ThetaStation

User Guide

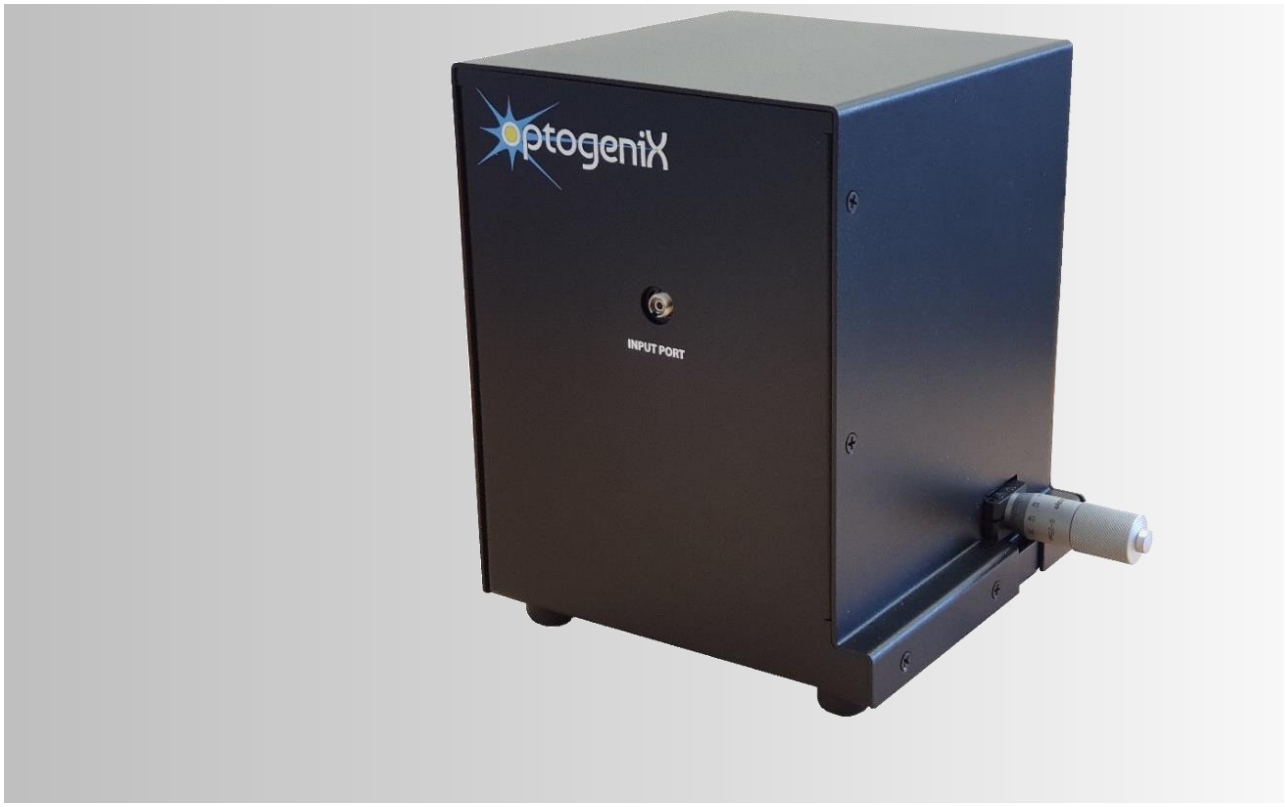


Table of Contents

Chapter 1 Warning Symbol Definition

Below is a list of warning symbols you may encounter in this manual or on your device.

NOTICE
Information considered important but not hazard related

CAUTION
Instructions for use that, if disregarded, might result in product damage.

WARNING
Instructions for use that, if disregarded, might result in personal injury or death

Caution: Risk of Electric Shock

Warning: Laser Radiation

Chapter 2 Safety

NOTICE

All statements regarding safety of operation and technical data in this instruction manual will only apply when the unit is operated correctly.

WARNING

Do not open housing. Do not operate without cover installed.

WARNING

Do not operate in wet or damp conditions.

WARNING

Light source must be turned off before connecting it to ThetaStation.

.

Chapter 3 Description

ThetaStation is an opto-mechanical tool designed to perform **site-selective light delivery** with OptogeniX tapered fibers[1]. ThetaStation can be used with any visible light source. Selection of the emitting sub-region of a Lambda fiber (or the active optical window of a Sigma fiber) is achieved by manual operation of a micrometric screw.

ThetaStation can also be used in combination with OptogeniX **Magic Box** to perform **site-selective fiber photometry**.

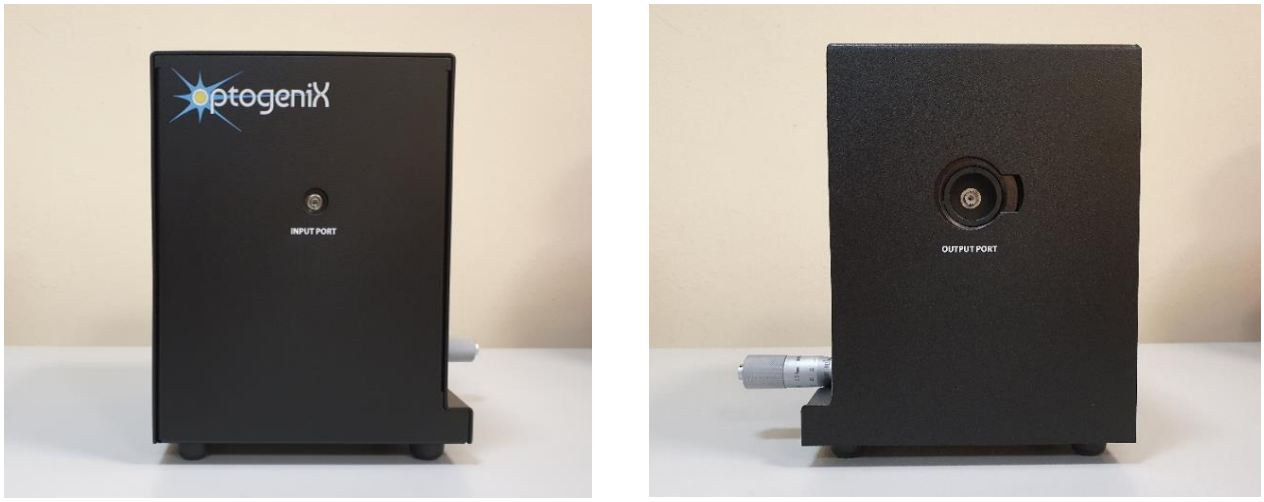


Figure 1 ThetaStation front view with *Input Port* (left) and rear view with *Output Port* (right)

[1] https://www.optogenix.com/applications/

Chapter 4 Configuration Guide

4.1 Definitions and abbreviations

NA – numerical aperture

Input Patch cord – Patch cord connecting the light source to the ThetaStation Input Port.

Input Power – Optical power at ThetaStation input (measured at the output of the input patch cord connected to light source).

Output Patch Cord - Patch cord connecting the tapered fiber to the ThetaStation output port

Output Fiber – Common fiber type (NA/core size) for both the output patch cord and the tapered fiber.

Output Power – Optical power at the output of the output patch cord.

Configuration – A specific combination of light source, input patch cord (NA/core size), output fiber (NA/core size), and rotary joint (if employed).

Transmission efficiency – Ratio between the output and the input power.

Working range – Range of positions of the micrometric screw within which site-selective operation is effective.

4.2 Use with Lambda fibers

The full set up for site-selective light delivery with ThetaStation and a generic light source is shown in **Figure 3**.

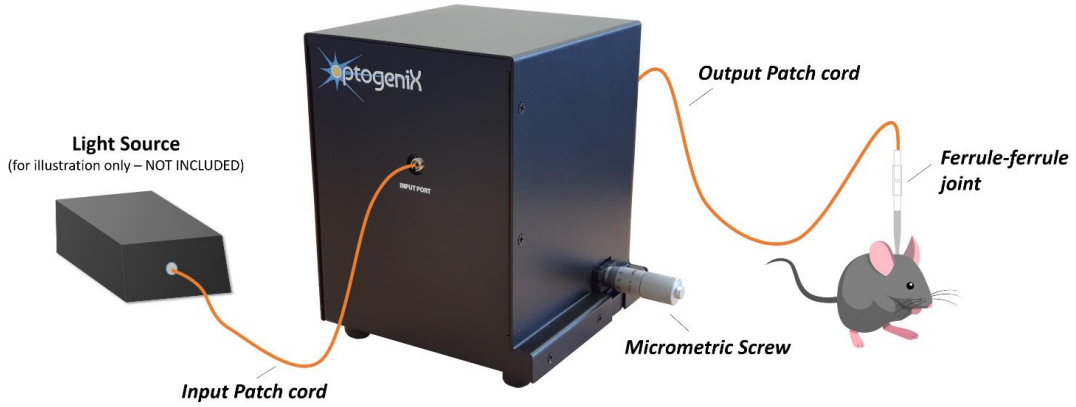


Figure 3 Connection scheme

LASER sources are recommended for Optogenetic stimulation. Transmission efficiency decreases with the position of the micrometric screw from 0mm to end of working range[2]. Values of maximum transmission efficiency achieved with a specific laser source and *recommended input patch cord and output fiber types* (NA/core size) are reported in TABLE 1 and can be used for estimating the transmission efficiency achievable with other laser sources with wavelength in the 400 - 490nm interval.

Source Type	***Input Patch Cord***	***Output Fiber***	***Transmission Efficiency (0mm)***	***Input Power for Optogenetic Stimulation***
LASER (Cobolt 06-MLD 473nm)	Single mode (SM)	.22NA/105µm	1%	30mW
		.39NA/200µm	5%	6mW
	Multimode (MM) .22NA/50µm	.22NA/105µm	0.8%	40mW
		.39NA/200µm	4.5%	7mW

TABLE I

The last column in TABLE I reports for each listed configuration the input power required for 1mW/mm2 emission out of a Lambda fiber having the

[2] Minimum value of transmission efficiency (typically 70% of maximum) depends on the actual configuration.

same NA/core size of the output fiber, and at 0mm position of the micrometric screw[3].

General recommendations

- Use a Single Mode (SM) input patch cord for best transmission efficiency;
- The use of a low NA/small core input patch cord gives acceptable transmission efficiency;
- Transmission efficiency increases with both NA and core size of the output fiber. Therefore, *small diameter tapered fibers should be preferred only for their reduced invasiveness*.

N.B.: *Site-selective operation is compatible with the use of a* ***rotary joint****.*

4.3 Use with .66NA Lambda fibers

With ThetaStation is possible to operate site-selective light delivery with Lambda fibers based on Plexon high efficiency .66NA fiber[4]. Values of transmission efficiency listed in TABLE I for the case of the .39NA fiber can be used as a reference for a .66NA output fiber type.

[3] Assuming 15% power loss at the ferrule-ferrule joint. Calculated from M. Pisanello et al., Scientific Reports (2018)8:4467

[4] Available at https://plexon.com/products/lambda-fibers/

Chapter 5 Setup and Operation

5.1 Connection Guide

1) Unbox the ThetaStation and remove all packaging material.
2) Connect all the setup components as in **Figure 3** of **Chapter 4**.

***N.B.**: Connection of the output patch cord is easier when the micrometric screw is at 0mm position.*

***N.B.**: For FC port type, to ensure a good connection pay attention that the connector key is oriented to enter within the receptacle slot.*

WARNING
The output port and attached patch cord connector move with the rotation of the micrometric screw. Do not touch them during operation.

5.2 Calibration

Position and size of light emitting sub-regions of Lambda fibers (or windows emission properties of Sigma fibers), and transmission efficiency need to be calibrated as a function of the micrometer screw position for:

- every different configuration
- every different-active-length Lambda fiber or different geometry Sigma fiber

Sub-regions position and size (Lambda Fibers)

Immerse the tapered fiber in a solution of fluorophore suitable for the wavelength in use and place it under a microscope equipped with the corresponding emission filter[5] (**Figure 4**).

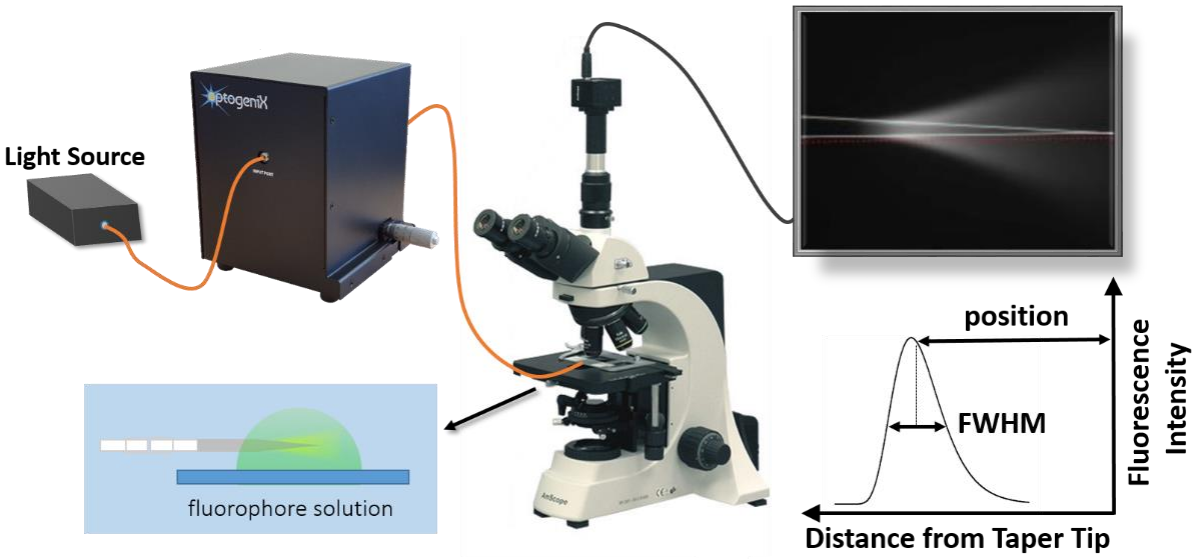


Figure 4 Characterization of the emitting profiles of a Lambda fiber.

With a microscope camera, image the fluorescence emission as a function of micrometric screw position. With the help of an image analysis software[6], for each collected image take the intensity profile along a line close to the taper edge. The sub-region width can be estimated as the FWHM of the intensity profile. The distance from the taper tip of the FWHM middle point is then conventionally defined as the sub-region position.

[5] e.g. PBS:fluorescein solution and FITC filter for blue light (450nm-480nm).
[6] e.g. *ImageJ* (free software)

NOTICE

Sub-regions definition is part of the design of the experiment. As the active sub-region varies continously with the position of the micrometric screw, the number and position of the active sub-regions are defined arbitrary by the end-user during calibration.

Optical Windows activation and emission profiles (Sigma Fibers)

Light emission properties of Sigma fibers as a function of the micrometric screw position can be verified with the same method described in the previous paragraph.

Transmission Efficiency

Transmission efficiency as a function of the micrometric screw position can be easily collected with a light intensity sensor (and related power meter) of the appropriate wavelenght range, as shown in **Figure 5**.

Figure 5 Setup for optical power calibration

5.3 Operation

Use with Lambda Fibers

Select an emitting sub-portion by setting the micrometric screw at the calibrated position.

For each position, set the light source power to achieve the desired sub-region output power as:

output power = (input power) x (transmission efficiency) x (F-F joint efficiency)

where *F-F joint efficiency* accounts for the power loss in the ferrule-ferrule joint between the output patch cord and the tapered fiber. The exact value of the *F-F joint efficiency* depends on the status of connectors (facet cleanliness and smoothness) and should be measured for any tapered fiber stub/output fiber combination. Assuming 85% as average value gives a good estimation of the output power.

Use with Sigma Fibers

Select the active optical window by setting the micrometric screw at the calibrated position.

The optical power emitted by the active window for a given position of the micrometric screw must be directly measured with a power meter as a function of the light source power **before fiber implantation.**

Chapter 6 Accessories

6.1 Patch cords

We recommend the use of OptogeniX patch cords for site-selective light delivery with .22NA or .39NA tapered fibers.

.66NA patch cords for use with .66NA Lambda fibers are provided by Plexon[7].

6.2 Calibration slides

Calibration slides are provided for easy collection of the light emission profiles of tapered fibers. Calibration slides consist of a specified tapered fiber stub attached on a microscope slide. The surface of the slide close to the fiber is covered with hydrophobic material to help formation of the droplet of fluorophore solution.

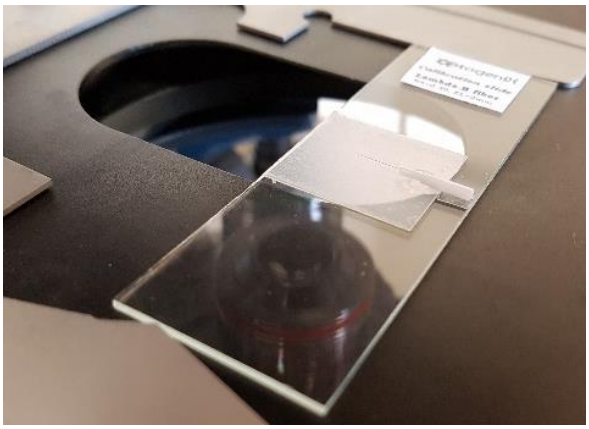


Figure 6 Calibration slide

[7] www.plexon.com

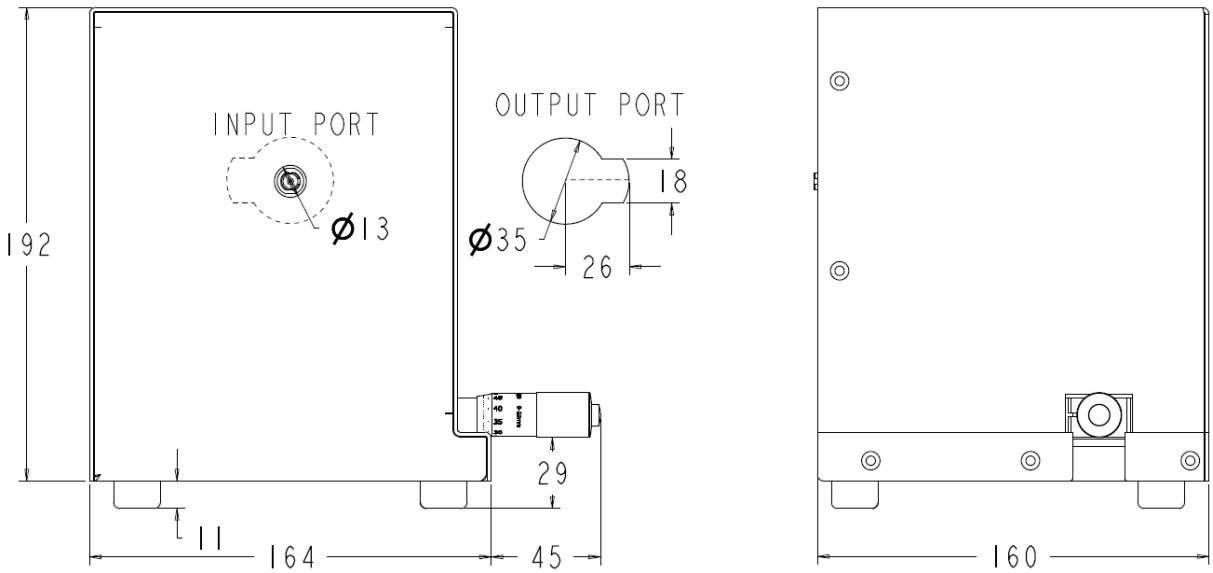
Chapter 7 Specifications

Weight (kg):	5
Enclosure material:	Aluminum 5052-H32
Input port:	FC/PC
Output port:	FC/PC or SMA
Working range (105/.22NA output fiber)	0-8mm
Working range (200/.39NA output fiber)	0-14mm
IEC protection class:	Classe III
Max Input Optical Power	500mW

Chapter 8 Troubleshooting

Problem	Solutions
Irregular light emission from the tapered fiber (e.g. contemporary light emission from two or more sub-regions of a Lambda fiber)	Output Patch Cord can be defective. Verify light emission from the Output Patch Cord by directing it toward a flat, non-reflecting object (like a piece of paper). Signature of regular emission as a function of the micrometric screw position is a filled circle at 0mm that transforms in increasing diameter rings until the end of the working range. Examples of irregular light emission: - separate rings emitted at the same position of the micrometric screw; - rings emitted independently from the micrometric screw position.
Transmission efficiency is significantly lower than the typical value reported in TABLE I for the same configuration.	Check patch cords connection to the device. Check for any abnormal operation of the light source. If the problem persists contact OptogeniX.

Chapter 9 Mechanical Drawing



N.B.: All dimensions in millimeters.

Chapter 10 Cleaning and Maintenance

ThetaStation can be cleaned using a soft cloth. If needed, the cloth can be dampened with some isopropyl alcohol.

DANGER

Disconnect the system from accessories and from the light source before any cleaning and maintenance operation.

Chapter 11 EU Declaration of Conformity

CE *EU Declaration of Conformity*

OptogeniX S.r.l.
Via Barsanti c/o Istituto Italiano di Tecnologia
73010, Arnesano (LE) – ITALIA
VAT n°: IT04644390751

This declaration of conformity is issued for:

Opto-mechanical instrument model name/number	**ThetaStation**
Serial Number of ThetaStation covered by this EU DoC	YYMMDDTS1XX (YY = year, MM = month DD = day, TS1 = product code, XX = serial number)

We, Optogenix Srl, declare under our sole responsibility that the above named product conform to the essential requirements of the following European Union directives and normative documents:

- Direttive 2014/35/UE, Low Voltage (LVD)
- Directive 2014/30/UE, Electromagnetic Compatibility (EMCD)
- Direttiva 2011/65/UE, 2015/863/UE **R**estriction **o**f the use of certain **H**azardous **S**ubstances
- Direttiva 2001/95/CE, General Product Safety
- Directive 2012/19/UE, "RAEE"
- IEC 61511-1:2016 Functional safety - Safety instrumented systems for the process industry sector - Part 1: Framework, definitions, system, hardware and application programming requirements
- IEC 61882:2016 risk analysis by the HAZOP method

Issued On: April 1rst, 2020

Leonardo Sileo
Sole administrator

Chapter 12 Warranty

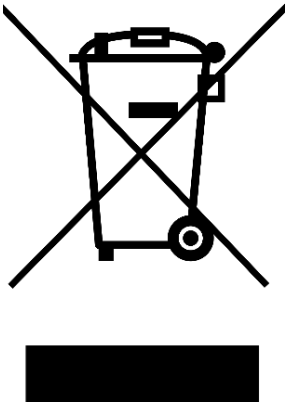
ThetaStation (the Product) is guaranteed to perform per advertised specifications and is covered against material, manufacturing or design defects for two (2) years following the date of delivery to buyer. If, prior to the expiration of the Warranty Period, the Buyer informs OptogeniX in writing of any breach of this limited warranty, then OptogeniX may repair or replace the Product that gave rise to the breach or, in OptogeniX's sole and exclusive discretion, refund the amounts that Buyer paid for the Product.

Buyer will bear the costs of access, de-installation, re-installation and transportation of the Product to OptogeniX and back to Buyer. Any repair or replacement pursuant to this limited warranty will not extend the Warranty Period. OptogeniX does not warrant the Product, or any repaired or replacement parts, against normal wear and tear or corrosion. This limited warranty and remedy are expressly conditioned upon: (i) Buyer's payment of the purchase price in full, (ii) Buyer giving written notice of the defect, reasonably described, to OptogeniX within ten (10) days of the time when Buyer discovers or ought to have discovered the defect, (iii) the storage, installation, operation, use, and maintenance of the Product in compliance with the Instructions, (iv) the existence of proper records of Buyer's operation and maintenance of the Product during the Warranty Period, (v) Buyer providing OptogeniX with a reasonable opportunity to examine the Product and the aforementioned records, and (vi) the absence of any unauthorized modification or repair of the Product, including without limitation the removal or alteration of any serial numbers or warranty date decals.

Before any test may be used to evaluate the Products, Buyer will: (i) provide OptogeniX with reasonable written notification of the test, (ii) allow OptogeniX to be present during the test, and (iii) receive OptogeniX's consent to the conditions of the test, which consent will not be unreasonably withheld. If a test is performed on the Products, and OptogeniX has not consented to the conditions of the test, then this limited warranty will be void.

Chapter 13 Regulatory

As required by the (Waste Electrical and Electronic Equipment Directive 2012/19/UE) of the European Community (EC) and the corresponding national laws, OptogeniX offers all end users in the EC the possibility to return “end of life” units without incurring disposal charges.



This offer is valid for OptogeniX equipment:

- Sold after June 26, 2020
- Marked correspondingly with the crossed out “wheelie bin” logo (see right)
- Sold to a company or institute within the EC
- Currently owned by a company or institute within the EC
- Still complete, not disassembled and not contaminated

If you wish to return an OptogeniX unit for waste recovery, please contact OptogeniX or your nearest dealer for further information.

Waste Treatment

If you do not return an “end of life” unit to OptogeniX, you must hand it to a company specialized in waste recovery. Do not dispose of the unit in a litter bin or at a public waste disposal site.

Chapter 14 OptogeniX Contacts

For technical support or sales inquiries, please write us at info@optogenix.com or visit www.optogenix.com.