

User-Manual

UVE

System for Photochemical Derivatization



Dear Sir or Madam,

You have decided in favor of UVE, the high-performance photochemical derivatization system.

Thank you very much for your confidence! We will do anything we can to keep you satisfied with the system in the long run.

The working principle is easy and effective - you needn't prepare derivatization solutions any more.

The system is designed especially robust for daily operation in laboratories; it is user-friendly, flexible and well-priced.

The UVE thus offers you a range of advantages and facilitations, which will make routine analysis noticeably easier for you.

The LCTech team wishes you all the best!



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P.S. Do you want to let us know changes or suggestions to this manual?
Please feel free to contact us directly via e-mail to service@LCTech.de.

Content

I) Important Safety Instructions	4
II) Operating UVE.....	8
1. Analysis of Aflatoxins with UVE	9
1.1 General Information	9
1.2 Method Description.....	10
1.3 HPLC-Parameter	11
2. Exchanging UV Lamp and Reactor Coil	12
2.1 Exchanging UV Lamp.....	12
2.2 Exchanging Reactor Coil	13
3. Troubleshooting	14
4. Ordering information	15
5. Notice	16

I) Important Safety Instructions

DANGER

Immediate danger! Death or severe injuries as well as damage to equipment and environment may occur.

WARNING

Warning, possible danger! Severe injuries as well as damage to equipment and environment may occur.

CAUTION

Caution, possible danger! Slight injuries as well as damage to equipment and environment may occur.

NOTICE

Note! Disregarding of notes may cause damage to the product.



Isolate equipment from mains before removing the cover.



After end of lifetime please properly dispose of the device.

Do not dispose to domestic waste.

NOTICE**General Information**

Read and comply with this manual. Take care of the safety instructions. They are the very precondition for no person being exposed to danger and for the correct use of the device.

Abuse of the system and use for other purposes than those originally intended, as well as inappropriate repairs by non-authorized service providers and the use of other than the original parts and accessories negate any possible claims against the manufacturer.

Remove all packing material, inspect the product for completeness and damages, and retain all delivered parts.

WARNING**Intended Use**

The system and all system parts are not to be used on humans or animals.

Prevent any part of the human body from coming into contact with UV radiation.

Make sure that the individual components are only connected, combined and operated according to their design and as indicated in the instructions for use.

CAUTION

UVE is designed for ambient temperatures during operation between +10 °C and +40°C. Check the maximum temperatures if installing UVE in a cabinet or a housing and make sure ventilation is adequate. Make sure that the maximum temperature is not exceeded. Install an external automatic ventilation system if necessary.

NOTICE

UVE and all system parts may only be used for the intended purpose, i.e. for derivatization in devices determined for this purpose.

DANGER**Setting up and Installing UVE**

Equipment shall be connected to a suitable fused and protected electrical supply and a suitable earth point.

Avoid strong over pressure. Risk of bursting!

Maximum permissible pressure 21 bar / 300 psi.

CAUTION

Comply with maximum permissible pressures at inlet and outlet.

Check that mains voltage and current conform to the equipment.

Connect tight at inlet and outlet of UVE.

NOTICE

Provide a firm level platform for the equipment and check that the system to be in a mechanically stable position and that all fittings are secure. Ensure a stable position without any mechanical contact except of the feet. Comply with all applicable safety regulations. Clean dirty ventilation slots in order to avoid any restriction of the air supply; this is particularly important for systems with an installed fan (P/N 10742)

If the equipment is brought from cold environment into a room of operation, allow the equipment to warm up (pay attention to water condensation on cold surface).

Comply with all applicable and relevant safety requirements (regulations and guidelines), implement the required actions and adopt suitable safety measures.

NOTICE**Ambient Conditions**

To the best of our knowledge the equipment is in compliance with the requirements of the applicable EC-directives and harmonized standard with regard to design, type and model. Directive IEC 1010 gives in detail conditions,

under which the equipment can be operated safely (see also IP degree of protection).

Adopt suitable measures in case of differences, e.g. using the equipment outdoors, conductive pollution or bedewing.

Pay attention to the permissible maximum ambient and internal temperatures.

DANGER

Operating Conditions for UVE

UVE is not suitable for being operated in explosive areas.

UVE is not suitable for

- - unstable substances
- - substances which react explosively under impact (mechanical stress) and/or when being exposed to elevated temperatures without air
- - self inflammable substances
- - explosive substances

The user must take suitable precautions for a breakdown or dysfunction. A breakdown of UVE (e.g. by power failure) and connected components, a breakdown of supply parts or changed parameters must never lead to a dangerous situation.

Attention: If ON is preselected the process runs immediately after a power failure without pressing any further key. It is the user's responsibility to ensure that no dangerous status of the system due to the automatic start-up can occur and to provide appropriate safety measures.

WARNING

Starting UVE must not lead to a dangerous situation under any circumstances.

Avoid plugging of the tube by disposal.

Check tightness of the tube connections regularly and mend if necessary.

Avoid leakage of solvents.

The user must take care that the system can be lead to a safe status even in case of a mistake.

Pay attention to the interactions and chemical reactions of the media.

Current-carrying parts, such as the power cord, must never be brought into contact with water. As with any other mains-operated device, danger of short-circuits and thus for life exists.

The system may only be operated with alternating current.

The voltage of the local mains supply must correspond with the specifications detailed on the system label.

Do not use a defective cable for the operation of the system. The mains lead and all possible extension cables must be fitted with sealed contacts and possess a minimum cross-section of 1 mm².

DANGER

The casing of the mains lead should be in accordance with all relevant requirements for external, current-carrying cables in the laboratory environment.

DANGER

Safety during Operation

Adopt suitable measures to prevent the release of dangerous, toxic, explosive, corrosive, noxious or polluting fluids, vapors and gases. In case install an

CAUTION

appropriate collecting and disposal system and take protective action for UVE and environment.

The user must take suitable precautions to prevent any formation of explosive mixtures in the expansion chamber or at the outlet. Mechanically generated sparks, hot surfaces or static electricity may ignite these mixtures.

The system should not be operated without flow of fluid over a longer period of time. Risk of overheating!

Use only genuine spare parts and accessories. Otherwise safety and performance of the equipment as well as the electromagnetic compatibility of the equipment might be reduced.

UV irradiation is dangerous. Never work with unprotected eyes and skin close to an illuminated UV lamp.

NOTICE**Maintenance and Repair**

Wear parts have to be replaced if necessary.

Isolate equipment from mains before maintenance.

Never operate the system with opened housing. Ensure that the system cannot be operated accidentally. Never operate a defective or damaged UVE.



Attention: The system might be contaminated with hazardous or otherwise dangerous substances. If necessary decontaminate before contact.

WARNING

Ensure that maintenance is done only by suitably trained and supervised technicians. Ensure that the maintenance technician is familiar with the safety procedures. A system returned to the manufacturer can be repaired only when certain procedures (safety, environmental protection, decontamination) are followed.

II) Operating UVE

UVE is delivered ready-to-use. Unpack the device and place it between your HPLC system and the fluorescence detector.

Ensure that the HPLC flow path is cleaned and excessively flushed with acetonitrile/water or methanol/water before connection. Impurities from other derivatization reagents may affect the performance of the device.

Afterwards connect one of the two 1/16" capillaries with the column outlet, the other one with the detector inlet. The two 1/16" capillaries are arbitrarily interchangeable regarding the connection. If an elongation to the detector is needed or if the UVE is directly connected to the detector cell, we recommend to use a PEEK capillary to avoid that scattered light can get into the detector cell.

The soft rubber tube is the drain of the reflection tube of the UV lamp. Only in case of a leakage fluidity can leak from this tube. As this is a safety device, it is to be controlled for leaking fluidity from time to time.

In the UVE a pressure of 21 bar / 300 psi mustn't be exceeded in the reactor coil!

Switch on the HPLC system and the UVE now; the power switch is on the front side of the device and shows green light when switched on. To ensure a proper equilibration of the system, please wait another 15 minutes before operation.

In particular cases the use of the HPLC eluent has to be checked regarding the stability due to photochemically induced reactions (e.g. the generation of radicals.)

In some cases it might make sense to connect a cooling loop (e.g. 2 m 1/16" PTFE-tube) between UVE and the detector.

Technische Daten

Power supply	P/N 10519: 220 - 265 V, 50/60 Hz; P/N 10742: 90 - 126 V, 50/60 Hz
Power input	100 W
Measurements (B x H x T)	145 x 100 x 275 mm
Weight	3 kg
UV-lamp	254 nm
Reactor coil	1 mL
CE-certified	

1. Analysis of Aflatoxins with UVE

1.1 General Information

Aflatoxins are naturally occurring toxins (mycotoxins = fungal toxins), which are highly toxic for humans and mammals. The aflatoxins are produced by molds (e. g. *Aspergillus flavus*). They are highly cancer-causing, especially in the liver.

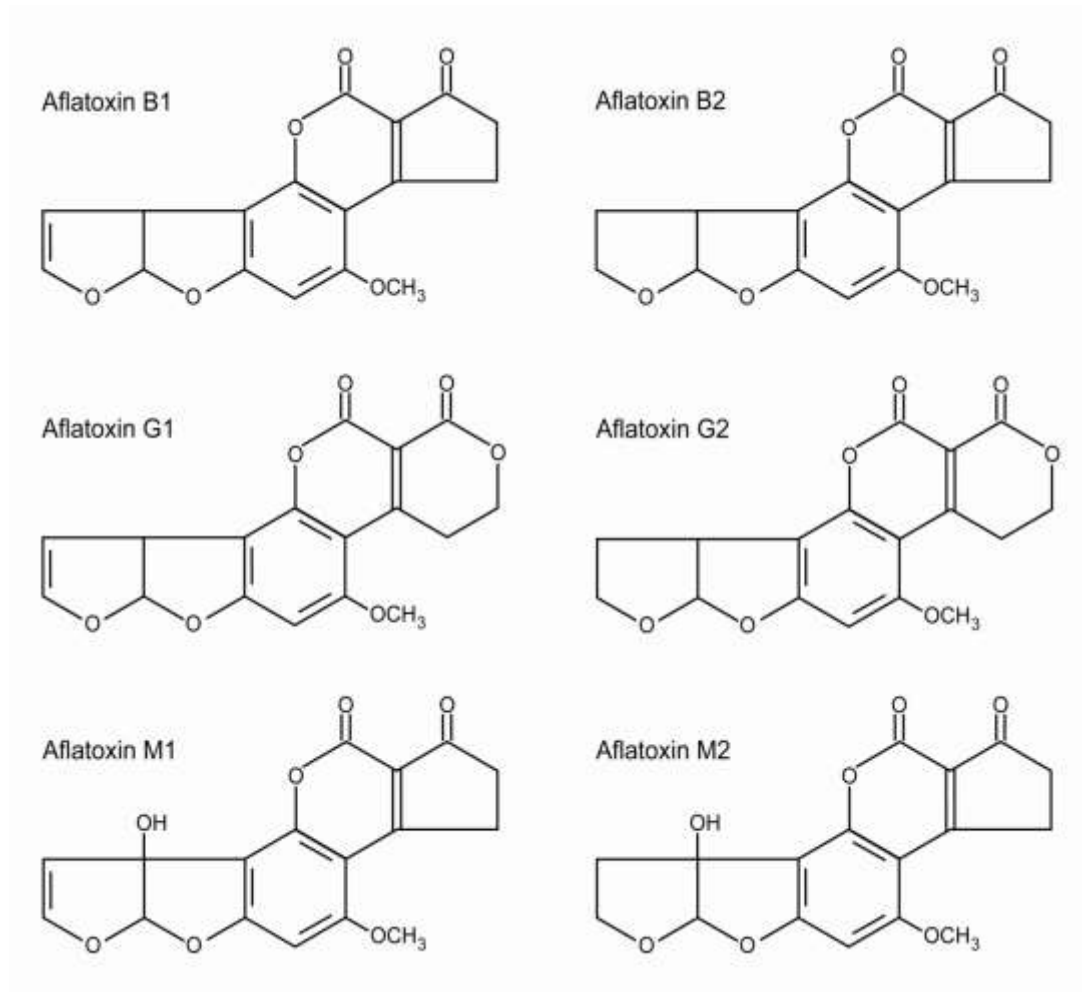


Figure 1 Structure of aflatoxins

Aflatoxins are found in nuts, figs, dates, corn, dried paprika, spices and many other vegetable foodstuffs. The aflatoxins M1 and M2 can be found in the milk of cows fed with contaminated feed. They are the metabolized aflatoxins B1 and B2 (14-hydroxylated). The abbreviations of an aflatoxin consist of the color of the fluorescence (blue or green) or the occurrence (milk) and its relative chromatographic mobility (1 or 2). Due to their high toxicity, the maximum contents in food are regulated world-wide.

1.2 Method Description

Depending on the matrix, the aflatoxins are extracted from the corresponding matrix, cleaned up with immunoaffinity columns (e.g. LCTech AflaCLEAN) and afterwards measured with HPLC and fluorescence detection.

Since only the aflatoxins B2 and G2 show natural fluorescence, B1 and G1 must be derivatized prior to detection. This can be done photochemically using the UVE by irradiation with UV light at 254 nm. The aflatoxins B1 and G1 are thus hydroxylated, leading to stable and measureable fluorescence. The chemical or measurement-related properties of the other aflatoxins (B2, G2, M1, M2) are not changed by this step.

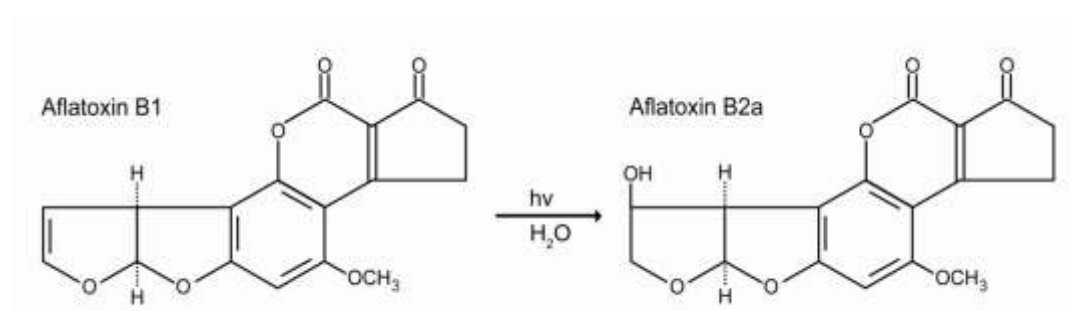


Figure 2 Mechanism of photochemical derivatization

The comparability of this method and the electrochemical derivatization was shown. (A. Papadopoulou-Bouraoui, J. Stroka, E. Anklam, J. AOAC Int. Vol. 85, No. 2, 2002, 411 - 416 as well as AOAC Official Method 2005.08).

A validation study according to EU directions was performed by Muscarella et al. (Muscarella, Marilena, Iammarino, Marco, Nardiello, Donatella, Lo Magro, Sonia, Palermo, Carmen, Centonze, Diego and Palermo, Domenico (2009) 'Validation of a confirmatory analytical method for the determination of aflatoxins B1, B2, G1 and G2 in foods and feed materials by HPLC with on-line photochemical derivatization and fluorescence detection', Food Additives & Contaminants: Part A, 26:10, 1402 – 1410)

The achievable limits of detection and quantitation mainly depend on the performance of the used fluorescence detector and do not depend on the derivatization.

1.3 HPLC-Parameter

HPLC-column	150 x 4.6 mm with guard 8 x 4.6 mm; C-18; 3 μ M
Eluent	Water / acetonitrile / methanol (HPLC grade) (60 / 15 / 30 // v / v / v) isocratic <i>suitable for fluorescence analyses is highly recommended</i>
Flow rate	1,2 mL/min
Column temp.	36 °C
Injection volume	10 – 100 μ L
λ – Excitation	365 nm
λ - Emission	460 nm

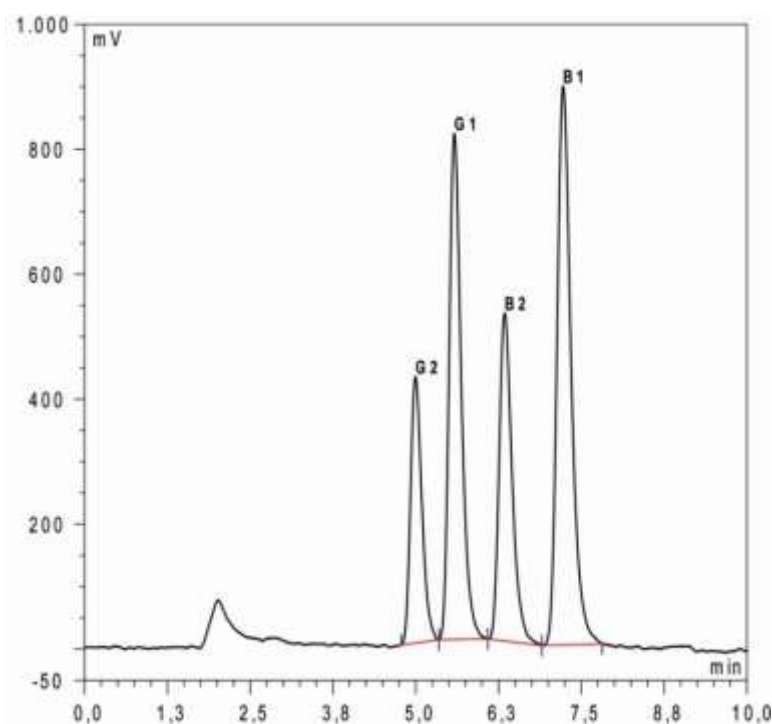


Figure 3 Chromatogram of the aflatoxins G2, G1, B2 and B1 (left to right) with photochemical derivatization, e.g. dates, spiked with 10 ng aflatoxin

Concentrations in origin standard solution:

B1, G1 2 ng each and B2, G2 0.5 ng each

Injection volume: 60 μ L

Absolute concentrations:

0,06 ng G1, B1

0,0015 ng G2, B2

The special geometry of the reactor coil allows the elution of aflatoxin B1 already after at about 8 minutes without noteworthy band broadening.

2. Exchanging UV Lamp and Reactor Coil

As a rule, both UV lamp and reactor coil can be operated without any loss of performance for a long time. In case you nevertheless have to exchange them, please proceed as follows:

Caution! Never operate the UV lamp with open housing!

2.1 Exchanging UV Lamp

- Switch off the HPLC system and the device and unplug the UVE from electricity.
- Unscrew the screws of the housing and take it off.
- Please wear laboratory gloves for all further steps in order to avoid any hand contact with the glass surface of the UV lamp.
- Remove the faceplate at the reflection tube by loosening the two hexagon sockets. Pay attention to the tubes while removing the faceplate! Do not squeeze them!
- Grip the UV lamp with thumb and index finger and straightly pull it out of the reflection tube.
- In order to exchange the UV lamp carefully remove the reactor coil from the lamp.
- It is not necessary to disconnect the fittings.
- Dispose the defective lamp as hazardous waste.
- Now push back the reactor coil onto the UV lamp. Straightly insert the new lamp until it tangibly locks.
- Embed the faceplate of the reflection tube and tighten it with the screws. The two tubes must be led through the vertical slot and must not be squeezed!
- Attach the housing and plug the system in electricity.
- Your device is again ready to operate.

2.2 Exchanging Reactor Coil

- Switch off the HPLC system and the device and unplug the UVE from electricity.
- Unscrew the screws of the housing and take it off.
- Please wear laboratory gloves for all further steps in order to avoid any hand contact with the glass surface of the UV lamp.
- Remove the faceplate at the reflection tube by loosening the two hexagon sockets. Pay attention to the tubes while removing the faceplate!
- Loosen the fittings of the reactor screw.
- Grip the UV lamp with thumb and index finger and straightly pull it out of the reflection tube.
- Carefully remove the reactor coil from the lamp.
- Dispose of reactor coil as hazardous waste.
- Now push back the new reactor coil onto the UV lamp.
- Straightly insert the new lamp until it tangibly locks.
- Connect the new reactor coil by using the fittings.
- Embed the faceplate of the reflection tube and tighten it with the screws.
- The two tubes must be led through the vertical slot and must not be squeezed!
- Attach the housing and plug the system.
- Your device is again ready to operate.

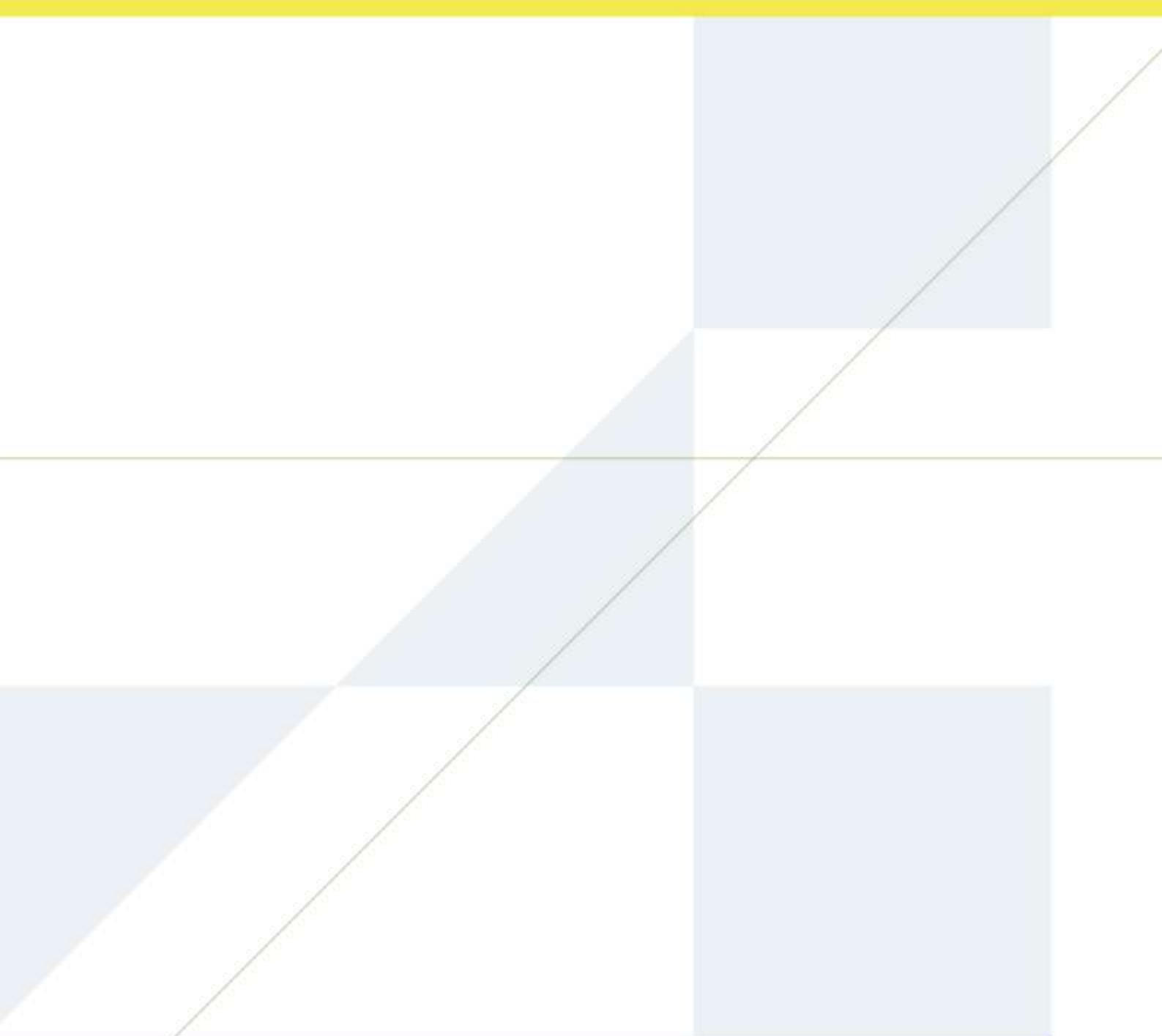
3. Troubleshooting

Problem	Reason	Action
UV lamp is off	No power supply	Check power plug and mains supply
	Defective UV lamp	Exchange
Fluidity is leaking from drain tube	Defective reactor coil	Exchange
	Defective HPLC system	Check
	Degraded standard	Check
	Aged reactor coil	Exchange
Signal intensity decreases	Aged UV lamp	Exchange
	Interfering substances in mobile phase	Wash and condition with methanol. Change solvents to higher quality. Use HPLC grade, water from water purifiers e.g. MilliQ) is not recommended. Methanol (HPLC quality) should be suitable for fluorescence analysis.

4. Ordering information

Description	P / N
UVE, photochemical reactor for aflatoxin analysis, 254 nm, 1mL reactor volume, 220 - 265 V	10519
UVE, photochemical reactor for aflatoxin analysis, 254 nm, 1mL reactor volume, 90 - 126 V	10742
UV replacement lamp	10563
Reactor coil, 1.0 mL	10520
HPLC column for aflatoxin analysis, RP C18	10522
Guard cartridge holder	10750
Guard cartridges, RP C18, 3 pcs.	10523
AflaCLEAN, aflatoxin, 3 mL, widebore immunoaffinity column, 25 columns/pack	10514
AflaCLEAN, aflatoxin, 3 mL, widebore immunoaffinity column, 500 columns/pack	11721
AflaCLEAN Select, aflatoxin, 3 mL, widebore immunoaffinity column, 25 columns/pack	12058
AflaCLEAN Select, aflatoxin, 3 mL, widebore immunoaffinity column, 500 columns/pack	12059
AflaCLEAN Select, aflatoxin, 1 mL, widebore immunoaffinity column, 25 columns/pack	12062
AflaCLEAN Select, aflatoxin, 1 mL, widebore immunoaffinity column, 500 columns/pack	12063

5. Notice



Any Questions?
Do not hesitate to contact us: