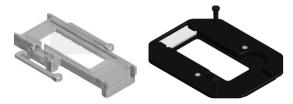
7 Options (for STX-E upgrade)

Optional common add-on options.

Following options can be purchased individually. Please contact Tokai Hit for more information.

STX-CSG

Dish Holder set for chamber slide, Slide glass and chambered coverglass



ATX-D35-2-SET

Dish Holder set for 35mm dish (2pc)



TIZWX-SP12-A21K

Add-on perfusion spacer with 12 access ports for Synvivo Tygon 0.6" tube.



STX-C0202

External Gas mixer for hypoxia.

CO2: 5.0 - 20.0% O2 : 0.1 - 18.0%



Tokai Hit Customer Support

Please contact our support team with following information.

- 1. Description of inquiry or problem
 - Inquiry of regarding usage,
 - Details of malfunction or defect
 - Request of additional accessories and optional products
- 2. Serial Number

- Six-digit number at chamber cable and controller

Email: solution@tokaihit.com

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