

OLYMPUS[®]

INSTRUCTIONS

IX81-ZDC

FOCUS DRIFT COMPENSATING MICROSCOPE

This instruction manual is for the Olympus Focus Drift Compensating Microscope Model IX81-ZDC. To ensure the safety, obtain optimum performance and to familiarize yourself fully with the use of this microscope, we recommend that you study this manual thoroughly before operating the microscope. Retain this instruction manual in an easily accessible place near the work desk for future reference.



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CE Compliance

This device complies with the requirements of both directive **2004/108/EC** concerning electromagnetic compatibility and directive **2006/95/EC** concerning low voltage. The CE marking indicates compliance with the above directives.

Use in domestic area

EN61326-1 defines two categories according to the location for use.

Class A: Equipment suitable for use in establishments other than domestic, and those directly connected to a low voltage power supply network which supplies buildings used for domestic purposes.

Class B: Equipment for use in domestic establishments, and in establishments directly connected to a low voltage power supply network which supplies buildings used for domestic purposes.

This system is applied Class A. Some interference may occur if this system is used in domestic location.

Safety and EMC Precautions

1. Use only power cord which OLYMPUS specifies. Otherwise the safety and EMC performance of the product can not be assured.
2. Be sure to ground the product. Otherwise our intended electric safety and EMC performance of the product can not be assured.

NOTE: This equipment has been tested and found to comply with the limits for a Class A digital device, pursuant to Part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this equipment in a residential area is likely to cause harmful interference in which case the user will be required to correct the interference at his own expense.

FCC WARNING: Changes or modifications not expressly approved by the party responsible for compliance could void the user's authority to operate the equipment.

CONTENTS

IMPORTANT – Be sure to read this section for safe use of the equipment. – 1-5
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1	NOMENCLATURE	6
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2	FOCUS DRIFT COMPENSATION OPERATION	7
----------	---	----------

3	SPECIFICATIONS	7
----------	-----------------------	----------

4	TROUBLESHOOTING GUIDE	8
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IMPORTANT

- ◎ The IX81-ZDC microscope features the capability of compensating for the focus drift that poses problems during long hours of time-lapse observation of live cells.
However, as the IX81-ZDC compensates for focus drift by detecting the primary image (1X) of the objective, the correct focus drift compensation cannot be obtained unless the detection light path and TV light path (right, left or bottom port) offers the primary image (1X) of the same condition.
- ◎ This manual pertains only to the information related to the ZDC (focus drift compensation) facility of the microscope. Please also refer to the instruction manuals for the microscope and associated modules.
- ◎ The application software can be controlled with the following software (before using the software, consult its manufacturer if its version is compatible with the IX81-ZDC):
 - 3I (Slide Book); • OBS (Experimental Manager); • UIC (MetaMorph).

Safety Precautions




1. The focus drift compensation function uses a laser diode (wavelength 785 nm) for autofocusing. It is designed to be safe by reducing the laser power (Class 1), it is still not recommended to view the laser light directly by removing the cover, etc. (Before maintenance, be sure to set the power switch to "○" (OFF).)
Never remove the warning and caution labels on the microscope.
The semiconductor laser for autofocusing incorporated in this unit makes it designated as a product of the following class.

CLASS 1 (IEC60825-1:1993+A1:1997+A2:2001)
CLASS I (CDRH21 CFR1040)

2. Always use the power cord provided by Olympus. If the proper power cord is not used, product safety performance cannot be warranted.
3. Always ensure that the **grounding terminal** is properly grounded. If the equipment is not grounded, Olympus can no longer warrant the electrical safety performance of the equipment.
4. Distribute the connection cables at a distance from the lamp housing. If a connection cable contacts the lamp housing or its surroundings, the cable may be melted and cause an electric shock hazard.
5. Never insert metallic objects into the air vents of the microscope as this could result in electrical shock, personal injury and equipment damage.
6. To prevent the microscope system from falling down, do not install modules which cause the microscope system height to exceed 1 meter.

Safety Symbols

The following symbols are found on the microscope. Study the meaning of the symbols and always use the equipment in the safest possible manner.

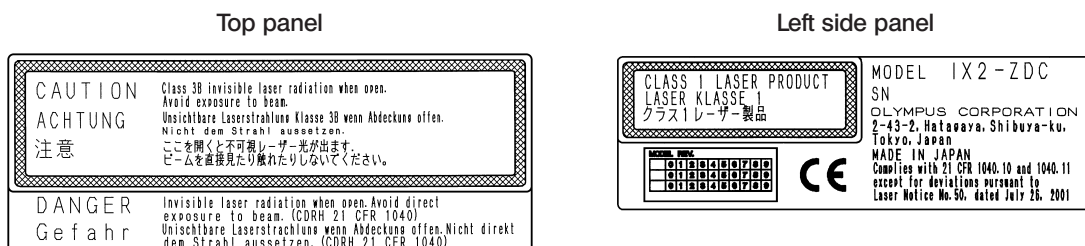
Symbol	Explanation
	Before use, carefully read the instruction manual. Improper use could result in personal injury to the user and/or damage to the equipment.
	Indicates that the main switch is ON.
	Indicates that the main switch is OFF.

CAUTION Control or adjustments using procedures other than those described in this manual may result in hazardous exposure to the laser radiation.

Warning labels

Warning labels related to laser beam are attached to the microscope. Always heed the warnings.

Warning label position (AF sensor unit)



When a warning label becomes dirty or is peeled off, contact Olympus for replacement.

1 Getting Ready

1. The microscope is a precision instrument. Handle it with care and avoid subjecting it to sudden or severe impact.
2. Do not use the microscope where it is subjected to direct sunlight, high temperature and humidity, dust or vibrations. (For the operating conditions, refer to chapter 3, "SPECIFICATIONS" on page 7)
3. For the objectives applicable to the microscope, see the list on page 4.

2 Maintenance and Storage

1. Do not attempt to use organic solvents to clean the microscope and the modules but wipe them dry with a soft cloth. When they are extremely dirty, use a lint-free, soft cloth slightly moistened with a diluted neutral detergent.
2. Do not disassemble any part of the unit as this could result in malfunction or reduced performance. Particularly, the stage support cannot be removed because it has been bonded after adjustment.
3. When not using the microscope system, make sure that the lamp housing is cool enough, and cover the microscope system with the dust cover before storage.

3 Caution

If the microscope is used in a manner not specified by this manual, the safety of the user may be imperiled. In addition, the equipment may also be damaged. Always use the equipment as outlined in this instruction manual.

The following symbols are used to set off text in this instruction manual.

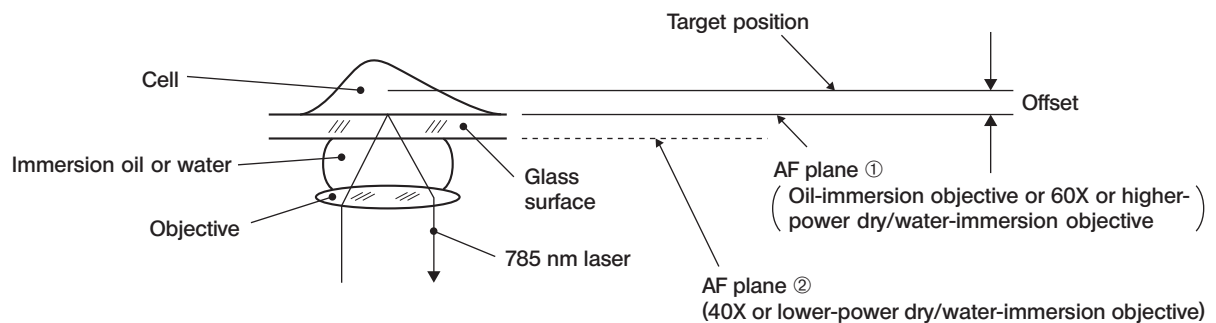
- ▲ : Indicates that failure to follow the instructions in the warning could result in bodily harm to the user and/or damage to equipment (including objects in the vicinity of the equipment).
- ★ : Indicates that failure to follow the instructions could result in damage to equipment.
- ◎ : Indicates commentary (for ease of operation and maintenance).

Notes on ZDC (Focus Drift Compensation)

1. Principles

The focus drift compensation focuses a 785 nm laser beam on the glass surface of a glass bottom dish*, etc., and applies software offsetting to the observation target position to reproduce the desired focus position. This is the difference from the autofocusing which focuses directly on the desired cell. The focus drift compensation is mainly used in fluorescent light observations.

* **CAUTION** Focus drift compensation is not available with other materials than glass (such as plastic).



The focal plane of oil-immersion objectives is focal plane ①.

The focal plane of dry objectives with 40X or lower power is focal plane ② when the glass thickness of the glass bottom dish is below 0.17 mm. A 60X objective has two focal planes ① and ② provided that the glass thickness is 0.17 mm or more.

The focal plane of water-immersion objectives varies depending on the AF zone setup and glass thickness.

When the glass thickness is below 0.17 mm or when a 40X objective is used, the focal plane is usually focal plane ②. When the glass thickness is 0.17 mm or more and a 60X objective is used, there are two focal planes ① and ②.

The selection of focal plane ① or ② is performed by setup of the AF zone (it is usually selected automatically by the application software). Focusing on focal plane ② can be prevented by setting the AF zone in a range that contains focal plane ① but does not contain focal plane ②. When the AF zone is set so that it contains focal plane ② but does not contain focal plane ①, it is possible to prevent focusing on focal plane ①. If the AF zone is set so that it contains both focal planes ① and ②, the focus point comes on focal plane ① or ② that is closer to the AF start point than the other focal plane.

(When focusing on either focal plane ① or ② is required, set the AF zone width within $\pm 20 \mu\text{m}$.)

© For details on the AF zone, refer to "Application Software."

2. Applicable microscope model: IX81-ZDC

3. Applicable objectives: ★ If objective has an aperture, set it wide open.

★ The AF focusing speed is less than 1 sec. in the focusing range, but slows down depending on the AF search range setting.

UIS2 Series

Objective Name	NA	WD (mm)
PlanApoN 60XO	1.42	0.15
UPlanSApo 20X	0.75	0.60
UPlanSApo 40X	0.90	0.18
UPlanSApo 40X2	0.95	0.18
UPlanSApo 60X O	1.35	0.15
UPlanSApo 60X W	1.20	0.28
UPlanSApo 100X O	1.40	0.13
UPlanFLN 20X	0.50	2.10
UPlanFLN 40X	0.75	0.51
UPlanFLN 60X	0.90	0.20
UPlanFLN 100X O/O2	1.30	0.20
UPlanFLN 100X OI/OI2	1.3-0.6	0.20
LUCPlanFLN 20X	0.45	6.6-7.8
LUCPlanFLN 40X	0.60	2.7-4.0
LUCPlanFLN 60X	0.70	1.5-2.2

★ When using a large specimen such as a micro-test plate, focusing may not be achieved due to its inclination.
The focusing is guaranteed only with a glass bottom dish with a diameter of 35 mm.

UIS Series

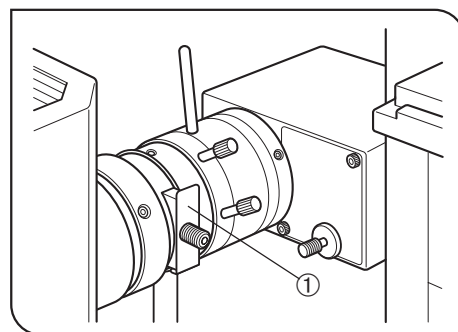
Objective Name*	NA	WD (mm)
PlanApo 60X O3	1.40	0.15
PlanApo 100X O3	1.40	0.10
UPlanApo 60X	0.90	0.20
UPlanApo 60X W3	1.20	0.25
UPlanApo 60X W3/IR	1.20	0.28
UPlanApo 100X OI3	1.35	0.10
UApo 20X 3/340	0.75	0.55
UApo 40X 3/340	0.90	0.20
UApo 40X W3/340	0.70	0.40
UPlanFI 20X	0.50	1.60
UPlanFI 40X	0.75	0.51
UPlanFI 100X O3	1.30	0.10
PlanApo 40X W/LSM	0.90	0.16
PlanApo 60X O/LSM	1.10	0.13
PlanApo 60X O TIRFM-SP	1.45	0.15
UPlanFI 40X O-SP	1.30	0.12
LCPlanFI 40X	0.60	2.60
SLCPlanFI 40X	0.55	6.4 - 8.3
LUCPlanFI 40X	0.60	3.40
LCPlanFI 60X	0.70	1.70

(Special order)
(Special order)

★ When using a large specimen such as a micro-test plate, focusing may not be achieved due to its inclination.
The focusing is guaranteed only with a glass bottom dish with a diameter of 35 mm.

* These objectives can be used regardless of the model number (3, 2 or none).

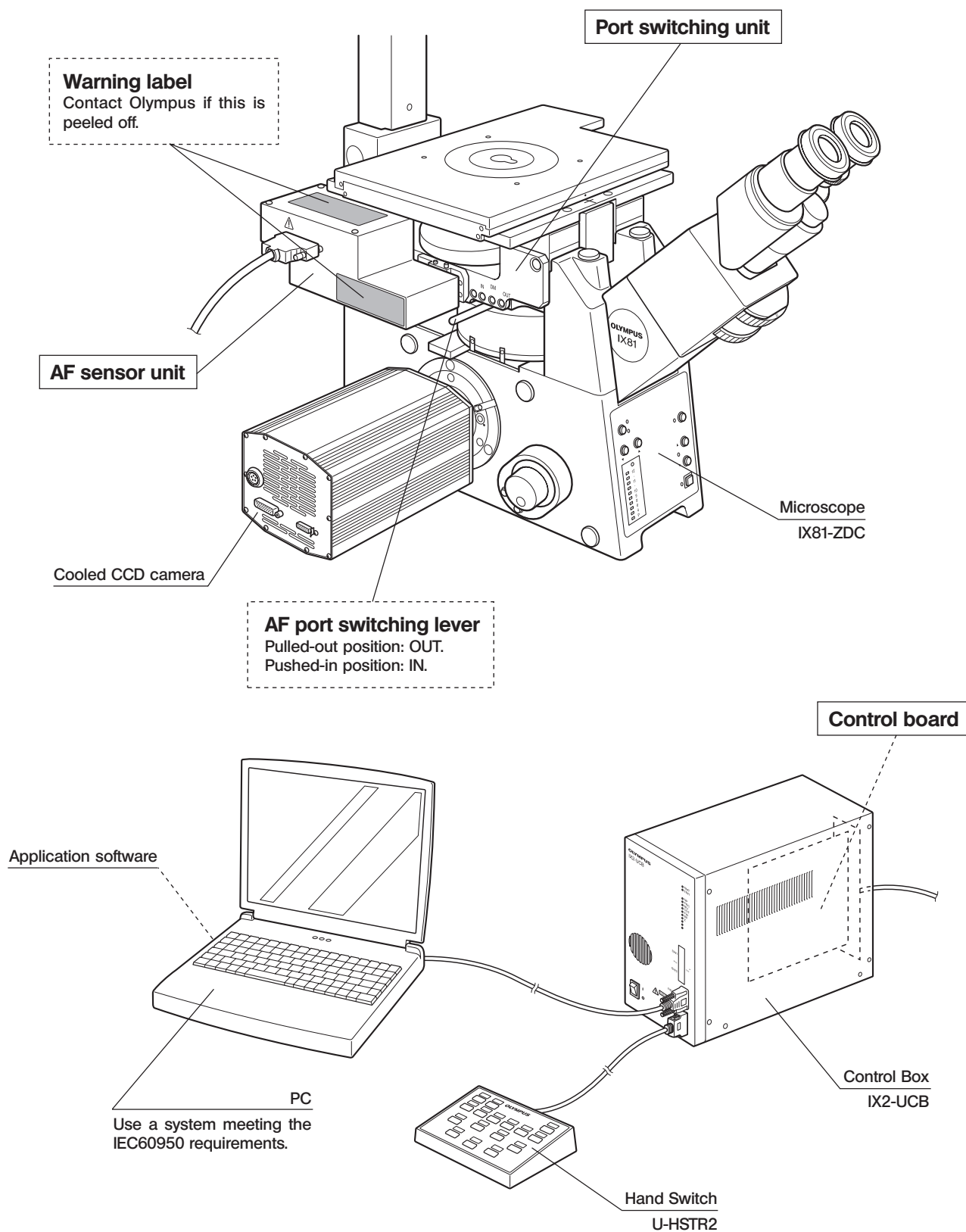
4. If the target position is apart from the glass surface, real-time (tracking) AF is not possible due to the necessity of offsetting.
5. With DIC (Differential Interference Contrast) observation, focus compensation is possible only with the gray sensitive color. It is neither possible in phase contrast observation.
6. Operating environmental temperatures: 5 to 40°C (41 to 104°F)
(Focus compensation is possible under a temperature variation range within 5°C (9°F) and when the microscope is protected against the wind from air conditioners.)
When the FV (FLUOVIEW) system is combined, the compensation performance may drop slightly due to focus error produced inside the scan unit.
7. Restrictions on combined modules
 - The module that can be mounted on the left side port is only a TV adapter that can project the primary image in 1X magnification, that is, the U-TV1X, U-DPCAD or U-SIP. (Optimal AF performance cannot be achieved with other combination.)
 - Notes on observation tube
 - a) When the U-TBI90 observation tube is used, the image in the peripheral part of eyepiece's visual field may be obscured (about 5%). In addition, no intermediate attachment can be used with the U-TBI90.
 - b) When the U-BI90/BI90CT observation tube is used together with a 4X or 10X objective, the image in the peripheral part of eyepiece's visual field may be obscured. In addition, the IX2-CA2X magnification changer lens unit cannot be used with the U-BI90/BI90CT (it becomes usable by mounting the U-EPA2 eye-point adjuster (occupying only 1 stage) on the U-BI90/U-BI90CT).
 - Notes on intermediate attachments
 - a) Only one U-EPA2 eye-point adjuster can be mounted (it cannot be used together with other intermediate attachments).
 - b) Only one U-FWO filter wheel can be mounted (it cannot be used together with other intermediate attachments).
 - c) When the IX-ATU intermediate attachment and GX-SPU side port unit are combined, the image in the peripheral part of eyepiece's visual field may be obscured if the magnification changer knob of the microscope frame is set to 1X.
 - d) The U-CA/U-ECA/U-ECA1.6X magnification changer cannot be used in combination with the microscope frame.
 - When the IX-ATU intermediate attachment and U-TR30H trinocular observation tube are combined, the only TV adapter that can be used is the U-TV1X.
 - When the GX-SPU side port unit and a TV camera are used, the only TV adapter that can be used is the U-TV1X.
 - The FN of the right side port is specified as 16. When a 10X or lower-power objective a CCD camera are used, the brightness in the peripheral area may be slightly insufficient.
 - When a 2X, 4X or 10X objective is used with FN 22, the brightness in the peripheral area may be slightly insufficient. (This problem does not occur with FN 16.)
 - When a 10X objective is used in transmitted light observation using the IX2-ULWCD condenser, the illumination NA may become insufficient and the resolution may degrade. Also, optimum phase contrast observation is not available using a phase contrast objective (PH1 or PH2).
 - When using the IX2-RFAW, remove the light shield ① in the excitation filter slider on the left illuminator to prevent it from coming in the way of operation.



- Optimum phase contrast observation is not available when the IX2-ILL30 transmitted light illumination column and a phase contrast observation (PH1 or PH2) are used.
- AF does not function normally when the U-LH100IR transmitted light IR lamp housing is used. To achieve AF, it is required to close the shutter or turn off the light during AF.
- During TIRFM observation, autofocusing may malfunction when the laser light of TIRFM penetrates in the AF sensor. To prevent this, block the TIRFM observation light (laser light) when the IX81-ZDC is used for AF detection.
- The IBMU/IBML temperature retention box cannot be used because a clearance is produced between the temperature retention box and transmitted light illumination column.
- Focus drift compensation of the IX81-ZDC is impossible with the MI-IBC carbon dioxide culture system of the previous version due to focus pulsing of the MI-IBC. If you want to use a carbon dioxide culture system, be sure to use the MI-IBC of the new version (released in February 2004).

1 NOMENCLATURE

◎ The module names enclosed in are involved in focus drift compensation.
The assembly and adjustments are to be done by Olympus.



2 FOCUS DRIFT COMPENSATION OPERATION

1. Set the power switches of the IX2-UCB control box and PC to "I" (ON).
2. Set the AF port switching lever ① of the AF sensor unit to the pushed-in position (IN).

☉ This operation engages the dichroic mirror (DM) in the light path of the AF sensor unit, making AF detection possible.

If AF detection is not necessary, setting the lever ① to the OUT position makes it possible to eliminate the small loss of the observation light. This is also effective in observation with IR light (785 nm or higher wavelengths).

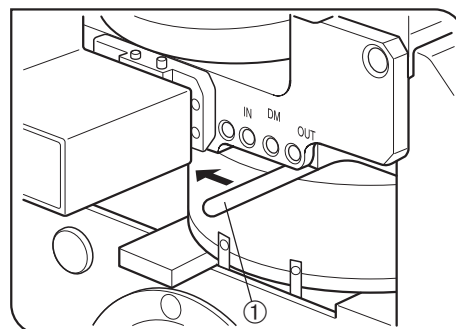
3. Run the "Application Software for PC."

4. Set up the AF (autofocusing) functions.

★ The following parameters should be set to start AF.

- Offset: Setup of the amount of compensation between the glass surface and target position.
- AF zone: AF search zone (AF center, width and range)

☉ For details, refer to the "Application Software."



3 SPECIFICATIONS

Item	Specifications
Applicable microscope frame	IX81-ZDC (Note: Set the magnification changer built into the IX81 to 1X when using the focus drift compensation function.)
Applicable observation tubes	U-BI90-CT, U-BI90, U-TBI90, U-TR30H (in combination with the IX-ATU)
Applicable control box	IX2-UCB (Control board and Application Software for PC (see page 1) are required.)
Controller	PC
Focus detection method	Pupil-division reflection active autofocusing using a laser diode and 2-division detector. Laser diode: 785 nm (Class 1: IEC60825, Class I : CDRH) Laser pulse duration: 0.5 to 2.5 ms. Frequency: 100 Hz. Momentary maximum output power; 100 μ W.
Applicable objectives	Refer to the list of applicable objectives (page 4).
Observation method	Reflected light fluorescence observation is recommended.
Field number	22
AF focusing rate	Less than 1 sec. from the focusing range (0.8 sec. excluding the offset drive time) (Note: The AF rate may slow down depending on the PC setup.)
AF repeatability	Provided that an objective other than the 20X objective is used: $\pm 0.1 \mu\text{m}$ (with constant temperature)/ $\pm 0.3 \mu\text{m}$ (with less than 5°C temperature change) (With the 20X objective, the above figures change to $\pm 0.3 \mu\text{m}/\pm 0.5 \mu\text{m}$ respectively.)
Dimensions & weight	IX81-ZDC : 303 (W) x 313 (H) x 434 (D) mm., 19.2 kg (42.2 lb) (AF sensor unit : 76 (W) x 76 (H) x 201 (D) mm., 0.9 kg (2 lb)) (* Image-forming lens section/excluding the connector)
Operating Environment	<ul style="list-style-type: none"> • Indoor use. • Altitude: Max. 2000 meters. • Ambient temperature: 5° to 40°C (41° to 104°F) • Maximum relative humidity: 80% for temperatures up to 31°C (88°F), decreasing linearly through 70% at 34°C (93°F), 60% at 37°C (99°F), to 50% relative humidity at 40°C (104°F). • Supply voltage fluctuations: $\pm 10\%$. • Pollution degree: 2 (in accordance with IEC60664) • Installation (overvoltage) category: II (in accordance with IEC60664)

4 TROUBLESHOOTING GUIDE

Under certain conditions, performance of the microscope may be adversely affected by factors other than defects. If problems occur, please review the following list and take remedial action as needed.

If you cannot solve the problem after checking the entire list, please contact Olympus for assistance.

Problem	Cause	Remedy	Page
a) Autofocusing is impossible.	The AF port selector lever is not set to IN.	Set it to IN.	7
	Bubbles are present in the oil or water.	Remove the bubbles.	–
	An objective other than specified is in use.	Use another objective or use manual focusing.	4
	The objective is mounted improperly.	Screw in the objective firmly.	–
	The transmitted light bulb generates infrared light.	Close the shutter or turn off the light bulb during AF.	5
	A plastic bottom dish is in use.	Use a glass bottom dish.	3
	Electrical noise is interfering with operation.	Ground the IX2-UCB. Be also sure to ground the ancillary equipment.	1
	The glass bottom dish has a large scratch.	Use another glass bottom dish or use manual focusing.	–
	The microscope is being used in DIC observation.	Adjust the retardation of the differential interference contrast to near a gray sensitive color.	5
	Water (or oil) of water-immersion (or oil-immersion) objective is dried.	Supply oil.	–
	A water-immersion objective is used and the interface between the water and glass bottom dish is focused.	Set the AF zone so that its center is located on the interface between the culture fluid and glass bottom dish and the AF zone width is $\pm 10 \mu\text{m}$.	7
b) Autofocusing takes long time or fails.	AF is set improperly.	Set up AF properly. (For the setting method, refer to the instruction manual for the application software.)	7
	Electrical noise is interfering with operation.	Ground the IX2-UCB. Be also sure to ground the ancillary equipment.	1
	The microscope is vibrating.	Reduce the microscope vibration by using an anti-vibration bench, etc.	–
	The specimen is vibrating.	Lock the specimen firmly. Also confirm that the stage center plate is free of abnormality.	–
c) AF is applied to a position deviated from the target position.	The offset value is input improperly.	Enter the correct offset value. (For the setting method, refer to the instruction manual for the application software.)	7
	Water (or oil) of water-immersion (or oil-immersion) objective is dried.	Supply oil.	–
	A water-immersion objective is used and the interface between the water and glass bottom dish is focused.	Set the AF zone so that its center is located on the interface between the culture fluid and glass bottom dish and the AF zone width is $\pm 10 \mu\text{m}$.	7
d) The specimen moves in the X-Y directions during autofocusing.	The specimen is not locked.	Lock the specimen by placing a weight on the glass bottom dish, for example. Also confirm that the stage center plate is free of abnormality.	–

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