



## NightSHADE evo LB 985N In vivo Imaging System with indiGO Software

Operating Manual 73037BA2

Rev. No.: 00, 09/2022



#### Not for use in in-vitro diagnostic (IVD) procedures.

The information in this guide is subject to change without notice.

DISCLAIMER

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This instrument is not designed or intended for use with installations or equipment in hazardous environments. Servicing of the instrument must only be performed by Berthold Technologies Field Service Engineers or service staff authorized by Berthold Technologies.

Please contact our Service Center at <u>service@berthold.com</u> if you have any operational issues.

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#### **Prefatory Comments** 1

## 1.1 History of Changes

Revision	Date	Chapter / Page	Change	Author
Rev. 00	09/2022		Initial release	R. Kembügler

## **1.2 Typographical conventions**

The following typographical conventions will be used in this operating manual:



Caution! Danger!

**⊠** Important information

	Example
Menus and options in the software are printed in bold type	Measurement, View
The <b>selection of a menu item</b> is written as a sequence of	
commands, separated by a vertical line	Device   Configuration
The names of dialog boxes and parameters are also	
printed in bold type	Filter Setting
Buttons are printed in bold type inside angular brackets	<start>, <stop></stop></start>
Enumerations are emphasized by	$\boxtimes$
Action <b>steps</b> are identified by	$\boxtimes$

## 1.3 Warning signs at the instrument

	<b>Wear eye protection</b> When using the LED panel an eye protection (DIN EN 166) is required to prevent from high intensity light caused by the LED lights.
	<b>Caution Danger</b> This sign indicates a general danger (e.g. switched power socket inside the instrument).
4	<b>Dangerous electrical voltage</b> The switched power socket inside the instrument carries high voltage. Be careful when handling this device.
<u>SSS</u>	<b>Hot surface</b> The excitation light source can become hot. Be careful when handling these device.

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## 2 Safety Instructions



**Caution!** This sign alerts you to important operating procedures with a potential danger of damaging the equipment and endangering your safety on disobeying. Refer to the user and instrument manuals for precautionary instructions.

This operating manual includes information and warnings that have to be observed by the user in order to ensure safe operation of the instruments.

Please do always act according to the following safety instructions, before as well as during operation of the system! Before set up and operation of the instrument it is necessary to read the instructions below as neither safe operation of the instrument nor safety of the user are guaranteed otherwise. Failure to follow the instructions may invalidate the warranty.

The instrument has been manufactured in accordance with the safety requirements for electronic and medical measuring systems. If the law lays down regulations on the installation and/or operation of sample measuring system, then it is the operator's responsibility to adhere to them.

The instrument is designed in a way that no harmful noise levels are emitted by the instrument itself. If the user detects unusual noise from the instrument the service must be contacted.

The manufacturer has done everything possible to guarantee that the equipment functions safely, both electrically and mechanically. The user has to make sure that the instrument will be set up and installed properly to guarantee safe operation.

The instruments are tested by the manufacturer and supplied in a condition that allows safe and reliable operation. Improper use of the instrument can disable safety features and could lead to an unsafe operation of the instrument

- □ This equipment must be installed and used in accordance with the manufacturer's recommendations. Installation must be performed by properly trained and authorized personnel.
- □ The over-voltage category is II.
- The instrument may only be operated by personnel who have been trained on the use of the system. It is strongly recommended that all users read this manual prior to use.
- Never reach into the instrument while the unit is in operation, neither with parts of your body nor with other instruments.
- **□** Remove the transportation lock before switching on the instrument.
- □ Use the instrument only for the designated application.
- □ The instrument is designed for indoor use only (dry environment).
- □ The instrument may not be operated at altitudes above 2000 m above sea level.
- □ The instrument is designed to be operated within a temperature range of 10 to 28 °C. Operating the instrument at higher temperatures will lead to problems with the camera cooling (safety shutdown of the camera cooler)
- □ The instrument is designed to be operated at a maximum relative humidity of 80 % for temperatures up to 28 °C (non condensing).

- BERTHOLD TECHNOLOGIES assumes no liability for any damages, including those to third parties, caused by improper use or handling of the instrument.
- □ The user is responsible for connecting the instrument in accordance with the valid regulations for electrical instruments.
- □ Set the instrument up to ensure easy access to the mains switch.
- □ The mains supply voltage fluctuations must not exceed +/- 10 %. Operating voltage is AC 100- 240V +/- 10 %.
- □ The instrument is provided with a 3-pole grounded plug. If your wall outlet does not allow connection of a 3-pole plug, have a suitable wall outlet installed by qualified personnel or use an adapter for safe grounding. Please observe the safety specifications of the grounded plug. Only use the provided power cable or a cable that fits the specifications of the power cable provided with the instrument.
- □ The instruments are designed according to the IEC 61010-1 or EN 610 10-1 regulations for electrical measuring systems.
- To disconnect the instrument from power the appliance coupler has to be removed from the mains. Ensure that the distance to the wall is high enough to allow an easy removal of the mains cable.
- Do not open any instrument doors as long as the instrument is in operation.
- Service and repair work must be carried out by certified personnel only. After any service work the secure operation of the instrument must be tested.
- The operator may only perform the maintenance work described in this user guide.
- There are no exchangeable electrical components in the instrument. In case of malfunction call authorized service personnel.
- □ Use only parts described in this manual for servicing.
- Disconnect power supply before opening the instrument.
- □ Pull the power cord to disconnect instrument from power supply.
- □ Turn instrument off before pulling the power cord.
- Upon removal of the front and top parts of the housing no safety measures are in effect. Be aware of any moving parts. The interior of the instrument may reach temperatures that can cause burns. Some parts of the instrument may remain hot without visual indication for some time after the power has been turned off.
- The internal battery can explode if not properly inserted. The internal battery must be replaced with the original spare part (ID 17391) by certified service personnel. Used batteries must be recycled based on the manufacturer's directions.
- If you can see that the instrument has become unsafe to use, switch it off and disconnect it from power supply.
- □ If liquid gets inside the instruments, pull the power cord. Clean the unit or have it cleaned by an authorized service center.
- Protect yourself from electrostatic charge, as discharge could damage sensitive instrument parts, especially sensitive parts of the computer and electronics boards.





- When the lid is opened (e.g. filter change) ESD can no longer be guaranteed. To avoid any damages to the electronic parts it is recommended to take precautions (touching the metal case of a safety grounded object, wearing a grounding strap, etc.).
- □ The system always has to be primed with solutions recommended by the kit manufacturer.
- □ Use only reagents recommended by the kit manufacturer.
- **Use reagents only in accordance with the kit manufacturer's instructions.**
- Do not use any flammable or explosive solutions or liquids whose mixture is flammable or explosive.
- □ For instrument cleaning, please refer to the respective sections in this manual.
- Reliable instrument function can be guaranteed only when original spare parts are used.
- □ Pollution degree is 2.
- □ The instrument may only be used in rooms with a max. pollution degree of 2.
- □ This instrument must not be used in areas with potentially explosive atmosphere.

The tests and service work recommended by the manufacturer has to be performed to make sure that the operator remains safe and that the instrument continues to work correctly. Any service and maintenance work not described in this user guide has to be performed by authorized service personnel. Use the instruments only for the designated application.

## 2.1 Consignes de Sécurité

Attention! Ce symbole d'alarme, vous avertit de prêter attention aux consignes opératoires. En effet si vous ne suivez pas ces instructions, il peu y avoir un risque d'endommagement du matériel et également vous faire ecourir des risques pour votre propre sécurité. Il est impératif de respecter les instructions du mode d'emploi et de les respecter.

Ce mode d'emploi contient des informations et avertissements qui doivent être suivis par l'utilisateur afin de garantir un fonctionnement sûr des instruments.

Il est impératif de respecter les consignes de sécurité suivantes non seulement avant la mise en service mais aussi pendant le fonctionnement de l'appareil! Avant l'installation et mise en service de l'instrument tous les utilisateurs des appareils sont tenus de lire d'abord ces instructions de service, autrement ni le fonctionnement correct de l'appareil ni la sécurité de l'utilisateur peuvent être garantis. Ne pas suivre ces instructions de service peut invalider la garantie.

Le appareil a été fabriqué conformément aux prescriptions de sécurité en vigueur pour les appareils de mesure électroniques et médicaux. Si l'installation et/ou l'utilisation des appareils de mesure de prélèvements-échantillons sont/est soumise(s) à des réglementations prescrites par la loi, il appartient à l'utilisateur de les respecter.

Le constructeur a fait tout le nécessaire pour assurer le fonctionnement sûr des appareils (du point de vue électrique, électronique et mécanique). L'utilisateur est tenu de veiller à ce que les appareils soient installés correctement afin d'éviter toute altération de leur utilisation sûre.

Les appareils sont contrôlés à l'usine et livrés dans un état assurant la sécurité de fonctionnement.

- Les appareils ne peuvent être mis en service que par des personnes autorisées et ne peuvent être utilisés que par un personnel formé. Tous les utilisateurs qui travaillent avec les appareils doivent d'abord lire ce mode d'emploi.
- La catégorie de surtension est II.
- Les appareils ne doivent être utilisés que par du personnel formé à cet effet. Il est recommandé à tous les utilisateurs de lire ce mode d'emploi avant d'utiliser l'appareil.
- **D** Retirer les sécurités de transport avant la mise en marche.
- Les appareils ne doivent être utilisés que pour l'usage prévu.
- Berthold Technologies n'assume aucune garantie, y compris pour les dommages causés à des tiers, en cas de manipulation non conforme des appareils.
- Les appareils ne doivent être utilisés qu'à l'intérieur de locaux fermés (environnement sec).
- Les appareils ne doivent pas être utilisés à des altitudes supérieures à 2000 m au-dessus du niveau de la mer.



- Les appareils sont conçus pour fonctionner dans une plage de température de 10 à 28 °C. Des températures ambiantes plus élevées affectent le refroidissement de la caméra (arrêt de sécurité du refroidissement de la caméra).
- □ Les appareils sont conçus pour fonctionner avec une humidité relative maximale de 80 % (jusqu'à 28 °C) (sans condensation).
- □ L'alimentation électrique ne doit pas dépasser ±10 % de la valeur nominale. La plage de tension est de 100 240 V AC ±10 %.
- Il est de la responsabilité de l'utilisateur de s'assurer que les appareils sont installés conformément aux réglementations électriques locales.
- Les appareils sont équipés d'un câble d'alimentation à 3 pôles. Il s'agit d'un équipement de sécurité. Si la prise de courant ne prend pas en charge une connexion à 3 pôles, un électricien spécialisé doit installer une prise de courant à 3 pôles appropriée ou fournir un adaptateur approprié pour la mise à la terre de la connexion. Ne détruisez jamais les dispositifs de sécurité du raccordement à la terre. Utilisez uniquement le câble d'alimentation fourni ou un câble de même spécification et de même certification.
- ❑ Les appareils sont conformes aux prescriptions des normes CEI 61010-1 et EN 61010-1 relatives aux appareils de mesure électriques.
- □ Ne pas ouvrir l'appareil lorsqu'il est en service.
- Les travaux de service et de réparation ne doivent être effectués que par des spécialistes. Après les travaux, le fonctionnement sûr doit être contrôlé.
- Seuls les travaux de maintenance décrits dans le manuel peuvent être effectués par l'utilisateur.
- Lors des travaux d'entretien, seules les pièces indiquées doivent être utilisées.
- Avant d'ouvrir l'appareil, l'alimentation électrique doit être coupée.
- Pour déconnecter complètement l'appareil du secteur, il est possible de débrancher le câble d'alimentation. Il faut veiller à ce que la distance par rapport au mur soit suffisante pour que le câble d'alimentation puisse être retiré sans problème.
- Éteindre l'appareil avant de le débrancher.
- Lorsque l'appareil est ouvert, les mesures de sécurité ne fonctionnent plus. Attention aux composants mobiles ! L'intérieur des appareils peut atteindre des températures qui peuvent provoquer des brûlures. Certaines pièces peuvent rester chaudes sans signe visible, même après l'arrêt de l'appareil.
- Tous les appareils et accessoires livrés doivent être raccordés au réseau avec mise à la terre. Utiliser une fiche de sécurité !
- Attention : la batterie interne peut exploser si elle est mal installée ! La batterie interne ne peut être remplacée que par le même modèle ou par un modèle de remplacement recommandé (ID 17391) par une personne autorisée par Berthold Technologies. Les piles usagées doivent être éliminées conformément aux instructions du fabricant.



- Installez l'appareil de manière à pouvoir le mettre en marche et l'arrêter facilement.
- Si la sécurité de fonctionnement est compromise, les appareils doivent être éteints et débranchés.
- Si du liquide a pénétré à l'intérieur de l'appareil, débrancher la fiche secteur. Ouvrir l'appareil et le nettoyer ou le faire nettoyer par un centre de service agréé.
- Lors de l'ouverture de l'appareil, il faut éviter les charges électrostatiques (p. ex. dues à la moquette), car les décharges sur l'appareil peuvent endommager les pièces électroniques.
- Ne pas utiliser de liquides inflammables ou explosifs, ni de liquides dont le mélange est inflammable ou explosif.
- Respectez toutes les dispositions légales relatives à la manipulation des déchets biologiques, des réactifs et des échantillons de patients.
- Pour le nettoyage de l'appareil, veuillez consulter la partie correspondante de ces instructions d'utilisation.
- □ Le bon fonctionnement ne peut être garanti que si les pièces de rechange d'origine sont utilisées.
- Le degré de pollution est de 2.
- L'appareil ne doit pas être utilisé dans des zones à risque d'explosion.
- L'appareil ne peut être utilisé que dans des locaux présentant un degré de pollution maximal de 2.

Afin d'assurer la sécurité de l'utilisateur et le bon fonctionnement des appareils, effectuer les travaux d'inspection et d'entretien recommandés par le fabricant. Toutes les mesures d'entretien et de réparation allant au-delà de celles spécifiées dans ce manuel sont réservées aux techniciens autorisés.

## 2.2 Sicherheitshinweise



Die vorliegende Bedienungsanweisung enthält Informationen und Warnungen, die vom Benutzer befolgt werden müssen, um einen sicheren Betrieb der Geräte zu ermöglichen.

Dieses Zeichen weist den Benutzer auf wichtige Punkte hin, deren Beachtung unerlässlich ist.



Die folgenden Sicherheitshinweise sind sowohl vor der Inbetriebnahme als auch während des Betriebs des Gerätes unbedingt zu beachten. Vor Inbetriebnahme des Gerätes ist es zwingend erforderlich, die Bedienungsanleitung zu lesen, da ansonsten die Sicherheit des Gerätes und des Benutzers nicht gewährleistet wird.

Das Gerät wurde in Übereinstimmung mit den Sicherheitsanforderungen für elektronische und medizinische Messgeräte hergestellt. Bestehen für die Errichtung und/oder den Betrieb von Probenmessgeräten gesetzlich vorgeschriebene Regelungen, so ist es die Aufgabe des Errichters und Betreibers, diese einzuhalten.

Das Gerät emittiert keine gefährlichen Schallpegel. Sollten dennoch Geräusche wahrgenommen werden, die auf einen fehlerhaften Betrieb hinweise ist der Service zu rufen.

Der Hersteller hat alles unternommen, um ein sicheres Arbeiten der Geräte (bezüglich Elektrik, Elektronik und Mechanik) zu gewährleisten. Der Benutzer muss dafür sorgen, dass die Geräte so aufgestellt und installiert werden, dass ihr sicherer Gebrauch nicht beeinträchtigt wird.

Die Geräte sind werkgeprüft und wurden in betriebssicherem Zustand ausgeliefert.

- Die Geräte dürfen nur von autorisierten Personen in Betrieb genommen und nur von eingewiesenem Personal bedient werden. Alle Benutzer, die mit den Geräten arbeiten, müssen zuerst diese Bedienungsanleitung lesen.
- Uberspannungskategorie ist II.
- Die Geräte dürfen nur von dafür geschultem Personal betrieben werden. Es wird allen Anwendern empfohlen, diese Bedienungsanleitung vor Benutzung zu lesen.
- **D** Transportsicherungen vor dem Einschalten entfernen.
- Die Geräte dürfen nur für den vorgesehenen Zweck eingesetzt werden.
- Berthold Technologies übernimmt keinerlei Gewährleistung, auch für Schäden gegenüber Dritten, die durch unsachgemäße Handhabung der Geräte hervorgerufen werden.
- Die Geräte dürfen nur innerhalb geschlossener Räume betrieben werden (trockene Umgebung).
- Die Geräte dürfen nicht in Höhen von mehr als 2000 m über dem Meeresspiegel betrieben werden.



- Die Geräte sind dafür ausgelegt, innerhalb des Temperaturbereiches von 10 bis 28 °C betrieben zu werden. Höhere Umgebungstemperaturen beeinträchtigen die Kühlung der Kamera (Sicherheitsabschaltung der Kamerakühlung).
- Die Geräte sind dafür ausgelegt, bei einer maximalen relativen Luftfeuchte von 80 % (bis zu 28 °C) betrieben zu werden (nicht kondensierend)
- Die Stromversorgung darf nicht mehr als ±10 % des Nominalwertes aufweisen. Der Spannungsbereich beträgt AC 100 – 240 V ±10 %
- Es liegt im Verantwortungsbereich des Anwenders, dass die Geräte nach den lokalen elektrischen Vorschriften installiert werden.
- Die Geräte sind mit einem 3-poligen Netzkabel ausgestattet. Dies ist eine Sicherheitsausstattung. Wenn die Steckdose keinen 3-poligen Anschluss unterstützt, muss ein Fachelektriker eine passende 3-polige Steckdose installieren oder einen passenden Adapter zur Erdung des Anschlusses bereitstellen. Zerstören Sie niemals die Sicherheitsvorkehrungen des geerdeten Anschlusses. Verwenden sie nur das beigelegte Netzkabel oder eines mit der gleichen Spezifikation und Zertifizierung.
- Die Geräte entsprechen den Vorschriften der IEC 61010-1 und EN 61010-1 für elektrische Messgeräte.
- □ Nicht öffnen, wenn das Gerät in Betrieb ist.
- Service- und Reparaturarbeiten dürfen nur von Fachleuten ausgeführt werden. Nach den Arbeiten muss der sichere Betrieb geprüft werden.
- Es dürfen nur die im Handbuch beschriebenen Wartungsarbeiten vom Anwender ausgeführt werden.
- Bei Wartungsarbeiten dürfen nur die angegebenen Teile verwendet werden.
- Vor dem Öffnen des Gerätes ist die Stromzufuhr zu unterbrechen.
- Um das Gerät vollkommen vom Netz zu trennen, kann das Netzkabel gezogen werden. Es ist auf ausreichend Abstand zur Wand zu achten, damit das Netzkabel problemlos abgezogen werden kann.
- Gerät ausschalten, bevor der Stecker gezogen wird.
- Wenn das Gerät geöffnet ist sind Sicherheitsmaßnahmen nicht mehr in Betrieb. Auf bewegliche Komponenten achten! Das Innere der Geräte kann Temperaturen erreichen, die Verbrennungen verursachen können. Einige Teile können heiß bleiben ohne sichtbare Zeichen, auch nachdem das Gerät abgeschaltet worden ist.
- Alle gelieferten Geräte und Zusatzgeräte sind geerdet ans Netz anzuschließen. Schutzkontaktstecker verwenden!
- Achtung: Die interene Batterie kann bei falschem Einbau explodieren! Die interne Batterie darf nur mit dem gleichen oder einem empfohlenen Ersatzmodell (ID 17391) von einer von Berthold Technologies autorisierten Person ausgetauscht werden. Verbrauchte Batterien müssen entsprechend den Herstelleranweisungen entsorgt werden.



- Stellen Sie das Gerät so auf, dass Sie es leicht ein- und ausschalten können.
- Bei Beeinträchtigung der Betriebssicherheit sind die Geräte abzuschalten und vom Netz zu trennen.
- Ist Flüssigkeit in das Innere des Gerätes gelangt, Netzstecker ziehen. Das Gerät öffnen und reinigen bzw. durch eine autorisierte Servicestelle reinigen lassen.
- Elektrostatische Aufladungen (z.B. durch Teppichböden) müssen beim Öffnen des Gerätes verhindert werden, da Entladungen am Gerät zur Beschädigung elektronischer Teile führen können.
- Es dürfen keine entzündlichen oder explosiven Flüssigkeiten oder keine Flüssigkeiten, deren Mischung entzündlich oder explosiv ist, verwendet werden.
- Beachten Sie alle gesetzlichen Vorschriften f
  ür den Umgang mit biologischem Abfall, mit Reagenzien und Patientenproben.
- Zum Reinigen des Gerätes bitte den entsprechenden Teil dieser Bedienungsanleitungen beachten.
- Ordungsgemäße Funktionalität kann nur bei Verwendung der Originalersatzteile garantiert werden.
- □ Verschmutzungsgrad ist 2.
- Das Gerät darf nicht in explosionsgefährdeten Bereichen verwendet werden.
- Das Gerät darf nur in Räumlickeiten mit einem maximalen Verschmutzungsgrad von 2 betrieben werden.

Für die Sicherheit des Benutzers und die Funktionsfähigkeit der Geräte sind die vom Hersteller empfohlenen Überprüfungen und Wartungsmaßnahmen durchzuführen. Alle über die Betriebsanleitung hinausgehenden Wartungs- und Instandhaltungsmaßnahmen dürfen nur von autorisierten Technikern ausgeführt werden.

## 3 Warranty and Customer Service

### 3.1 Warranty Statement

The instrument is sold in accordance with the general conditions of sale of Berthold Technologies GmbH & Co KG and its affiliates and representatives.

Berthold Technologies warrants this product to be free of defects in material and workmanship for a period of 12 months from the date of delivery, ex works Bad Wildbad.

Berthold Technologies or its authorized representative will repair or replace, at its option and free of charge, any product that under proper and normal use proves to be defective during the warranty period.

Berthold Technologies shall in no event be liable or responsible for any incidental or consequential damage, either direct or indirect.

The above warranty shall not apply if:

- a) the product has not been operated in accordance with the operating manual
- b) the product has not been regularly and correctly maintained
- c) the product has not been repaired or modified by a Berthold Technologies authorized representative or user
- d) parts other than original Berthold Technologies parts are used
- e) the product and parts thereof have been altered without written authorization from Berthold Technologies GmbH & Co KG
- e) the product has not been returned properly packed in the original Berthold Technologies packaging

This warranty does not apply to any third-party product involved in the application.

Berthold Technologies reserves the right to refuse to accept the return of any product that has been used with radioactive or (micro)-biological substances, or any other material that may be deemed hazardous to employees of Berthold Technologies. Such products have to be properly decontaminated and marked.

Before returning products to Berthold Technologies ensure the devices are properly decontaminated.

A return material authorization (RMA) number must be obtained and clearly identified on the packing and documents. Please visit the Berthold Technologies website and fill in the required information to obtain the number.

https://www.berthold.com/en/company/service/form-rma-return-material-authorization/

Retain the original packaging for use if the instrument needs to be returned to Berthold Technologies.

## 3.2 Customer Service

**Customer service will be provided in the first instance by the network of Berthold Technologies representatives**. In the event of any problem experienced with your instrument, the first recourse should be **your local Berthold Technologies representative**. For further problems requiring hardware or software expertise, the Technical Support group at Berthold Technologies GmbH & Co KG will be available by phone, fax or email to deal with your queries.

Please use the following contact for your enquiries.

Berthold Technologies GmbH & Co.KG Technical Support Calmbacher Str. 22 75323 Bad Wildbad Germany Phone: +49 7081 177 171 Fax: +49 7081 177 301 Email: service@berthold.com https://www.berthold.com/en/company/service/form-more-services/

Please visit the Berthold Technologies website for any service inquiries.

Please also make sure that you have the relevant information available before contacting Berthold Technologies. Helpful information would include:

- serial numbers, part number, revision: see production label on instrument
- set production laber on institutien
- software and firmware versions
- monitor and log files (refer to the respective service manuals)

## 4 Quick Start

These quick start instructions provide a quick access to the program. You are guided step by step through the major program functions. The page numbers in the headers refer to the individual sections in the manual that provide more detailed information about the respective functions.

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#### 4.1 Installation

#### 4.1.1 Requirements

Please observe the following requirements when working with the NightSHADE evo:

- 2-5 mains outlets (depending on accessories). The maximum power consumption of the NightSHADE evo without PC and external devices is max. 600 W. The power consumption of the external cooling unit is 340 W. To power on/off the system with one switch we recommend connecting all devices to a common outlet strip.
- At least 160 cm width of workplace (the load-carrying capacity of the table or the desktop should be approx 100 kg). Make sure that the table or desktop stands firm and does not wobble.
- To use network resources (network printer, file server, etc.) a connection to a network is essential. A connection to the Internet is recommended for easier support, but it is not essential for operation of the NightSHADE evo.
- To allow ergonomic working, the NightSHADE evo should always be set up to the left of the PC, as the door hinges are on the left.
- To fill the cooling unit for the LED panels up to 4 liters of destilled water are required. It is recommended to place the external cooler on the floor (under the lab table etc.). Please note that a tubing connection to the NightSHADE evo is required for the coolant. In case the cooler is placed unter the lab table a cut-out on the table area is required to get shortest possible tubing.



#### 4.1.2 Unpacking and Carrying

The NightSHADE is delivered in a so-called flight case. The case consists of 3 parts, the lower base and two upper shells. The upper shells are locked to the base with butterfly locks and can be removed after releasing the locks.

Since the lower base of the flightcase is equipped with wheels, the instrument can be placed in front of the table to allow an easy placement on the table top.



# The NightSHADE evo is heavy. To unpack the instrument from the flight case and lift it to the table at least 2 persons are required.

When the upper shells are removed the black handles at the side of the instrument can be popped out to carry the instrument.

One person on each side of the instrument must grab the handle to lift up the instrument carefully.





The wheels must be secured using the built-in brake when the box is not used or stored to avoid uncontrolled movement of the flight case.

To activate the break just push the pedal (see image) at the corresponding wheel. Pull back the pedal to release the brake.

To lift up the instrument to the table both persons holding the

handle must lift up the instrument simultaneously and lift it out of the lower base and up on the table. A third person that secures the instrument with both hands at the base plate after lifting it out of the lower base is recommended. Once the instrument is placed on the table the handfles can be folded in.

- □ Unpack the boxes and make sure that the delivery is complete. If you should discover any sign of damage on the instruments or instrument parts, please notify the shipping agent and/or the distributor immediately.
- Provide adequate space for setting up the instruments. To set up the NightSHADE evo, the computer and the printer next to each other about 2 meters of desk space are required.
- □ The device feet of the NightSHADE evo are height-adjustable to compensate a possible unevenness of the table. By turning the device feet, their height can be adjusted to ensure a safe foothold of the NightSHADE evo.



#### 4.1.3 Camera Installation

For safety reasons, the camera is delivered in a special case to prevent damage during transport. Therefore, the camera has to be installed first before it is taken into operation. **Please note: Do not turn on the NightSHADE evo as long a no camera has been installed**.

The camera is connected to the instrument using two cables. One cable proviodes the supply voltages (2), the other USB-cable (3) is to control the camera and readout the data.



The cable (1) is used to supply the fan in the top cover to allow a proper air exchange. Make sure this cable is plugged in correctly as otherwise the fan will not work and the camera could get cooling problems during longer operations.

After switch on, the device is initialized.

If the device is transported over larger distances (e.g. in a car or truck), you have to dismantle the camera prior to the transport to rule out that the camera will get damaged. To prevent damage please remove or secure the base plate whenever you transport the device.

#### 4.1.3.1 Top view camera

The NightSHADE evo camera has a USB port to control the camera and to transfer data. The camera is delivered in a separate case which also accommodates other accessories, such as the connection cable etc. Make sure no protective stickers are in front of the camera (protecting the shutter hole).



Install the camera that the red dot located on the bajonet lock is matching the red dot on the NightSHADE evo socket and fix it in the bayonet socket until it clicks into place. Connect the power supply cable (1) to the camera and as well as the outlet socket and screw it.

Make sure that the USB-connector (2) is plugged into the camera and outlet socket properly.

With this the installation of the camera is completed.

#### 4.1.3.2 Side view Camera

The NightSHADE evo offers the special option that the camera can be mounted on the side, too. This feature enables the system to take images of plants from the side or to measure the sample dishes in an upright standing position. The upright standing position is used when the sample changer is assembled into the NightSHADE evo.

The power supply and the USB connection for the camera is located at the rear side of the instrument. Please use the supplied special cables to connect the camera to the instrument.

To assemble the camera at the side proceed as follows:

- □ Turn off the NightSHADE evo.
- Open the top cover and remove the USB cable and the power supply cable from the camera.
- Remove the blind plug from the side view camera holder. To remove the cover, turn it slightly.
- Dismantle the camera by turning it counterclockwise until it can be removed
- Place the camera in the same way at the camera holder at the left side of the instrument. Make sure that the red dot at the camera is matching the red dot at the instrument bajonett.
- □ Plug the power supply cable to the rear panel and then to the camera.
- Plug the USB control cable to the rear panel and then to the camera.
- □ Put the blind plug to the top position of the camera to avoid incoming light.
- □ Switch on the NightSHADE evo.





#### 4.1.4 Connecting

The power supply and device communication ports are located on the rear panel of the NightSHADE evo. Plug the power cord into the mains inlet unit(1). The main power switch is located next to the mains inlet.

Connect the NightSHADE evo to the PC via the USB3 B-socket (2) on the rear panel using a USB3 cable (A to B).



The 7-pin Tuchel socket (4) is the power supply for the side mounted camera, the USB3 socket (3) is used for the control of the side view camera only.

To ensure secure operation only connect certified IT equipment to the instrument.

## Check to make sure that the system voltage indicated on the instruments matches the line voltage.

If possible connect all devices via one outlet strip to the mains supply.

The NightSHADE evo is initialized after power on; a control lamp on the front panel is flashing.

The NightSHADE evo camera is ready for operation after it has cooled down to operating temperature; this takes approximately 10 minutes.

# The cooling of the camera is initialized when the indiGO software is started (controller startup). Switching on the NightSHADE evo without starting the software will not cool down the camera.

The NightSHADE should only be connected to mains power supply systems that are secured by a 10A automatic circuit breaker.

#### 4.1.5 Power On

After power on, the device starts an automatic initialization where all relevant components will be initialized. During this time, the LED on the front panel is flashing red. After the successful initialization the NightSHADE evo is ready for operation.

The indioGO software automatically recognizes the NightSHADE evo. No further steps are necessary.

#### 4.1.6 Connecting to the PC

To assure the proper operation of the NightSHADE evo device drivers are necessary. All device drivers are automatically installed through the indiGO software setup.

When connecting the instrument to PC, new devices are detected through the windows device manager. Using the automatic installation (not using the Windows update driver repository) the drivers are installed automatically. This procedure can take a while.

Do not connect the instrument to the PC when the indiGO software has not been installed yet.

#### 4.1.7 Software Installation

The indiGO software is included on the enclosed USB stick.

Use this media to install the software in order to use the correct individual calibration data which is automatically copied during installation. Keep this media in a secure place. You will need it again e.g. for a new installation on a different PC.

For the operation of the NightSHADE evo the indiGO software version 2.1 or higher is necessary.

We recommend using Windows 10 as operating system. Older operating systems than Windows 7 are not supported.

To start the installation program, click on the file **SETUP.EXE** on the data carrier. After launching the installation program a welcome screen appears:



After the welcome screen the folder for the software installation must be defined. If possible confirm the suggested folder. Click on Letter to get to the next page.

<b>Indigo</b> 2.1.0.0	đ	<b>Indigo</b> 2.1.0.0	ā
Enter a Destination for the Installation C:\Program Files (v86)\IndiGo	Browse		Install indiGO as © Evaluation - Demo Version © Full/Update Version with Instrument control
< Back	Cancel		<back next=""> Cancel</back>

IndiGo can be installed in two different configurations. The evaluation or demo version will install a preconfigured system with instrument simulation to work without an instrument connected or to use it on office PCs for offline data evaluation. In this case select "Evaluation – Demo Version". The installation will start immediately. To update an existing indiGO installation (Evaluation or full version) or new installtion with full instrument control, select "Full/Update Version with Instrument control".

Depending on the hardware, a default selection of system filters might be available and can be installed through the setup. If you want to install a set of default filter slider and wheel tick the box **"Install default filters"**. The default filters are empty filter sliders without any filters.





In this case the default systems filter management file will be copied to your computer. If you reinstall the software the current active systems filter management file will be overwritten.

To get to the next screen asking for the drivers to be installed on the system click on **metalled** 

<b>Indigo</b> 2.1.0.0	
Install Ad	ditional Device Driver
	<back next=""> Cancel</back>

When working with the NightSHADE evo the PCI drivers are not required. Use the selections as suggested above.

**Note:** Due to the fact that the setup will install device drivers for the NightSHADE evo hardware on the computer, the Windows-system might show a warning that a signature of a Windows driver cannot be verified. In this case allow the setup to install these Windows drivers.

After the installation the controller will be initialized for the first time.

If the installation was performed using an instrument specific installation media (USB Stick) the configuration procedure can be slightly different since the installer has already copied the instrument configuration to disk from the USB Stick during installation.

#### 4.1.8 Instrument Controller First Time Initialization Setup

The **Instrument Controller First Time Initialization Setup** starts via the setup. Please note that this Setup will only be executed when the software is installed on a computer for the first time without calibration data.

indige
Welcome to the Instrument Controller First Time Initialization Sctup
You are just a view steps away from beginning your work.
Please ensure that all devices are powered on and has been detected by Windows. You can validate the devices using the Windows Device Manager.
<ul> <li>Select <b>Hext</b> to setup your Instrument now.</li> <li>Select <b>Cancel</b> to shutdown the application and setup your Instrument later.</li> </ul>

Select <u>Next></u> to continue.

			i	ndige
Setup Serial Nur Please enter the In Number.	m <b>ber</b> nstrument	Ident	Numbera	nd the Serial
Ident Number:	055393	1.4	101	- 20
UND THE TRUTTLET		S	ican Device	B
	Lo	ad Fac	tory Defau	ít Settings

Please enter the instrument ident-number and the serial number. You can find both numbers on the back of the instrument.

You can check the instrument ident-number by starting **"Scan Devices"**. If the calibration for your device was stored on the NightSHADE evo by Berthold Technologies before it was delivered you can download the stored calibration parameters from the instruments flash memory now by confirming **"Yes"**.

owlCtrl		8
?	Devices found at BT040508. Query default configuration from flash?	
	Ja Nein	

If you want to recalibrate your instrument please select "No".

You can load a default calibration for your instrument type via **"Load factory default settings"**. The factory default settings are chosen by the system via the given instrument ident-number.

Select <u>Next></u> to continue.



This step of the NightSHADE evo Controller First Time Initialization Setup allows the import of further created configuration backups and calibration files.

Select <u>Next></u> to continue.

Welcome				
	indige			
Setu	Setup NightOwl Administrator			
The I main for t	The NightOwl Controller needs to create an administrator to maintain your NightOwl. Please enter a <b>Name</b> and a <b>Password</b> for the administrator account.			
War	Warning			
If yo prote	If you don't enter a password, the administrator account is not protected.			
	Name: admin			
Pas	sword: admin			
Device Manager	< Back Next > Cancel			

To maintain the instrument the instruments controller needs to create an administrator account. Please enter a name and a password for the administrator account.

Select <u>Next></u> to continue.

Welcome			
indice			
Validate NightOwl Administrator			
To validate the administrator account logon now.			
Logon			
Name:	admin		
Password:	•••••		
Device Manager	< Back Next > Cancel		

In the next screen log on in order to verify the administrator account.

Select <u>Next></u> to continue.

Welcome	×
	indige
	NightOwl Controller Startup
	Please select, when you want to start the NightOwl Controller on your system.
	Note
	It is recommended that the controller is started with Windows. This option will give you the best support for scheduled measurements. Anyway, if you don't use the scheduler you can start the Controller with IndiGo.
	Start Controller with IndiGo     Start Controller with Windows
Device Manager	< Back

Next, choose a start method for the controller. . With regular installations it is recommended to select the first option ("Start controller with indiGO"). The indiGO controller takes care of the hardware control and performs the measurements even when indiGO itself is not running. Choosing this option the controller will be started everytime indigo is launched (if not already running). After a reboot it will not restart automatically.

With instruments running unattended for a longer period it is recommended to choose the second option (Start controller with Windows) to have a working system even after a power failure when the PC is rebooting. In this case the controller is added to the startup group of Windows and starts automatically after login.

Select	Next >	to continue.
--------	--------	--------------

Welcome	
	indige
	Congratulations
	You have successfully managed the First Time Setup and the NightOwl Controller is now ready to support your work.
	A quick start guide is available in the owners handbook or through the help menu.
	Select Finish to restart and initialize the NightOwl Controller now.
	You can logon to the Controller Setup through the menu by activating the controller icon on the taskbar.
Device Manager	< Back Finish Cancel

With this screen the NightSHADE evo Controller First Time Initialization Setup is completed. Select **"Finish"**, to restart and initialize the instrument controller now.

#### 4.1.9 First Time Initialization Setup

To configure indiGO launch the software by double-clicking on the indiGO icon.



The first time initialization setup will start.

Welcome		Welcome	
	indice		indige
	Welcome to the India Inst Time Initialization Setup You are just a view steps away from beginning your work with India. The wiese of a guide you through the India First Time India between the guide you through the India First Time India between the steps India man. Setect Enverse I solution the application and setue India bater.		Setup as Cleant or Server If this ratialation is a Server installation, please ensure that you accounting. If this installation is a Cleant installation, the First Time Setup on the Server must have been finding audicastily and the OWL Controllermants active in the Server on. Please choose one of the following Server: A Nontroll is connected to the computer. © Cleant A Nontroll is connected to a different computer.
	<red. [lext="">] Cancel</red.>		< Back Next> Cancel

If the PC is directly conneted to the NightSHADE evo via USB choose "Server" to install the instrument controller as well as the evaluation part.

In case of a network connection to a NightSHADE evo, select the "Client" installation.

Click <u>Next></u> to continue.

Welcome	indice	
	Setup Database Nesse setup the location for the IndiCe database. This installation care orable a new database on this computer or can be connected to an airsady existing distabase. If you choose an existing distabase, the database will be attached	indigo
	o un restation. Detabase Location: Surnave didictored	The folder 'ci\indigoDatabase' does not exist. Do you want to create it?
	<bed: next=""> Cancel</bed:>	La Nein

If indigo is installed for the first time the first time initialization setup will create a new and empty database.

If indiGO should be connected to an existing database, it has to be attached to this database. Use the browse command to allocate and define a suitable database

#### Please note:

IndiGO manages all measuring data in a file-based database.

BERTHOLD

The indiGO software is able to manage the database on the local computer as well as on a network location. Therefore the database location can be set up to an individual folder.

The benefit of storage on the network is that one has access to the data from several evaluation stations, which is a major advantage in a multi-user environment.

Due to the huge amount of data indiGO does not use a server-based database like Microsoft SQL-server. Therefore the sharing of measurement data through a multi-user environment is not possible. For this reason measurement data should only be edited by one user at a time.

Old databases created with indiGO Version 1.x are not compatible. But data created with these indiGO versions or WinLight can be imported into the new indiGO database using the new indiGO software.

Click <u>Next</u> to continue.

elcome	
	indice
	Setup Controller Connection
	Please setup the address to the OWI. Controller.
	Valid addresses might be:
	<ul> <li>192, 168, 0, 1-1024</li> <li>bk-rightCwl-1024</li> <li>bk-rightCwl: 1024</li> </ul>
	Controller Address:
	LCCALLEST 002
	< Badt Next > Cancel

This dialog box defines the connectionaddress to the NightSHADE evo controller. If the controller is running on the same computer the default settings "LOCALHOST-1024" can be applied.

If the NightSHADE evo controller is running on a remote PC in the network, please set up the correspondending IP-address or the host name like"192.168.1.2-1024" or "IMAGING\_PC-1024". The IP port number to contact the instrument controller is always 1024. Host and port number must be separated with a dash "-".

Welcome	Welcome	
	indice	indice
	Disable Inditio User Hanagement Been fift is not recommended, you have the opportunity to disable the User Management provided by Indita of the smallalation. Warning If the User Management is disabled, all data of the Indita disables is presented to any user using the indialation of Indita, Theorem any user inget view and meetify the data.	Setup an IndiGo Default User You have the apportunity to setup a default user for IndiGo. At starbup, JudiGo will login this user to the delabase. Viraming If the default user has administrative access to the delabase, all data of the JudiGo database will be presented to any user using the statistics of Pholoc.
	<ead: next=""> Cance</ead:>	< Eack Next > Cancel

IndiGO offers a user management. Three different modes are available:

- <Disable User Management> (not recommended)
- <Set a Default User Account>, user will be logged on automatically or
- User must log-in everytime the software starts.

In case no user management is required tick the box **<Disable User Manage-ment>**. No log-in is required and no user management is available. If the User Management has been disabled it can be re-activated via the system configuration at any time.

Welcome		X
	indice	Ď
	Validate Default User To validate the user account used for the automatic logon, plea logon now by using the designated account.	se
	Please logon now	
	Paesword:	
	< Back Next > Cance	a )

To set up an automatic log on to indiGO using a default user account, tick the box **<Set a default user account>**.

By selecting **<Next>** you will be asked to set up a user name and a passwort. Please note that the created account will have administrator rights.

Click Next> to continue.

At the end of the configuration asummary page is displayed.



To end the first time initialization setup select [Finish].

#### 4.1.10 User Management

IndiGO supports an integrated user management.

Instrument Controller Setup	User Management <b>User Manager</b>				indig	×
	Edit	User Name	Created 11/3/2009 11/6/2009 11/6/2009 11/6/2009	Expire	User Role Admin Guest Service User	
	Delete					
Instrument is offline					Clos	e

The User Management can be enabled or disabled by the system administrator. Refer to the **<User Management> <Advanced>** settings to enable or disable the user management.

Only admins can access the user management.

Users are organized in groups, so called roles. You can assign one out of five roles for each user.

- Service
- Admin
- Supervisor
- User
- Guest

#### Service

Service users can change all settings of the system. New users can be created and existing users can be deleted. The access to measurements is restricted through the user management. Other users can be invited to own projets.

#### Admin

This is the system administrator role. Administrators can change all settings of the system. New users can be created and existing users can be deleted. Administrators have access to all measurements created. Administrators can change the ownership of measurements and manage invitations of all users.

#### Supervisor

This is the system supervisor role. Supervisors can change most settings of the system like heater temperature, filter management and system status. Supervisors have access to all measurements created by themselves or where they got invitations. Supervisors can change the ownership of measurements and manage invitations of all projects they have been given access to.

#### User

Users can change any settings necessary to run measurements. Applications and templates available can be created and changed. Users can access and change the filter management. The access to the user management and any controller service functions such as the system



calibration is denied. The access to measurements is restricted by the user management. Other users can be invited to own projects.

#### Guest

Guests can run measurements in projects where they have been invited to.

#### **4.1.11 Installing Fluorescence Filters**

#### **Excitation Filter Slider**

The NightSHADE evo is equipped with automatic filter changers for the excitation and emission path. The excitation filter is located on a filter slider, which is accessible after opening the cover on the front panel of the NightSHADE evo. The filter slider can hold five excitation filters with a diameter of 25 mm (1 inch). The 6<sup>th</sup> position is reserved for luminescence measurements to avoid incoming light through the fibre optic.

Filters placed in the filter slider should be fixed with the snap rings provided with the device. Using excitation filters the inprinted arrow on the filter should match the arrow printed on the fluorescence filter housing. The arrow should face the sample, turning away from the light.



The filter slider can be ejected via the indiGO software (Filter Manager). The filter slider duct is protected by a light blocking plug, which must be mounted after inserting and loading the filter slider.

Instrument Controller Setup								? >
Instrument	^	Filter M <b>Excit</b>	lanagement ation Inver	nto <b>ry</b>			inc	GO
Camera A Camera Advanced Fiters Heater Sample Changer Daylight Simulation Calibration Galibration Serial Number Video Codecs				1:	GFP 475/20	475 [nm]	20 [nm]	70 [%]
Quick Check			Name	Filter 1	Filter 2	Filter 3	Filter 4	Filter 5
Advanced			Slider A	GFP 475/20				
Transport About Filter Management								
Emission Inventory			<					>
Advanced	~		Filter Inven	tory F	Filter Manager	. E	Eject	Load
Instrument is ready								Close

To eject the slider select **Eject**.

When loading the filter please observe the correct installation direction (see above). To reload the slider, select After that the slider is initialized by the NightSHADE evo.

#### **Emission Filter Wheel**

The emission filters are located on a filter wheel in front of the lens. A filter wheel can hold up to five filters with a diameter up to 50 mm using specific filter holder inserts.



The positions of the filters are numbered and identified by a respective number printed on the left side of the corresponding filter wheel position.

For luminescence and photographic measurements filter position 6 is used. Using emission filters the inprinted arrow on the filter should face the sample, turning away from the camera.

To insert the filter slides switch off the NightSHADE evo, open the filter flap at the front side of the instrument, move the wheel manually to the required position and slide in the filter holder. After moving the filter wheel manually it is required to initialize the wheel again. Otherwise the filter positioning might be wrong. It is recommended to change filters on the filter wheel while the instrument is turned off.


## 4.1.12 Filter Management

indiGO offers an integrated filter management. The filter management is used to describe the filter load on the excitation filter sliders as well as the emission filter wheels.

The system comes with a set of GFP filters. The recommended excitation position for the GFP exciter (478/20) is position 1.

The GFP emission filter (520/10) is recommended to be placed in postion 4. The IR-cutoff filter that comes with the NightSHADE evo is recommended to be placed in position 5



## 4.1.13 Filter Inventory

The filter inventory is used to collect the available filters. indiGO governs two filter inventory lists. One is used for excitation filters, the other for emission filters.

The inventory lists can be either reached via **<Excitation Inventory> <Filter Inventory>** for the excitations filters or via **<Emission Inventory> <Filter Inventory>** for the emission filters.



To add a new filter to the inventory list, select the **<New>** command.

Edit Filter		-	X
Setup Filter Proper	ties		
Name:	MyName		
CWL [nm]:	560	A	
HBW [nm]:	400	Л	_
T [%]:	100	Ц.	
			OK Cancel

Each filter is described by the following properties:

- The name of the filter
- The center wave length of the filter
- The half band width of the filter (FWHM)
- The transmission rate



### 4.1.14 Filter Manager

Once a filter has been added to the filter inventory list it can be used for an excitation filter slider or an emission filter wheel.

The filter manager can be reached via **<Excitation Inventory> <Filter Manager>** for the excitations filters or via **<Emission Inventory> <Filter Manager>** for the emission filter.



To add a new excitation filter slider or an emission filter wheel to the manager select **<New>**. To change or edit the load of a slider or a wheel select **<Edit>**.

Edit Filter		×	Edit Filter		×
Excitation	Filter e: Slider A		Setup I	Emission Filter Properties	
Excitation	Filter Properties		Setup	Emission Filter Properties	
		Filter 5:     Empty     ~       Filter 4:     Empty     ~       Filter 3:     Empty     ~       Filter 2:     Empty     ~       Filter 1:     GFP 475/20     ~			Filter 5:     IR cutoff       Filter 4:     GFP 520/10       Filter 3:     Open       Filter 2:     Open       Filter 1:     Open
Filter Inventory		OK Cancel	Filter Inventory		OK Cancel

The loading can be changed using the filter dropdown list **<Filter N>**. Please keep in mind, that only available and unused filters are displayed in the droplist. Before you can assign a new filter to a position , either a new filter has to added to the **<Filter Inventory>** or an existing filter has to be removed from its position.

### 4.1.15 Filter Manager – Backup and Restore

All data from the filter manager can be exported from the system and stored to an archive as a backup using the command **<Filter Management> <Advanced> <Export Filter Inventory>**.

A further created backup can be imported using the command **<Filter Management> <Advanced>** <**Import Filter Inventory>**.

# 4.2 First Acquisitions

# 4.2.1 Quick Start Glossary

### Projects, Applications and Identities:

All parameters necessary to run a measurement are defined in an application.

Individual samples, called identities, can be organized in a project. Each project is connected with an application.

To perform a measurement, select an identity and the system automatically refers all settings of the project and the correlated application.

After the measurement is accomplished all data will be automatically transferred to the preselected identity of the project.

All acquired data is stored and organized as a structured tree. The main nodes are the projects containing identities, containing measurement data.

### Example:

- Project 1
  - Identity 1
    - Measurement 1
    - Measurement n
  - Identity n
    - Measurement 1
    - Measurement n
- Project n

### **Projects and Applications**

Applications consist of a pre-measurement, a measurement and a post-measurement.

When a project is created a mirror copy of the used application is saved with that project. This means that changing the original application template later on will not affect the project's parameters. Consequently changing parameter settings in the project (e.g. exposure time) has no influence on the original application template.

On the other hand before each measurement the application settings of a project can be modified. The last modification is always safed with the project. Settings used for each specific measurement performed can be found in the report.

### **Projects and Users**

Projects can be associated to users. When created, a project belongs to a specified owner. The owner has the possibility to allow other users to access the project and to run measurements on identities associated with the project. To allow other users the access to a project, the owner has to create an invitation.



# 4.2.2 Launching the software

Depending on the system configuration it might be required to log-in after the indiGO has been started. When no other users are defined the standard log-in is as follows:

Log-in:	Admin
Password:	Admin

After log-in the main window of indiGO is shown:



# 4.2.3 Defining a Measuring Template

indigo defines different measurement methods (e.g. luminescence measurement, fluorescence measurement) by an measuring template. In each template the measurement mode, the exposure time, the measurement sequence and the filter settings, (if available), are assigned.

Administrator access rights are necessary, to set up a measuring template . If this is not guaranteed the function to define a measuring template is not available to rule out faulty operations.

To set up a new template refer to **<Applications & Templates> <Measuring Templates>**. Select **<New Measuring Template>** from the **<Measuring Templates>** context menu.



First enter a name after indiGO has started the wizard In addition a descriptive comment can be added. Due to Windows file and path name conventions special characters such as < ? " : | / / \*. are not allowed.



Select  $\longrightarrow$  to define the measurement workflow.

Four different kinds of measurements can be performed. Please note that depending on the system configuration some of the selections might not be available (e.g. due to a missing fluorescence illumination module).

Example:

To define a luminescence measurement, select "luminescence" from the dropdown list after clicking on Sade T

۵	Data Aquisition New Template
Setup Name	
Data Aquisition	Photo Luminescence
Sample Size	Fluorescence Transillumination

The Luminescence image wizard will pop up.

Luminescence		<b>×</b>
	Aquisition Settings	20
Aquisition	Exposure Time [s]: Gain:	High 👻
Post Processor	Read Out:	Slow 👻
	Binning Factors:	4x4 Binning 👻
Emission		
Quick Check	< Back Next > Fin	ish Cancel

• Exposure Time

is the length of the time the CCD chip is exposed to the sample

• Gain

is an internal conversion factor. Low Gain offers high dynamic range while high gain gives higher intensity numbers but limited dynamic range. When using the NC100 camera the difference in signal between low and high gain is about factor 4.

Read out

defines the speed to read out the camera. High speed is recommended for photographic images and high intensity measurements such as fluorescence since a higher



background noise will be obtained. Low speed is ideal for highest sensitivity luminescence measurements. Be aware that with low speed the camera readout can take up to 15 seconds.

### Binning Factor

is the combinated readout of neighbouring CCD pixels. The result is an increased signal and thus an improved sensitivity and a better signal-to-noise ratio, however, at the cost of loss in spatial resolution.

Select **\_\_\_\_** to get to the post processing definition.

Luminescence	Post Processor Cosmic Suppression: Subtract Bkgnd Image: Bkgnd Correction: Flatfield Correction: Image Accumulation	
Quick Check	Image Accumulation:	Solisabled Visabled Visabled

The Post Processor page offers additional image processing features which can be performed after the exposure to increase the image quality.

### • Cosmic suppression

is an imaging filter to get rid of small high intensity pixels caused by cosmic ray during the exposure.

### • Subtract background image

Thisfunction will subtract an individual background image from the taken image. The mode "Pre Image" or "Post Image" defines if the background image will be taken before or after the real image using the same settings. While taking the background image, the camera shutter is closed. The image background subtraction eliminates camera background pattern or memory images.

#### Background Correction

subtracts a specific amount of counts from the current image. The background is calculated using the lowest intensities on the image.

# Flatfield Correction

corrects an uneven image caused by the vignetting of the lens

#### Image Accumulation

With activated image accumulation more than one image is taken and added on top of the previous one. This allows a smoother background and offers a better cosmic suppression (if enabled). Up to 9 images can be added.

Enable the cosmic suppression to avoid spots caused by cosmic rays during longer exposures. Clicking on the second button will open the parameter box to adjust the image processing filters. For save cosmic suppression use about 50% strength. Increasing this value will increase the risk that real small spots will be treated as cosmics.

Since luminescence is measured no filter is necessary. That requires the selection of the empty / photo position in the emission filter settings. To ensure a proper energy calibration check the wavelength mentioned in the input box (560nm for firefly luciferase).

60	Emission Filter
Acuistion	Wheel: Wheel A 👻
	Filter: Photo 💌
	$\frown$
Post Processor	
se 1	
Emission	Photo
	Spectral Peak
	Wavelength [nm]: 560

To close the luminescence parameter wizard select **Frish**.

The summary of the new created luminescence measurement is shown in the right column:

Image: marked state       Image: marked	Luminescence	Aquisition Settings Exposure Time (s) 20     Binning x-Binning 4 y-Binning 4 Post Processor Cosmic Supersion enabled Emission Filter Name Photo Riter Wavelength (nm) -1 Spektral Peak (nm) 560
	Image: Add mark       Edit       Image: Down       Image: Down       Image: Down       Image: Down       Image: Down       Image: Down       Image: Down	Add     Luminescence       Edit       Lup       Down       Delete

To add a photographic exposure to the list of measurements select <Add>, <Photo>.

Edit Template		X
Setup Name	Data Aquisition Edit Template	indige
Data Aquisition Data Aquisition Sample Size Cegion Settings	Ad     Luminescence       Photo     Luminescence       Fluorescence     Fluorescence       Transillumination     Zolete	Aquisition Settings     Exposure Time (s) 20     Binning          x-Binning 4         y-Binning 4      Post Processor     Coartic Supression enabled     Emission Filter     Name Photo     Filter Wavelength [rm] -1     Spektral Peak [rm] 550
	< Back	Next > Finish Cancel

The default parameters (100ms exposure time, 10% illumination intensity) are suitable for most applications. Disable all post processors in the corresponding section. The "close iris" feature improves image quality (depth of focus). This feature is not available with in the NightSHADE evo

To close the photographic parameter wizard select **Ense**.





Aquisition	Exposure and Illumination Exposure Time [s]: Illumination Intensity [%]: 10	Aquistion	Post Processor Cosmic Suppression: Disabled Bignd Correction: Disabled Flatfield Correction: Disabled Enable 3D Filter: Disabled Image: More
Quick Check	<back next=""> Finish Cancel</back>	Quick Check	<back next=""> Finish Cancel</back>

For the sample size definition dialog box select **\_\_\_\_**.

	Sample Size New Template		indice
Setup Name		Setup Sample Size	
Data Aquisition		Sample Size [mm]:	150
2		Sample Height [mm]:	20
Sample Size		Comment:	
1			Quick Check
Region Settings			
		Preset	
			Load Store

You can either select an already existing format using the load button or define a new sample format. The following parameters are required:

#### • Sample Size:

The sample size refers to the image size (width) in mm. To make sure that the sample will be measured completely it is recommended to define the image size a little bit larger than the actual size of the sample (hier ist bei der Shade doch egal, was man einträgt, sollen wir das nicht ändern?)

#### • Sample Height:

The sample heights refers to the focus height. If you are measuring flat samples (gels, blots) located on the base plate the sample height is 0 mm. If elevated samples are measured (e.g. animals or plants) the sample height has to be set to a relevant range (e.g. height of a special leaf). If you are analyzing animals, it is advisable to use half the height of the animal as sample height. If you are working with sample holders or other extra parts (e.g. transilluminators), you have to take their height into account when calculating the sample height.

#### • Comment:

You can enter a comment on the sample format which describes the format.

The Quick Check can be used to start an acquisition using the entered values. The result is shown in a preview where you can either check the focus or whether the entered sample size is adequate for the acquisition of the sample.

If the focus is not OK it can be adjusted by manually adjusting the focus ring at the lens.

To go to the next page select  $\square$ 

New Template			X
*	Region Settings New Template		indige
Setup Name			
		Region of Interest	
Data Aquisition		X - Range Min [pixel]:	1
		X - Range Max [pixel]:	1024
		Y - Range Min [pixel]:	1
Sample Size		Y - Range Max [pixel]:	1024
24		s	et Maximum Frame Size
Region Settings		Multi Sample	
		Y - Images:	1
		A inages.	•
		Y - Images:	1
		<back next=""> Fit</back>	nish Cancel

On this page you can define the setting for the "Region of Interest" as well as the "Multi Sample" parameters.

By using the "Region of Interest" a section of the CCD chip can be defined to get faster framerates. By default the whole chip area from 1 to 1024 for X and Y is used

The command "Set maximum Frame Size" will adjust the frame size according to the selected binning factor since some cameras cannot handle full frame at different binning factors.

The MultiSample feature allows the possibility to split the image into image sections. Each section is treated as a single sample identity. This feature might be useful for animal imaging in case that more than one animal at a time is measured. The X-Images and Y-Images define the amount of samples in X and Y direction. This feature is limited to 5 images in each direction (max 25 sample-ID's per measurement). Be aware that working with individual images will also require to run individual reports which is not useful for some applications.

#### Multisample X1 and Y1



#### Multisample X1 and Y5





In case of multi sample the required identities will be defined before starting a measurement.

To close the application wizard select **Email**.

## 4.2.4 Defining a Controller Template

When creating a controller template the workflow is about the same as with a measuringtemplate. . But instead of defining measurement settings controller templates are used to control additional devices such as the LED panels in order to simulate environmental parameters (e.g. day & night cycle). Furthermore control templates are used to define pre- and postmeasurement settings such as "Daylight off and on" which allow to interrupt the light simulation and go back to its original setting after the measurement.

A controller template consist of a list of single commands. This list will be splitted in its single commands when executed using the so called scheduler. Therefore it is possible to perform measurements inbetween

To set up a new controller template refer to **<Applications & Templates> <Measuring Templates>**.Select **<New Controller Template>** from the **<Controller Templates>** context menu.

Ava 	ailable Te	mplates er Templates	
<u>آ</u>	Mei	New Controller Template	
		Expand Collapse	
		Properties	

indiGO starts the wizard. In the first window the controller template can be named and a descriptive comment be added

*	Setup Name New Template		indi
Setup Name		Template	
Command Set		Template Name:	Test Template
		Comment:	

Select **\_\_\_\_\_** to define the control workflow.

Different kinds of control actions can be performed. Please note that depending on the system configuration some of the selections might not be available (e.g. due to a missing LED illumination).

Template		
ø	Command Set New Template	indi
Setup Name		
124	🐣 Add 🔻 🌞 Artificial Daylight	Estimated Start of Execution
	Artificial Davlight	t0 + [h:mm:ss] 0:00:00
w	Archiclar bayingiran	Artificial Daylight
Command Set	Daylight Off	Intensity Left [µE] 8
	Daylight On	17
		13
	Illumination	13
	Heater	Intensity Right (µE) 8
	Relay Out	17
		13
	Delay	13
	More	Sample Changer
		7 Auia
		2-7643
	Banat	Halogen
	Report	Positioning Table
	< Back	Next > Finish Cancel

To turn on the LED illumination (Option) select **<Artificial Daylight>** from the list. Now the intensities of the different wavelength can be adjusted

(C)	Artificial Daylight	
Artificial Daylight	Channel — Panel 1 —	Panel 2
	green [µE]	8
	red [µE] 17	
	blue [µE] 13	
	Far Red [µE] 13	
		<ul> <li>✓ Syncronize Panels</li> <li>✓ Use Energy Values</li> </ul>

Adjust the required intensities and click on **read** to accept the parameters. The control command shows up in the work flow list.

v Template			-
\$	Command Set New Template		indice
Setup Name			
1000	🝳 Add 🔻 🗄	+ Artificial Daylight	Estimated Start of Execution
			t0 + [h:mm:ss] 0:00:00
- W	Edit		<ul> <li>Artificial Daylight</li> </ul>
Command Set			Intensity Left [µE] 8
			17
	Let Up		13
	T Down		13
	- Domi		Intensity Right [µE] 8
			17
	X Delete		13
			13
	Report		
		< Back	Next > Finish Cancel

After setting all commands, close the dialog box and save the template by clicking on Friend

Daylight Intensity (option)

This command sets the LED illumination panel to a defined intensity.

- Daylight Off (option) Turns off the LED Panels. The previously used intensities are temporarily saved for future use (when using "Daylight On"). Should be added to a project as Pre-Measurement action to make sure the artificial daylight is off while measuring.
- Daylight On (option) Turns on the LED panel using the last used intensities saved by the "Daylight Off" command. Should be added to a project as Post-Measurement action to make sure



the artificial daylight is turned back on to the previously used intensity after a measurement.

• Illumination

Controls the intensity of the white or infrared (option) LEDs in the dark room for photographic exposures.

100	Illumination Properties	
lumination	Output Level [%]:	
20		_

• Heater

Sets the target temperature of the temperature controlled measuring table (Option).

Heater	Heater Properties
	Temperature (*):

### • Relay Out

Controls the power socket inside the dark room (Relay 1). Relay 2 is not used with this instrument.

(O).	Relay Output Properties	
Relay Output	Active Relay: Rela	e1 •
	Value: Lo-4	off 🔸

• Delay

Defines a waiting time within the control sequence. When inserting a delay command the relative time of the control sequence (t0) will increase. The delay time can be inserted either in seconds or in h:m:s format (hour : minute :second).

October Delay	Delay State Properties			
Delay State	Delay Time [s]:			
	Delay Time [h:m:s]: 0:00:01			
Execute	< Back Next > Finish Cancel			
Execute				

-	🔆 Artificial Daylight	Ξ	Estimated Start of Execution
	🖑 Delay 180 [s]		t0 + [h:mm:ss] 0:03:00
	🔆 Artificial Daylight	Ξ	Artificial Daylight
			Intensity Left [µE] 0
			0

# Sample Changer (option)

Moves the sample changer to a defined position.

Controller Event	
Sample Changer	Sample Changer Properties Changer Postion: Postion 1 Postion 3 Postion 4 Postion 5 Postion 6
Execute	< Back Next > Finish Cancel

### • Halogen

This command turns on the excitation light of the fluorescence illumination..

101	Halogen Properties	
Halogen	Output Level (%i):	

• Positioning Plate (available with older NightSHADEs only) The positioning plate is an optional table to move the sample in X and Y direction. The required position can be entered in mm or a predefined position can be selected from the dropdown list.



Controller Event	
0	Positioning Table Properties
Postioning Table	X-Position [mm]: 56
	Y-Position [mm]: 58
	Stored Positions:
	MTP 24 well A1
00	
Execute	< Back Next > Finish Cancel

# 4.2.5 Defining a Project

To define a new project refer to **<Project & Analysis> <Available Projects>**. Select **<New Project >** from the **<Available Projects >** context menu.



Projects are used to define non measurement-specific parameters, like user access rights, scale settings, etc.

To create a new project indiGO starts the wizard:

#	Project Name New Project		indige
Project Name		Project	
Mentiny Namina		Project Name:	My first Project
Č.		Comment:	Just e comment
Aquistion Settings		Import Project	
*			Import Project
Analyse Sattings			
S Ibe Date			

The following parameters are necessary:

• Project Name:

Enter the project name under which the project is to be saved. Due to Windows file and path name conventions special characters such as < > ? " : | / / \*. are not allowed.

• Comment:

Enter a comment which describes the project.

To go to the next page select  $\square$ 

*	Identity Naming New Project		indice
Nget Nane		Identity and Sample Rames	
Vietty Names		Prefixi	sample
A.			Tinckole Year
Aquetion Settings		Suffix:	test
*		fieuit:	sample-[580]-2010-04-test
Analyse Settings		Create Identities	
Seer Rights		Number of Sdentities to Creater	1

On this page the number of sample identities and the sample names can be defined. A prefix, the current year and month, a 3 digit sample ID counter and a suffix can be specified if required. The resulting sample ID name can be seen under **"Result"**. The box **"Number of Identities to Create"** defines the amount of sample identities which will be created. More ID's can be added to the project later, if necessary.

To go to the next page select  $\square$ 

Choose the "Acquisition Settings" used for the project.

\$	Aquisition Settings New Project		indige
Project Name		Templates	
identity Naming		Pre Measurement Steps:	None •
An inter Settere		Measurement:	Acceleration
Aparta analys			G Edt
Analyse Settings		Post Measurement Steps:	None 💌
8			GgEdt
User Rights			
		< Back Next >	Finish Cancel

a "**Pre Measurement**" and a "**Post Measurement**" can be defined to be executed before and after the measurement. Under "**Measurement**" select the required measuring template.

To go to the next page select  $\square$ 

۲	Analyse Settings New Project		indi
Project Name		Photo Display Range	-
1		Diable Default Settings	
Identity Naming		Use Coston Settings	
075		Maximum Value:	
Aquation Settings		Pleanum Volkae:	
bi.		Data Range	
-		Use Default Settings	
Analyze Settings		🙁 Use Coston Settings	
ci i		Maximum Value:	
User Rights		Selection value:	

The display ranges for the b/w photographic and colored signal (data) image are defined on this page. If "**Use Default Setting**" is selected the system will calculate default values based on the photograph and the measured data. Individual settings for the minimum and maximum values can be selected in the "**Use Custom Settings**" mode.

To go to the next page select  $\square$ 

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On the last page users can be assigned to the project. The projects creator automatically receives ownership of the project. Other users can be added to the project. To create new users refer to the indiGO user manager.

All defined users are listed in the left column and can be added to the project using the add command (). All users assigned to the project are listed in the right column; Users can be removed using the remove command ().

Users assigned to the project are authorized to accomplish measurements for the project. Users, defined as "**users**" can't change the measurement settings for the project. For users defined as "**guests**" the access to measure is denied.

To complete the project definition wizard select **Finit**.

### 4.2.6 Starting an Acquisition

The system can carry out measurements on demand or scheduled using a time table.

First we will show here how to create an acquisitions on demand. Further details about how to use the scheduler can be found at the end of the chapter.

To start an acquisition refer to <Acquisition & Scheduling> <Start an Acquisition>.

The acquisition wizard will be shown:

Start an Aquisition	5	indige
Selec	t Project and Identity to Aquire	
Acqui	Service Training-001-Test	Multi Sample  Tools
	Measurement Settings	Init Wheel
	Pre Controller Steps	Load Slider
	Post Controller Steps	Quick Check
	< Back	Start Cancel

The project and identity might be pre-selected. Otherwise select the project and the identity from the provided drop down lists.

By selecting Quick Check a photographic preview of the sample is created using the project settings

This preview might look as follows.



If the "Auto Loop" checkbox is activated the camera will take an image continuously. This might be useful for sample positioning or when adjusting the focus at the lens.

To focus the image just turn the manual focus wheel located next to the camera on the front left side. Note that it might take some time until the new image will show up on the screen.





To start a measurement select set. Upon completion of the measurement, the data is automatically stored. The data is displayed by double clicking on the sampleID.



The measurement consists of a photographic image in grayscales and the luminescence image in pseudo color.

# 4.2.7 Image Sequences

In addition to single measurements indigo also offers automatic sequence measurements. These kind of measurements allow automatically triggered measurements in a predefined schedule.

To start a sequence measurement refer to **<Acquisition & Scheduling>**, **<Create a new Sched-ule>**.

The sequence wizard will be shown:

Create a new Schedule		×
		indige
	Select Project and Identity to Aquire	
	Sample-001-2010-07-Test	Multi Sample
	Advanced	Tools
	Aquisition Settings	Init Wheel
	Pre Controller Settings	Load Slider
	Post Controller Settings	Eject Slider
		Quick Check
	< Back Nex	t > Cancel

First of all specify the type of scheduled event. To set up a scheduler for measurements choose "Schedule Measurement", to set up an schedule for controll actions (lights on, mains switch, etc.) choose "Schedule Controller Template".

If choosing the measurement mode, first a project and sample ID has to be selected. In case of the controller template mode, the corresponding controller template has to be selected from the list.

Create a new Schedule	
	indige
	Controller Template to Schedule
	Turn Light on
	< Back Next > Cancel

Create a new Schedule	×
	indige
Contents to Schedule	
Schedule Measurement     Schedule Controller Template	
< Back 1	Next > Cancel

Clicking on leads to the following dialog box.

reate a new Schedule	
	indice
	Start Execution
	Mittwoch , 16. Mai 2012 🕶 15:00:00 🚖
	Start Straight Away
	Execution Calendar
	Overall Cycles: 20
	Delay/Cycle Time: 2 Minutes
	< Back Finish Cancel

In this dialog box the starting time for the measurement- or control sequence can be defined. Alternatively "Start right away" can be selected by checking the box. This will enable an immediate start when is clicked. All other events will be relative to this starting time.

In the "Execution calendar" the total number of measurements / sequences as well as the interval time can bespecified. Clicking on will close the sequence wizard and the sequence is handed over to the scheduler.

Now the sequence is active and the instrument controller is taking care of the measurements. It's possible to evaluate other measurements or even exit indigo. Only the instrument controller must be running. This is the case as long as the indigo icon is visible in the taskbar.

To check the scheduler click on Oseneduce. The scheduler will show up displaying all queued jobs with absolute and relative times.

Instrument Given State Instrument Camera	State Scheduler				indig	Ċ
Scheduler	Delete	Date	Time	t0 + 🛦	Comment	•
	Delete All	05/16	3:00:00 PM	0:00:00	Prj: Dummy / ID: 002-T.	
	Delete Alt	05/16	3:02:00 PM	0:02:00	Prj: Dummy / ID: 002-T.	
		05/16	3:04:00 PM	0:04:00	Prj: Dummy / ID: 002-T.	
		05/16	3:06:00 PM	0:06:00	Prj: Dummy / ID: 002-T.	
		05/16	3:08:00 PM	0:08:00	Prj: Dummy / ID: 002-T.	Ξ
		05/16	3:10:00 PM	0:10:00	Prj: Dummy / ID: 002-T.	
		05/16	3:12:00 PM	0:12:00	Prj: Dummy / ID: 002-T.	
		05/16	3:14:00 PM	0:14:00	Prj: Dummy / ID: 002-T.	
		05/16	3:16:00 PM	0:16:00	Prj: Dummy / ID: 002-T.	
	? Info	05/16	3:18:00 PM	0:18:00	Prj: Dummy / ID: 002-T.	
	4 Peload	05/16	3:20:00 PM	0:20:00	Prj: Dummy / ID: 002-T.	
	······································	05/16	3:22:00 PM	0:22:00	Prj: Dummy / ID: 002-T.	
	t0: 3:00:00 PM	05/16	3:24:00 PM	0:24:00	Prj: Dummy / ID: 002-T.	
		05/16	3:26:00 PM	0:26:00	Prj: Dummy / ID: 002-T.	Ŧ
	t0: 05/16/2012	•			- F	

Job details can be seen by doubleclicking the corresponding item in the list.

05/16 3:00:00 PM	<b>×</b>
Aquisition Settings	~
Exposure Time [s]	180
Binning	
x-Binning	10 ⋿
y-Binning	10
Post Processor	
Cosmic Supression	enabled
Emission Filter	
Name	
Filter Wavelength [nm]	• 0



# 4.3 Image Processing and Evaluation Functions

# 4.3.1 Load Images

During the first acquisition images have been created and stored in the project folder. To re-open the previously taken images refer to the project folder first and select the appropriate identity. All identities associated to the project are located in the project folder. The measurements including the images are stored under the context of the identity and can be opened by either double-clicking on the identity or using the identity context menu.

#### Please note:

The contents of the project folder shown depend on the user account, based on the indiGO user access management.

The identity number is displayed on top of the data view. If several sample IDs are open, a cardfile tab for each identity is displayed, each cardfile tab has its own color.



Below the identity number a row of index cards is shown containing the measurements acquired. The last acquired measurement is shown on the very right.

Using the arrows you can scroll back and forth within the measurements or the identities.

To display a complete overview of the images, the so-called **"thumbnail view"** select  $\mathbb{B}$ . To close the identity select  $\mathbb{X}$ .

Using the **<create movie>** command in the **<Report>** menu a movie of all images in the current identity can be created.



# 4.3.2 Image Window

After an acquisition or the opening of an identity the measurement and the photographic image are displayed. By default the photographic image is overlaid by the measurement or signal image using pseudocolors.



At the left margin of the screen you see the intensity scale and histogram of the photographic image, together with the associated sliders. These sliders can be used to change the brightness intensity of the photographic image. The top slider defines the upper brightness value, whereas the bottom slider defines the lower brightness limit.

At the right margin of the screen you see the intensity scale and histogram of the measurement, together with the associated sliders. With these sliders the scaling of the pseudocolor signal image can be set. The top slider defines which intensity value has been assigned to the highest color value. Values above this threshold are not displayed (transparent area). The bottom slider defines which intensity in the signal image is displayed using the lowest color value. All values below this threshold are not displayed.

To modify the scale move the slider to the position required. Moving the mouse to the center of both sliders a center mark is displayed which enables you to move both sliders at once.

To enter a certain value rightclick on the slider and enter the new value in the input field.



cts

The system offers the possibility to zoom the histogram by using  $\bigcirc 3277 \bigcirc 327$ 

\$ <u>R</u> eload Settings	The scaling method can be changed via the context menu which can be
Scale Linear	invoked by clicking on the arrow next to the @ r icon
Scale Log. Base 10	invoked by clicking on the unow next to the States income
Scale Square Root	
Scale Log. Base e	
Align None	
Align Best Fit 1	
Align Best Fit 2	
Align Best Fit 3	

The following options for the scaling are available:

- Linear scaling
- Logaritmic scaling (base 10)
- Square root scaling
- Logaritmic scaling (base e)

\$	Reload Settings
•	Scale Linear
	Scale Log. Base 10
	Scale Square Root
	Scale Log. Base e
	Align None
	Align Best Fit 1
	Align Best Fit 2
	Align Best Fit 3

In addition different autoalignment preset features (Best fit 1 to Best fit 3) are listed to adjust the min / max value based on the histogram.

At the bottom margin of the screen a transparency slider is available. This slider can be used to define the transparency of the overlay. If the slider is in the 100% position, the overlay is completely opaque, whereas the overlay is fully invisible if the slider is set to the 0%.



Changes of the intensity scaling or the color threshold can be applied easily to all other measurements within the current identity using the Appy to identity command. This simplifies the post run changes on measurements, as it is not necessary to modify every single image.

If the current changes should be applied to all identities measured within the same project, the **Apply to Project** command can be used.

The command has to be confirmed and can be parameterized using the dialog shown below.



The following options are available:

<Apply Image and Contrast settings> will apply the image and contrast settings from the current image to all other measurements in the project.

If **<Use current Settings as default for new Measurements>** is selected, these settings are also valid for all future measurements in the project.

The selections **<Annotation Layer>** and **<Manually Layer>** will copy the areas and annotations to all other measurements within the project.

The **<Run Peak Search>** option will perform a peaksearch on all other measurements in the active project using the current settings.

The process will be started by selecting  $\square$ .

This opens all data of the current project (Apply to project) and applies the changes. To get back just close the data without saving.

#### Please note:

Depending on the selections and the amount of data this function may take some time to accomplish.



# 4.3.3 Line Plot

For further analysis of the measurements it might be helpful to create a 1D-line plot of the measured data. This function is recommendet to evaluate the measuring data.

### To use this function select **1**

An additional window is opened where the line profile created from a given lead is displayed.



The pixel intensity along the selected path is displayed as a line diagram. The average width of the lead can be selected. Moving the lead updates the line profile.

# 4.3.4 Surface Plot

To get a 3D surface plot select **<Surface>** from the **<Report>** menu.The surface can be used for 3D visualization of the measured data.



To create a 3D surface plot from the photographic image only please refer to associated command. The same applies if you want to create a 3D surface plot from the measurement data only.

From the dialog **<Photo Properties>** or **<Data Properties >** select **<Range>** and start **<Surface>**.

Photo Properties	Data Properties
Photo Unit Range	Data Unit Range Tilter
Current Background Photo	Data
photo.001 • LUT	lumin.000 • LUT
Surface	Surfece
Photo Display Range	Data Range
Maximum Value: 7634.99	Maximum Value:
Minimum Value: 1051.68	Minimum Value: 25
	Overlay Level: 83.92
CK Cancel Apply	CK Cancel Apply

To change the perspective of the 3D-view pick the graph and move the image to the position required. To zoom in use the mouse wheel.

To animate the surface double-click on the graph.

### Please note:

The windows displaying the surface can be resized.

## 4.3.5 Peaksearch

Once the image has been loaded and the color view has been set up, you can start to analyse the evaluation.

In the first step the desired areas has to be marked; now indiGO can calculate the intensity values.

The easiest method to find evaluation relevant data is to use the peaksearch.

Using the peaksearch all areas lying above a threshold value will be marked. With this method it is possible to evaluate different measurements under exactly identical evaluation conditions.

To start the peak search select <a>Peak Search</a>.

The displayed overlay properties window gives an overview of all automatically found peaks in the current image.



To run a new search select <u>Search</u>.

The following parameters can be edited:

#### • Peak Threshold - Based on

Using a droplist different search algorhytms are available:

Area

The area of every possible spot is analyzed and if the area is larger than a size specified in a parameter box below, the spot is validated as a peak. This is useful to get rid of smaller, unwanted spots.

Boundary

The most common algorithm is the boundary discrimination. The boundary of every possible spot is calculated and if the boundary is larger than a specified size from a parameter box below, the spot is validated as peak. This is useful to get rid of smaller, unwanted spots.

#### Noise detection

A special noise analysis filter is used to analyse the image and identify the peaks. For this method no parametars are required.

#### None

No discrimination method is being used. Every spot which is above the given lower threshold will be treated as peak.

### Advanced - Remove Areas inside Areas

If this option is set areas which are located inside other areas will be declined.

### • Maximum Areas

This parameter defines the maximum amount of peaks. If this amount of peaks is found the peak search stops despite of being completed. This will protect from long waiting times in case a very noisy image is used with hundreds of possible peaks.

To start the peaksearch select Run Search.

## 4.3.6 Define Areas Manually

The **"Manual Areas"** are available via the command **<Manually>**. They are used for the manual definition of integration areas. Here you have the possibility to define free forms, such as circles, rectangles or polygons.

To start this function select Manually .

A window is opened where an overview of all currently defined areas is displayed. If you have not defined any areas the list will be empty.

Annotation Manually	Peak Search	1:54:08 AMJ			
View  Import	Label	Index	Area [mm²]	Counts [cts]	
Ling Export					
Down     Delete					
Hide Layer					
			ок	Cancel Ap	ply



If you want to create new areas open the context menu by using the arrow on the button **<Manually>** and select the required shape (rectangle, circle or free-hand shape) from the context menu.

With the mouse curser shown on the image the selected shape can be drawn by dragging into the required area of the image.



To define a freehand shape circle the desired area with several clicks on the left mouse button or hold it down completely (pencil mode). A double-click will end the procedure.

The painted shape will be connected automatically and the freehand shape will be closed by a frame.

If you click on an area that has already been created, you will see "handles" around the edge of the area. The area can be resized by dragging the "handles" to the desired size.



The shape can be moved to another position. To do this you have to move the mouse to the boundary of the shape. The mouse pointer will be changed automatically to a hand cursor. If the hand cursor is displayed press the left mouse button and drag the shape to the required position. When pressing **<CTRL>** and

**<Shift>** while moving the selected area this area will be duplicated. The copy mode is indicated with a little "+" next to the mouse cursor symbol. To select more than one area just press the **<CTRL>** key while selecting the area with a mouse click or drawing a box around the required areas.

	Duplicate
	Set Background
	<u>H</u> ide
	Lock
¥	Cut
	Сору
8	<u>P</u> aste
×	Delete
	Select All

A context menu for all shapes is available. To open the context menu select the shape and right-click inside the shape.

For editing the standard clipboard commands are available

To use the object for background subtraction select **<Set as background>**.

An object defined as background area is identified by a checkmark in front of the context menu entry. In this case the average intensity of the area is defined as background and subtracted from the intensities of the other areas in the report.

To create a copy of the current area select **<Duplicate>**.

To lock the whole layer, use the command **<Lock>**.

A locked area cannot be moved any longer unless it's being unlocked through the **"Overlay Properties" <Lock Layer>**.

The layer becomes invisible by selecting the command **<Hide>**.

To delete all areas refer to "Overlay Properties" <Delete All>.

eak Search Resu	lts —				
View 🔻	Label	Index	Area [mm²]	Boundary [mm]	Counts [cts]
Import	○P:1	1	191.102	53.4341	19580.9
Import	○P:2	2	187.111	53.7835	3912.06
Export	○P:3	3	1.60933	5.0871	258.5
▼ Down					
X Delete Delete All					
Hide Layer					
Lock Layer	•		111		

Further commands for the area management are available via the contect menu of the area items list.

To export areas the **<Export>** command from the **"Overlay Properties"** can be used.

To import areas the **<Import>** command from the **"Overlay Properties"** can be used.





# 4.4 Creating a Report

# 4.4.1 Creating a Rich-Text-Report

indiGO offers the possibility to create and display measurement reports. The output can be adapted to the user's requirements.

Using the **<Rich-Text-Report>** command in the **<Report>** menu will start the Rich-Text-Report-Creation-Wizard.



To create a report, follow the instructions given by the wizard. In brief the following options can be set.

#### • Create a Front Page

This will create a report front page. The contents of the page, like your company name, address or logo can be setup through the indiGO options.

Export indice	Export indice
Report Contents  Croate a Font Page  Croate a Font Page  Croate a Font Page  Tages  Tage Tages  Tages  Tage  Tages  Tages  Tage  Tages  Tages	Units Area && Elongating @ Milmeter [m] O centimeter [m] Measured Data @ Counts [gns] @ Photons [ph(s] @ Energy [n%]
< Back Next > Cancel	<badk next=""> Cancel</badk>

#### • Export Images

If this option is set, all overlay images will be included into the report. You can also select, whether the overlay areas will be included into the images. The meta data of each image will be displayed next to each image.

### • Units Area & Elongating

Different units for the areas and the elongatings can be selected. Millimeter and centimeter can be selected for geometrical values (e.G. area or size of an area).

### Measured Data

Different units for the measured data can be selected. The intensity data can be displayed in counts (raw values), the emitted photons (ph/s) or the

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emitted power (nW). The photons as well as the power values are besed on a calibration using a light standard. This calibration is only valid, when the sample is placed on the base plate and not higher than 50mm. In case of a different sample size height or position, the calibration is not valid and wrong results will be calculated using the default calibration. In this case a dedicated calibration must be performed using the light standard. This calibration can be applied to luminescence measurements only.

## Rich Text Export Options

The report can be opened to be printed or stored into a file for further usage.

RichText Export Options
Doen ReportView to review the Export     Stent export to File
Filename:
be possible a l'import U

### 4.4.2 Excel Export

indiGO provides a very comfortable report function, the possibility to create Excel exports. In an Excel export, a complete measurement report, including all images, analysis data and in addition, diagrams containing a graphical view of the measurement results can be created.

To use this function Excel 2013 or later is recommended. Other spreadsheet programs are not supported.

Using the **<Excel Report>** command in the **<Report>** menu will start the Excel-Report-Creation-Wizard.



To create a report, follow the instructions given by the wizard. In brief the following options can be set.



### • Create a Front Page

With this command you can create a report front page. The contents of the page, like your company name, address or logo can be setup through the indiGO options.

Export	indige	indice
	Report Contents	Units Area && Elongating
	< Back Next > Cancel	< Back Next > Cancel

### • Export Area Details & Graphs

You can select either bar graphs or line charts to be created.

• Export Images

If this option is set, all overlay images will be included into the report. You can also select, whether the overlay areas will be included into the images.

### • Units Area & Elongating

Different units for the areas and the elongatings can be selected.

Measured Data

Different units for the measured data can be selected.

• Excel Export Options

The report can be opened to be printed or stored into a file for further usage.

export	indice
	Open Excel to review the Export     Open Excel to review the Export     Open Excel to File     Element
	C:\20100805115023.xisx >
	< Back Next >>>> Cancel

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2 205,39 55,11 0,21 2019,13	31	1 250							2	205,39	55,11	0,21	2019,13			
32 3 180,05 52,4 0,16 1372,57	32	2							3	180,05	52,4	0,16	1372,57			
33 200 4 163,08 51,66 0,14 1098,31	33	3 200 -	×.						- 4	163,08	51,66	0,14	1098,31			
34 5 97,25 63,71 0,33 573,8	34	4						. f	5	97,25	63,71	0,13	573,8			
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The spreadsheet will contain one to three worksheets depending on the selected options:

• IndiGO

The **"indiGO" sheet** contains the front page

• Analysis

The **"Analysis" sheet** contains all measurement results and the diagrams, depending on the settings.

• Images

The **"Images" sheet** includes the overlay images and all additional measurement information e.g. file name, date of acquisition, sample description and measurement parameters, to ensure reproducibility of the measurements.
# 4.5 Using the Scheduler

Control sequences like sunset and sunrise or measurements for selected identities can be controlled via a scheduler. You can think of the scheduler as a simple list of time controlled events.

To start an acquisition, refer to <Acquisition & Scheduling> <Create a new Schedule>.



The system will start the **<Create a new Schedule>** wizard, which allows you to select the contents you want to schedule first.



To schedule measurements select **<Schedule Measurement>** and chose **wet>** to continue.



On the next page you can select the project and the identity you want to measure.

Using the **<Advanced>** or **<Tools>** commands you can validate the acquisition, pre- and postcontroller settings or initiate a **<Quick Check>** to create a test image.

Chose **letter** to confirm the selected options and continue.

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When selecting **<Schedule Controller Template>** a dialog box pops up to select the desired control sequence.



Chose **lext** to confirm the selected template and continue.

Create a new Schedule	×
	indice
	Start Execution
	Mttwoch , 16. Mai 2012 V 16:00
	Execution Calendar
	Overall Cycles: 20
	Delay/Cycle Time: 2 Minutes
	< Back Finish Cancel

On the next page the **<Start Execution>** time can be selected. In the sample above, the first measurement will be executed on Wednesday the 16th of May at 16:00:00. When "Start Straight Awyay" is checked the start time cannot be entered. In this case the action starts right after clicking on **\_\_\_\_\_\_**. All other repeats will start relative to this start time.

The **<Overall Measurements>** are set to 2 and the **<Delay/Cycle Time>** was set to 2, as well. Therefore the system will add two measurements into the scheduler. The first will be executed Wednesday the 16th of May at 16:00:00 and the second will be executed Wednesday the 16th of May at 16:02:00.

The measurements will be executed in the background by the scheduler and will be sorted automatically into the indiGO structured tree.

The contents of the scheduler can be verified or deleted via the **<View> <Scheduler>** command from the main menu of indiGO.



Instrument State Instrument Camera Scheduler	State Scheduler				indi	
	Delete	Date	Time	t0 + 🙈	Comment	-
	Delete All	3 05/16	4:00:00 PM	0:00:00	Artificial Daylight	
	Delete All	\$ 05/16	4:02:00 PM	0:02:00	Artificial Daylight	
		05/16	4:04:00 PM	0:04:00	Artificial Daylight	
		05/16	4:06:00 PM	0:06:00	Artificial Daylight	
		\$ 05/16	4:08:00 PM	0:08:00	Artificial Daylight	E
		05/16	4:10:00 PM	0:10:00	Artificial Daylight	
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		05/16	4:14:00 PM	0:14:00	Artificial Daylight	
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	Y Keload	4 05/16	4:22:00 PM	0:22:00	Artificial Daylight	
	t0: 4:00:00 PM	05/16	4:24:00 PM	0:24:00	Artificial Daylight	
		4 05/16	4:26:00 PM	0:26:00	Artificial Daylight	-
	t0: 05/16/2012	•				•

The scheduler shows the list of scheduled measurents and control commands with absolute starting time and relative time. The time of the first event in the list (when the scheduler was empty before) is treated as time zero.

To highlight an item just click on it. To highlight more than one item press the CTRL key while clicking on the items. A doubleclick on a single, highlighted item or clicking on reference will show detailed informationabout the event.

Ξ	Artificial Davlight	
	Intensity Left [µE]	20
		20
		20
		20
	Intensity Right (µE)	20
		20
		20
		20

Highlighted items can be deleted by clicking on <u>Delete...</u>. To delete the whole list of items click on <u>Delete Al...</u>.

To update the list, click on <u>release</u>. This will remove past events.

# 5 System Description

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5.2 Design of the Acquisition Device	79
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# 5.1 Intended Use

*NightSHADE evo LB 983* is a sensitive Imaging System for luminescence and fluorescence measurements to be used on the benchtop. It is used to acquire and evaluate images in the macro imaging area in a large intensity range.

Not for use in in-vitro diagnostic (IVD) procedures.

This instrument is not designed or intended for use with installations or equipment in hazardous environments. Servicing of the instrument must only be performed by Berthold Technologies Field Service Engineers or service staff authorized by Berthold Technologies.

NightSHADE evo LB 985N includes the following components:

- Acquisition device including the transport mechanism with camera and lens and the electronics. The high-resolution CCD camera and the special wide open lens guarantee excellent sensitivity and image quality.
- The control of the acquisition system and the image processing and evaluation take place under control of the effective **software system** *indiGO*. The easy-to-use and clearly structured Windows software covers the entire spectrum of image processing functions.
- The **PC system** including a standard computer (Pentium 4<sup>®</sup> or other processor) with high-resolution monitor. We recommend a resolution of at least 1024x768 pixels.
- Image output to any Windows printer (laser or ink-jet printer) or video printer.



# 5.2 Design of the Acquisition Device

The acquisition device includes the hardware and electronics required for image acquisition. It comprises the acquisition chamber (dark box) including the camera lift with camera, lens and filter wheel. The electronics and excitation light source for fluorescence measurements are accommodated in the upper part of the device. The ventilation cap is designed for drawing off the waste heat caused by the camera cooling.



#### Front side

- o Light-tight door with handle to close the acquisition chamber
- Door for loading and unloading the filter slider for fluorescence emission
- o Service flap for halogen lamp and excitation filter slider loading and unloading.
- o Operating status indicator
- Inside the *acquisition chamber*
- o Inductive sensor detecting the door position
- o Bottom plate (removable) to position the acquisition objects
- o 4 LED's illuminating the acquisition chamber
- o Connector field
- Liquid cooling connectors (for LED panel)
- LED panel control connector
- Outlet of the fluorescence light for connection of the illumination light guide with optical switch
- $\circ~$  4 fibre optic illumination light guides for fluorescence illumination (with -40 verion only)

#### Rear panel/Top part

- Connection ports
- On/off switch
- o Mains fuse
- Mains connector
- USB B port (in)
- USB A port (out for side view camera)
- Camera power outlet (for side view camera)
- Liquid cooling in/outlet (for LED panels)
- o Ventilation slits

#### At the side

- Carrying handles
- Camera bayonet for side view (left)
- Flange port for external feed through

#### Bottom

The NightSHADE evo stands on four feet; the height of these feet can be adjusted to balance uneven setup locations.



# 5.3 Acquisition Chamber

#### 5.3.1 Light-tight Door with Door Button

To close the acquisition chamber light-tight, the door frame is provided with an elastic sealing and a handle which, when turned by 180°, will pull the door into the frame and squeeze it against the sealing. An inductive sensor indicates if the door is closed correctly.

Always make sure that the door is properly closed. If the door is not closed and you try to take an image, you will be alerted by the program.

Images can be taken even while the door is open. The camera is insensitive to daylight, i.e. the camera will not be destroyed by overexposure.

### 5.3.2 Illumination of the Acquisition Chamber (Dark Box)

Four LED's installed on the walls of the dark box ensure constant illumination of the acquisition chamber. However, the illumination depends on the camera setting and with very low camera position it can be affected adversely. If necessary, an additional light source has to be installed (power is supplied via the outlet strip in the acquisition chamber). The illumination of the acquisition chamber is automatically controlled by the software.

#### 5.3.3 Connector Field

The connector field is located in the right rear corner of the acquisition chamber; it includes the following ports:

Round sockets:	Control connector for LED panels (left and right)	
CAN socket:	System connectors to control instrument op- tions (e.g. turn table)	100V / 240V 100V
Mains socket:	Mains socket enabled via software	- Prov.

The mains socket is controlled through the software (see chapter 4.2.4) To prevent exposure to dangerous UV light when using e.g. a UV transilluminator the socket will switch off while the door is open.

Max. power outlet is 200VA (AC 100 – 240V +/- 10% according to the supply voltage).



The CAN socket controls external system options such as the turn table (see chapter 10.1.2) or the temperature controlled measurement table (see chapter 10.1.2).

When no external equipment is connected the dummy plug (terminator) must be connected to this socket to ensure a proper termination of the internal bus.

outlet for the optional LED panels (see chapter10.1.1). Connect the LED panels only while the instrument is powered off to prevent electrical damage of

The two round connectors at both sides of the connection box are the power supply and control

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the LED panel.

5.3.4 Camera and Lens

*NightSHADE evo LB 985N* uses a slow scan CCD camera, hermetically vacuum encapsulated and cooled down by a Peltier element to -70°C (the operating temperature is reached after about 10 minutes).

To ensure the proper operation of the camera cooling the ambient temperature should not exceed  $28^{\circ}$ C.

The camera is connected with the electronics via two cables (voltage supply and USB communication) on its top side.

Focussing of the sample is done by turning the manual focus wheel of the lens, located at the front left side. The lenses are changeable using a quck change bajonet system. To mount the lens both red dots of lens and carrier plate must match.

#### Maximum sample size for standard resolution images:

Ikon (with 25mm lens) : 266 x 266mm at baseplate level up to 127 x 127mm (sample must be raised up).

With Macro lens: 130 x 130mm at baseplate level up to 60 x 60mm (sample must be rasied up).

#### Discharge electrostatics before taking out the camera

Clean the camera and lens only with special cleaning products from your photo shop (see page 115).

#### 5.3.5 Filter Wheel

The filter wheel is installed below the camera. The filters are located on specific filter holders. These holders can be slided into the filter wheel.

Please observe the correct installation direction (arrow on the filter wheel) when loading the filter. Please refer to the filter manufacturer's description. The filters should be secured from above against falling out by the snap rings which are included with the device. In the filter manager you can specify filter properties. Make sure the filter numbers printed on the filter holder matches the filter position indicated on the filter wheel. *Filter position 5 should remain empty for the acquisition of photographic images.* 



Do not reach into the filter wheel while the wheel is moving  $\rightarrow$  Risk of injury. Filters can be exchanged by manually moving the wheel while the instrument is turned off.







#### 5.3.6 Filter Slider



The filter slider can accommodate five excitation filters with a diameter of 25 mm (or 1 inch). It is located under the flap on the right top side of the instrument. A light blocking knob keeps out light to ensure best performance. To get access to the slider just remove the knob.

The filter slider can be ejected via the indiGO software. The eject command can be found under Staten Acquisition.......

Start an Aquisition	
	indice
Select	Project and Identity to Aquire
Acquis	001-Probexytz   Multi Sample ition Settings Tools
	Measurement Settings Init Wheel Pre Controller Steps Load Slider
	Post Controller Steps Eject Slider
	<back cancel<="" start="" td=""></back>

To eject the slider select **Eject Slider**.

When loading the filter please observe the correct installation direction (arrow on the filter wheel).

To reload the slider select Load Slider. After that the slider will be initialized by the instrument.

The Berthold Application Support is available to recommend filters when you are selecting the optimum filter combination for your fluorescence application.

#### 5.3.7 Fluorescence Illumination

The built-in halogen light source together with the filter slider create the excitation light for flourescence measurements. The light is guided into the dark box to the ring light (1). For alternative illumination devices using standard 15mm fibre optic connectors an auxiliary light output port (2) is located at the right side at the ceiling of the dark box.



When the ring (2) is in the lower position, the light is guided to the built-in ring light. When pulling the ring to the upper position an optical switch guides the light to the auxiliary light output port. The light guide is fixed in the socket using internal spring loaded balls. To remove it pull the light guide slightly.

A blind plug (3) is placed inside the light output to prevent outside light entering the dark box. Please alyways keep this rubber plug in place if the auxiliary port is not required.

#### 5.3.8 Bottom Plate

The bottom plate helps you to position the acquisition objects. It includes two bolt heads (1) on the front panel to help you position and fix the samples or sample masks accurately. In addition, there are four wells (2) for accurate positioning of transilluminators.



The bottom plate can be removed for cleaning, if liquid was spilled.

In addition, the bottom plate can be pulled out from the front to facilitate the loading of samples. When you push the bottom plate back in again, make sure that the plate snaps into place with an audible click.



#### 5.3.9 Top Part of Acquisition Device

The upper part of the instrument contains the camera, the fluorescence light source (option) and the complete electronics.

To open lift up the lid.

Inside 2 connectors are visible. One is for the cmare power supplky, the other is the USB dataport to control the camera. Remove the cables before removing the camera. The camera can be taken out be turning it counter clockwise until the two red dots are matching each other. Now the camera is unlocked and can be removed by lifting up the camera.



#### Please note:

The ventilation slits of the cooler must always be open!

#### 5.3.10 Connections at the Rear Panel

The power supply and device communication ports are located on the rear panel of the NightSHADE evo.

Plug the power cord into the mains inlet (1). The main power switch is located next to the mains inlet.

Connect the NightSHADE evo to the PC via the USB B-socket (2) on the rear panel using a USB cable (A to B).



The 7-pin Tuchel socket (4) is the power supply for the side mounted camera, the USB A socket (3) is used for the control of the side view camera.

The cooling liquid connectors are located on the lower side of the rear panel. These quick tube connectors connect the chiller to cool down the LED panels the inlet connector is located in the center of the rear panel.



# Darkroom (rear view)

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# 6.1 Graphical User Interface

The indiGO graphical user interface is clearly structured and offers a variety of setup options.

Measurements are automatically displayed as overlay.



# 6.2 Application Toolbar

🛞 Cancel Measurement | 🛐 Start an Acquisition... 🛐 Create a new Schedule... 🔘 Open OWL View... 🦉 Quick Check... 🦉 Device State... 🕓 Scheduler... 🔀 Report... 🔕 🧇 👰

On the toolbar you find the following commands:

#### Cancel Measurement

Will cancel the currently running measurement. The foolowing message will show up in the system taskbar:



To continue the operations in the scheduler click on <u>contrue</u>. Otherwise all actions are paused.

#### Start an Acquisition

Start a new measurement for the current selected or any other project.

- Create a new Schedule Create a new schedule for the current selected or any other project.
- Open OWL View
   Start the indiGO image viewer.

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- Quick Check
   Start the <Quick Check> utility to create a photo or run a measurement for test circumstances.
   Device State
  - Open a window to display the current state of the NightSHADE evo.
- Scheduler
- Open the editor for the scheduler
- **Report** Create a report for the current active project
- Help Search
- Search the online help
- Help
  Start the online help
  About
- Display program version and copyright information

# 6.3 Identity Context



On the left of the context bar the actual selected identity name is displayed. On the right are three quick commands available.

#### • Apply to Project

With this command the current image and contrast settings, as well as the overlay properties of the annotation and the manual layers can be applied to the current project. You also have the possibility to start the peak search for all measurements of the project.

To perform this operation, all images assigned to the selected project will be opened. According to the amount of images the operation can take some seconds up to minutes.

Apply Changes	-	
Im	age Properties to Apply to	Project.
	Apply Image and Cont	rast Settings
	Use current Setting	gs as default for new Measurements
	This Opti Application	on allows to overwrite the default on Settings for new Measurements.
Ov	erlay Properties to Apply to	o Project.
	Annotation Layer	
	Manually Layer	Applying 'Image and Contrast Settings' or running the 'Peak Search' may take a minute to
	🔲 Run Peak Search	process ? Any changes to the project will be temporary until stored.
		OK Cancel



#### • Apply to Identity

With this command the current image and contrast settings, as well as the overlay properties of the annotation and the manual layers can be applied to all measurements of the current identity. You also have the possibility to start the peak search for all measurements of the identity.

Apply Changes	×.
Image Properties to Apply to	o Identitity.
Apply Image and Cor	ntrast Settings
Use current Setti Use This Op Applica	ngs as default for new Measurements tion allows to overwrite the default tion Settings for new Measurements.
Overlay Properties to Apply	to Identitity.
Annotation Layer	Applying 'Image and Contrast
V Manually Layer	Settings' or running the 'Peak Search' may take a minute to process ? Any changes to the project will be temporary until stored.
	OK Cancel

#### • Store Identity

Save all changes made to the identity to disk.



# 6.4 Identities and Measurements

N I	Mou	ıse	1		V	Te	stm	ous	e1			V	uh	r c	2																																					•	Ŀ		x
V.	AII		N	1		1	Y	2	T	N	3		1	Y	4		M	5		N	r e	5	I	Y	7		M	8		V	9		V	10	T	M	11		M	12		N	13		N	14		1	.5	1		4	Ŀ		82
ß	•	-	4	•	₽		(	Э,	•	2	6	1 %	6			•	•	1	L R	9	(cu		ß	An	nota	tion	•		e N	lanu	Jally	•	<	Pea	ak :	Sear	ch	•	<b>%</b>	<b>s</b>   (	¥	Rep	ort	Ŧ			þ	Ę		ß	•	4		•	

• Top Row

Currently opened sample identities

- Center Row
  - List of all measurements belonging to the current selected identity
- Bottom Row
   Measurement or data toolbar

# 6.5 Contrast Slider

<u>Q</u> 19661 <b>Q</b>	With the contrast slider on the left side of the image the brightness of the photographic display is defined. The top handle sets the upper brightness value, whereas the bottom handle is for the definition of the lower brightness limit.
	The brightness can be scaled using several scaling algorithms and can be aligned automatically. These operations can be reached via the <b><photo prop-erties=""></photo></b> .
·	The <b><photo properties=""></photo></b> can be reached through a dialog or the <b><photo menu="" prop-erties="" quick=""></photo></b> .

cts

#### 6.5.1 Photo Properties Quick Menu

To access the drop down menu simply click on the arrow next to the icon shown below.

🙆 🔻

. 🔊	•   4 • •   🤤 🍕   61	%
2	<u>R</u> eload Settings	
	Scale Linear	
	Scale Log. Base 10	
	Scale Square Root	
	Scale Log. Base e	
	Align None	
	Align Best Fit 1	
	Align Best Fit 2	
	Align Best Fit 3	
_		

- Reload Settings
   Reload the last stored settings for the scaling and the alignment.

   Scale
- Scale

Scale the contrast of the photo using the selected algorithm.

 Align Align the contrast sliders using the selected algorithm.

#### 6.5.2 Photo Properties Dialog

To access the photo properties dialog simply click the icon shown below

B

Photo

The Photo page is used to select the active photo.

If a measurement contains more than one photo, the photo displayed with the measurement can be selected.

Photo Properties	<b>— X —</b>					
Photo Unit Rang	e					
Select Background	Select Background Photo					
photo.000	20100301005800.photo.000					
none						
none						
none						
none						
none						
none						
🔘 none						
none						
none						
	OK Cancel Apply					



#### **Reference:**

The real name of the photo is displayed as a link. In our example: 20100301005800.photo.000, where *201003* is the date the photo was taken. In our example: 03. March 2010. 005800 is the recording time. In our example: 12:58:00 AM. By following the link, the indiGO image viewer will be opened and the photo will be displayed.

#### • Unit

The Unit page can be used to setup the unit of the contrast slider



#### Range

The Range page can be used to setup the contrast slider. For further analysis a LUT and a surface is supported.

Photo Properties	×
Photo Unit Range	
Current Background Photo	
photo.000 👻	LUT
	Surface
Photo Display Range	
Maximum Value:	16349.3
Minimum Value:	2246.91
ОК	Cancel Apply

# 6.6 Data Slider

On the right margin of the screen you see the intensity scale of the measurement, together with the associated slider. With these two sliders you can set the scaling of the pseudocolor signal image and the lower threshold of the color view. The top slider defines the intensity value.

The top slider defines which intensity value has been assigned to the highest valued color. Values above this threshold are displayed in white.

The bottom slider defines which intensity in the signal image is displayed with the lowest valued color. All values below this threshold are not displayed.

#### 6.6.1 Data Properties Quick Menu

The **<Data Properties Quick Menu>** supports the the same functions as the **<Photo Properties Quick Menu>**.

#### 6.6.2 Data Properties Dialog

• Data

The Data page is used to select the measurements displayed. Each data image recorded can be tagged to be relevant and displayed along with the photo.

Data Pro	perties	X			
Data	Unit	Range Filter			
Select Overlay Data					
	lumin.00	00 20100301005800.lumin.000			
	none				
		OK Cancel Apply			

Range

The Range page can be used to setup the data slider.

Data Properties
Data Unit Range Filter
Data
lumin.000 ▼ LUT
Surface
Data Range
Maximum Value: 16036.6
Minimum Value: 7390.52
Overlay Level: 80.00
OK Cancel Apply

For further analysis a LUT and a surface is supported. In addition the overlay intensity can be set through **<Overlay level>**.



#### • Filter

The Filter page can be used to change or setup the filter properties used to render the overlay. This might be useful when more than one signal layer must be displayed in order to assign different properties to each signal layer.

Data Properties
Data Unit Range Filter
Data
[umin.000]
Filter Properties
False Color Table
CWL [nm]: 560 🔒
HBW [nm]: 260
T [%]: 100
OK Cancel Apply

The following options are available:

- False Color Table
   Activates the system default f
- Activates the system default false color table
- Record Filter
   Activates the filter properties used for recording
- **Custom Filer** Enables the user to setup a custom filter, using a center wave length and a bandwidth.
- **Custom Gradient** Enables the user to define a gradient using a center wave length.

a Properties		×
Data Unit Rar	nge Filter	
Data		
lumin.000	•	
Filter Properties		
False Color Table	•	
Ealse Color Table		
Record Filter		
Record Filter Custom Filter		
Record Filter Custom Filter Custom Gradient T [%]: 1	100	



# 6.7 Toolbar

| 🔍 🍭 | 61 % 🔹 🔀 | 🔊 🍋 | 🌽 Annotation 🔻 🗃 Manually 🔻 🧠 Peak Search 🔻 | 🚿 | 🔂 Report... 🔻 | 🔒 🖺 🜉 拱

- Decrease the zoom Used to decrement the zoom
- Increase the zoom
   Used to increment the zoom
- Current zoom Used to setup the zoom manually, displays current zoom factor
- Align Align image into the view; used to calculate a best fit zoom factor
- Undo (area editing)
- Redo (area editing) Used for undo and redo while area editing is active
- Annotation (Menu)

Create annotation areas

Clipboard		
Extra		
<u>R</u> ect		
Circle	- 18	
<u>F</u> reehand		
Line		
Arrow		
Ruler		
Text		





#### • Manually (Menu) Create manual areas



#### • Peak Search (Menu) Starts the peak search



• Peak Search (Command)

Peak Sea	rch	<b>—</b> X—
Peak T	hreshold —	
	Based o	n: Noise Detection
Advand	ced	Remove Areas Inside Areas
Limitat	ion	
2	You can setup Peak Search wi the Peak Searc	an upper limit to the number of areas the ll identify. In case of exceeding the limit h will stop.
	Maximum Area	s: 0
		Run Search Close



#### • Overlay Properties (Dialog)

The **<Overlay Properties>** dialog can be opened via the **<Annotaion>**, **<Manually>** or **<Peak Search>** commands

View 🔻 L	abel	Index	Area [mm²]	Boundary [mm]	Counts [cts
Import	⇒P:1	1	3.34589	10.3352	2.75536e+000
	>P:2	2	2.34034	6.86205	1.52798e+00
Export	⇒P:3	3	0.80874	4.14286	46967
0	⇒P:4	4	0.644129	3.36695	48063
Search	⇒P:5	5	0.533196	3.01404	47952
0	⇒P:6	6	0.425841	3.11917	40589
Up	⇒P:7	7	0.529617	3.28236	37126
	⇒P:8	8	0.268387	2.0424	23762
Down	⇒P:9	9	0.293437	2.07744	26198
Delete Delete All					
lide Layer					
Lock Laver 🛛 🕢			111		

- Line Plot Starts the Line Plot Tool
- Report (Menu)

😰 R	leport 🔻 📑 📑 📑
×	Excel Report
<b>*</b>	<u>R</u> ichText Report
	<u>S</u> urface
5	Copy Image
	Export Image
	Create Video
	Print Image
	Print Report

- Report (Command)
  - Starts the wizard to create an Excel report. Same function as **<Excel Report>**.
- Export Rich Text Report
   Starts the wizard to export a Rich Text report. Same function as <Rich Text Report>.
- Copy current view to clipboard The current presentation of the measurement will be copied to the clipboard. Same function as <Copy Image>.
- Render Video
   Starts the wizard to create a video of the measurements. Same function as



The output name and destination of the video must be entered. Clicking on leads to the following dialog

Export	
	indige
	Composer Settings
	Frames/Second:
	Video Codec: 4 Reload Codecs
	<back next=""> Cancel</back>

The frames per seconds for the video can be entered here. Depending on the amount of sequence images this will define the length of the video. A codec must be selected. When running this function the first time, all available video codecs must be identified by clkicking on **<Reload Codecs...>** The "Microsoft Video 1" codec is recommended since it is available on almost any PC.

#### • Print Report

Starts the wizard to print a report. Same function as **<Print Report >**.

In the area below the image is a separate slider, which can be used to define the transparency of the data images.



## 6.8 Status Bar

Ready

🥥 Scheduler Idle 🔿 LEDs or Lights Off 🥥 Temp. Stabilized 🌍 Door Closed 🕘 Monitoring Instrument 🔿 --.-°C | 11/20/2012 | 10:45:05 AM

In the status bar the current status of the hardware and software is shown.

Ready shows the current status of the system. When a measurement is running a progress bar is shown.

<u>scheduler Idle</u> displays the staus of the Scheduler. If the scheduler is busy (measurement / control jobs pending) the icon is yellow.

O LEDs or Lights Off indicates if the optional LED panels inside the instrument are turned on or off.

• Temp. Stabilized shows if the camera is initailised and the cooling status.

• Door Closed displays the stautus of the dark room door. When the door is open an additional warning in the Windows status bar pops up.

Monitoring Instrument indicates if the instrument is initialized and properly connected to the PC.

• shows the current temperature of the heated sample tray (if installed)

11/20/2012 10:45:05 AM displays the current date and time.

# 6.9 IndiGO Menu

#### 6.9.1 New

#### Project

To create a new project, start the wizard. For further details please refer to section 4.2.5.

#### **Controller Template**

To create a new controller template, start the wizard. For further details please refer to section 4.2.4.

#### **Measuring Template**

To create a new measuring template, start the wizard. For further details please refer to section 4.2.3.

#### 6.9.2 Start a new Acquisition

This command will start the acquisition wizard for on demand measurements.

Start an Aquisition		
		indige
	Select Project and Identity to Aqu	iire
	Demo	•
	Service Training-001-Test	Multi Sample
	Acquisition Settings	Tools
	Measurement Settings	Init Wheel
	Pre Controller Steps	Load Slider
	Post Controller Steps	Eject Slider
		Quick Check
	< Back	Start Cancel

Several quick check commands are available to verify the settings made to the application. For further details please refer to chapter 4.2.6.

#### 6.9.3 Create a new Schedule

The system will start the **<Create a new Schedule>** wizard, which allows you to select the contents you want to schedule first.

For further details please refer to chapter 4.2.7

#### 6.9.4 Report

indiGO offers the possibility to create and display measurement reports. The output can be adapted to the user's requirements. For further details please refer to chapter 4.4

#### 6.9.5 Print Image

indiGO offers the possibility to create and display measurement reports. The output can be adapted to the user's requirements. For further details please refer to chapter 4.4



#### 6.9.6 Print Report

indiGO offers the possibility to create and display measurement reports. The output can be adapted to the user's requirements. For further details please refer to chapter 4.4

#### 6.9.7 Logoff

This command will change the current user account.



When clicking on "Change password" a new password can be assigned The old and new password must be entered as well as the user name.

Change Password	<b>×</b>
Change Password Pi	operties
Username:	
Old Password:	
New Password:	
Confirm Password:	
	OK Cancel

#### 6.9.8 Exit

Quit indiGO to come back later.

# 6.10Tools Menu

#### 6.10.1 Backup Projects

IndiGO can create backups of the database or selected projects. Backups are created using the **backup smart wizard**.

Beckup Database	<b>×</b>	Backup Database
	indige	indice
Select Projects to Backup:		
Projects	*	
GPP Pflance HO dual GPP Pflancen 365 GPP Pflancen 165 GPP Pflancen HO 405 GPP Stancen HO 404 GPP Stancen HO 405 GPP Stancen HO		
V Imported Projects		Testadup
	-	Destination:
Tag Al Untag Al		C1/Bedup
< Ead.	iext > Cancel	< Bok Rext> Cancel

The wizard will guide you through the steps necessary to successfully create a backup. When selecteing the Backup first define which projects have to be backed up. Clicking on leads to the dialog box to select the backup target directory. This should be a remote network drive or removable storage to assure a proper backup An Backup name can be assigned. This is the name of the file created by the backup (extension zip.dba). Depending on the amount of data and the computer- / transfer performance the backup may take a while.

After a successful backup a summary page will pop up.

Backup Database	X
	indice
	Creating Backup 114 files in 1 projects Done
	< Back Finish Cancel

### 6.10.2 External Tools

indiGO supports some external tools which can be accessed through this menu.

#### 6.10.3 Import indiGO 1.x Project

Projects and measurement data created with indiGO 1.4 can be imported into indiGO for reanalysis via this command. A smart wizard will guide you through the steps necessary to import the data. Please follow the instructions given by the wizard.



Import IndiGo 14 Project	Import IndiGo 14 Project
Source Project (Requires IndiGo 1.4 Format): C: (Patabase 1.0 User (Admin (Projects) Jum 2m Destination Project: Jum 2m < Back Next > Cancel	Select Identities to Import: Identity V 15 V nouve 1 V nouve 3 V nouve 5 V nouve 5

After launching the wizard just select the project file (.proj) of the indiGO 1.x project to import.

The "Destination Project" will define the Project name where the imported files will be copied to. An import project name cannot be used for data acquisitions.

Clicking on *Next>* will lead to a selection dialog box. Here you can select the Sample IDs to import. Uncheck the boxes next to the SampleID which should not be inported. The data import will start when clicking on *Next>*. After the successful import a summary page will pop up.

#### 6.10.4 Import Winlight Data

Projects and measurement data created with WinLight can be imported into indiGO for reanalysis via this command. A smart wizard will guide you through the steps necessary to import the data. Please follow the instructions given by the wizard.

The file structure of the files to be imported must fit the file structure of images created by the "Reportergen" application in WinLight.

Photographic image xxx\_yyy-p.fts Signal Image xxx\_yyy-F.fts

Only image pairs (signal image plus photo image) can be imported.

Import WinLight Data	× .	Import WinLight Data			×
	indige			ind	l <mark>i</mark> C©
	Source Directory:		Select Items to Import:		
	C: Users kenoueg pocurients (vightuwic priberto Kom 10 3		Source	Identity	Idx
	File Filter & Suffix:		W mouse3	nou	0
	V Enable Filter		MOUSE3	MOU	0
	Thouse		📝 mouse3b	nous	0
	Deather the Dealerty		✓ mouse3b	nous	0
	Destination Project:		M mouse3b	nous	0
	Import Data		V mouse+	nou	0
	Identity:		W mouser	nou	0
	Manual Identity		W mouse6	nou	ő
			Tag Al Untag	Al	
	< Back Next > Cancel			Back Next >	Cancel

Using this structure xxx will be taken as SampleID (Identity). The 3 digit number yyy represents the sequence image number. The suffix defines the image type (photo –p; signal –f).

# 6.11 Service Menu

#### 6.11.1 Filter Manager

This command will start the indiGO filter manager.

Instrument Controller Setup				? ×
Filter Management Filter Management Excitation Inventory Kottation Inventory Advanced	Filter Management Emission Inventory		i	ndige
		4: Shutter		
		2: orange 1: Photo	590 [nm] 23	3 [nm] 30 [%]
	Name	Filter 1 Filter 2	Filter 3	Filter 4
	🚯 whl 001	Photo orange		Shutter
	🚯 whi 002	Photo blue	green	Shutter
	Filter Invento	ry Filte	er Manager	
Instrument is offline				Close

For further details please refer to section 4.1.14

## 6.11.2 User Manager

This command will start the indiGO user manager.

User Management User Manager Advanced	ser Management ser Manager New Edit	User Name	Created E		li <b>g</b> e
	New Edit	User Name	Created E	voire User Rol	
	Edit	<u>\$2.a</u>		spire ober itor	e
		20 g	11/3/2009 11/6/2009	Admin Guest	
		S S	11/6/2009	Service	
	Delete	<b>&amp;</b> υ	11/6/2009	User	
Instrument is offline					Close

For further details please refer to section 4.1.12.

#### 6.11.3 Controller Service

The **<Controller Service>** command will start the **Instrument Controller Setup**.



Instrument 	Calibration Focus Table				indi	
	Ltem 🔻	Z-Pos 🔺	F-Pos [mm]	Size [mm]	Image	-
Energy	Graph V	200	4.03626	279.759		
Lamp Energy		220	4.03626	270.038		
Video Codosa	🔅 Tools 🔻	240	4.03626	260.255		
Ouick Check		260	4.03626	250.408		E
State		280	4.03626	240.495		
Advanced		300	4.03626	230.516		
About		320	4.03626	220.47		
🚞 Filter Management		340	4.04387	210.354		
🚞 User Management		360	4.13608	200.167		
🣴 Flash Services		380	4.25108	189.909		
		400	4.38886	179.576		
		420	4.54943	169.168		
		440	4.73277	158.684		-
		Align Size		Adapt	Calibrat	te

The Instrument Controller Setup is used to setup your NightSHADE evo or NightOwl device and to perform all system calibration tasks.

#### 6.11.4 Device State

The **<Device State>** command can be used to display the current states of the instrument, the camera and the scheduler.

Instrument Controller Setup	State Instrument		indi	
Scheduler	Reference V	System		
		Busy	Offline (	
	Position	Referenced	Not Referenced	
		Error Info	Ū.	=
	Environment •	Axis State		-
<u> </u>	🔅 Ping 🔻	Emission	-1 回	
		Excitation	-1 💿	
		Focus	-1 💿	
		Z-Axis	-1 💿	
		Sample Changer	-1 💿	
		lris	-1 💿	
		Position		
		Focus Position	-1 [steps]	
		Z-Axis Position	-1 [steps]	
		Heater		
	Stop Aviel	Available	Offline	-
	A Stop Axis:	Decet and the	v 🙈	
A Tratura at is afflice				

Raw device commands and test operations can be executed using the context command buttons.

#### 6.11.5 Transport

This menu item has no functionality using the NightSHADE evo.

#### 6.11.6 Quick Check

The **<Quick Check>** command can be used to get access to highlevel quickcheck commands for photo, luminescence and fluorescence.

Instrument Controller Setup			5 X
Instrument Quick Check G-G State	Instrument Quick Check		indice
		Photo Quick Check	
			Photo
		Measurement Quick	Check
			Luminescence
			Fluorescence
Instrument is offline			Close

Each highlevel quickcheck command will start a smart wizard which will guide you through the neccessary steps to perform the quickcheck.

#### 6.11.7 Controller Version

The command will display the current controller and instrument version information.

Instrument Controller Setup			<u> २</u>
Instrument	Instrument About		indice
		Camera	
		Version	
		F losta ment	
		Version	
		Model	
		Controller	
		Product Name	IndiGo
		Company Name	Berthold Technologies
		File Description	Instrument Controller
		Hie Version	2.0.0.23
		Legal Copyright	© 2010. All rights reserved.
		Virtual Memory	
		- maa romoy	
Instrument is offline			Close

#### Please note:

Watch the owl while moving the mouse.

#### 6.11.8 Messages

The **<Messages>** command will start the systems **Message Monitor** which can be used to review internal indiGO trace and error messages.


<u> </u>	lessage Monitor		<b>1</b>		X
lerr	ninal <u>V</u> iew <u>?</u>				
\$\$	Connect 🔻 😆 📋 Cle	ar Window 🛛 🔁 🔚	New Terminal	∃ <b>\$ \$ \$</b>	
	Time	ID	Server	Message	^
0	10/7/2010 7:03:30 PM	00000ed8	Instrument	flatfield correction is not calibrated	
0	10/7/2010 7:03:36 PM	00000ed8	Instrument	flatfield correction is not calibrated	
	10/7/2010 7:03:41 PM	00000ed8	Instrument	flatfield correction is not calibrated	
	10/7/2010 7:09:35 PM	00000860	Instrument	shutdown	
	10/8/2010 9:21:42 AM	00000e50	Instrument	controller startup	
	10/8/2010 9:22:20 AM	0000072c	owl	device is referenced now	
	10/8/2010 10:13:08 AM	00000e50	Instrument	shutdown	
	10/8/2010 10:14:34 AM	00000e38	Instrument	controller startup	
	10/8/2010 10:14:56 AM	00000Ь20	owl	device is referenced now	
0	10/8/2010 10:14:59 AM	00000e38	Instrument	user onki logged on	
	10/8/2010 10:21:38 AM	00000e38	Instrument	shutdown	
	10/8/2010 10:21:40 AM	00000f54	Instrument	controller startup	
	10/8/2010 10:21:52 AM	00000b64	owl	device is referenced now	
0	10/8/2010 10:21:53 AM	00000f54	Instrument	user onki logged on	
0	10/8/2010 10:30:27 AM	00000590	Instrument	flatfield correction is not calibrated	
	10/8/2010 10:31:00 AM	00000590	Instrument	flatfield correction is not calibrated	-
Conr	nected to 'Trace' at 'LOCALI	HOST:1025'		10/10/2010 12:32:0	01 PM

# 6.11.9 Options

The Options command allows you to setup further indiGO options.

IndiGo Options			×
Overlay  Coverlay  Coverlay  Manually Layer  Peak Search  Coverlage  Coverlage  Peak Search  Coverlage  Coverlage  Peak Search  Coverlage  Coverlage  Peak Search  Coverlage  Peak Search  Peak Search	Annotation Overlay		indige
Setup		Annotation	
Export About		Item Color:	Light Blue
		Item Selected:	Yellow 🔻
		Grid and Ruler Color:	Brown 🔻
		Grid and Ruler Selected:	Brown 🗸
		Default Font:	Arial 👻
		Default Font Size:	12 🔹
			Close

#### Overlay

Access to overlay colour options. Here user specific colors for the different data types (peak search areas, manual areas etc.) can be defined.

#### IndiGO

Access to setup configuration wizards and connectivity settings necessary for update and support mail access.





#### **Setup Database Connection**

Using this wizard the connection type can be changed. This might be required when the computer was moved away from the system or in case the database was changed from the local disk to a network drive.

Enter the type of connection first (usually direct connection to the instrument) and select the location of the database. If the database is not present at the selected location, a new, empty one will be created.



Finally the connection must be entered. In case of a local connection, the default "LOCALHOST-1024" is OK.



#### Setup Default User

With this wizard a default user can be assigned who logs in automatically after launching indiGO.



The user management can be disabled completely or a default user can be assigned.

#### First Time Initialisation Wizard

This button will start the initial initialization. See chapter 0 for details





### Export

Export report settings like address, company logo and preferred format.



# 6.12View Menu

# 6.12.1 Toolbar

This command shows or hides the toolbar. Starten Acquisition ... Command shows or hides the toolbar. This option is not persistent and the toolbar will be shown on the next start of indiGO.

### 6.12.2 Status Bar

This command shows or hides the status bar.

 Ready
 Scheduler Idle
 LEDs or Lights Off
 Temp. Stabilized
 Door Closed
 Monitoring Instrument
 ---\*C
 11/20/2012
 10:45:05 AM

 This option is not persistent and the statusbar will be shown on the next start of indiGO.

# 6.12.3 Scheduler

Open the editor for the scheduler. For further details please refer to section 0.

# 6.13Batch Image Export

IndiGo allows the batch export of images within a sample identity. The images are exported as 16 bit gray scale TIFF files with meta data attached as XML file. To start the export, select the required sample ID from the tree and right click on it.



Select "Export Identity" and a dialog box to select the destination folder will pop up.



Click on the arrow to select the desired folder and on  $\square$  to start the image export. A subfolder with the sample ID name will be generated. Depending on the amount of images, the export could take a while and requires a large amount of disk space. The file name structure is as follows:

Sequence number (3 digits)\_original filename (date coded 20120523123456).image type (photo; lumin;fluor).imagenumber (000-009).tif



# 7 Trouble Shooting

# 7.1 Hardware

### 7.1.1 Door status always open

If the door status is always open, the reason could be a disaligned door sensor. The door sensor is located at the upper right front side of the dark box. To adjust the door sensor refer to 8.2.3

## 7.1.2 Fluorescence lamp not working

The built in halogen fluorescence excitation light source has a lifetime of approx. 30h. When the lamp is no longer working replace the lamp. Refer to 8.2.4

## 7.1.3 No Fluorescence light output

When using the 4 light guides for fluorescence illumination it could happen, that the fluorescence light is not present at the light guides. In this case check the position of the optical switch (see chapter 5.3.7) the optical switch must be in the lower position to get light output at the 4 fibre optic light guides.

# 7.2 Software

### 7.2.1 Surface plot not visible

When running the surface plot from the Report menu a box with an error message (D3D device error) pops up instead of the surface plot. This indicates that the installed graphics card or the corresponding driver does not support Microsoft Windows DirectX 9 features.

In this case a driver update will solve the problem since many older PC do not have the lates drivers installed. In case the graphics card does not support DirectX 9 a new graphics card is required.

# 8 Cleaning and Maintenance

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# 8.1 Camera and Lens

### 8.1.1 Removing the bottom plate

The retractable bottom plate is removable. The bottom plate is locked by a latch underneath the telescopic rail. To remove the plate open the latch on both rails. The black plastic latch is located inside the rail. After unlocking the plate it can be carefully slided out to the front.

To reinstall it carefully slide in both rails at plate to the counterpart located at the bottom of the instrument until both latches lock with an audible click. Make sure the plate is inserted properly with the knobs facing the front.

## 8.1.2 Cleaning the Dark Box

The dark box should only be cleaned using a mild detergent and a lint-free tissue or a vacuum cleaner. To this end, the bottom plate which is locked by a latch underneth the telescopic rail should be removed (see above).

## 8.1.3 Cleaning the Camera

Clean the camera only with pressurized air spray or optical paper!

Remove the camera for cleaning.

### Dismantling and cleaning the camera

- Turn the camera to unlock the bayonet lock.
- Carefully lift the camera off from above.
- Do *not* detach the cable from the camera.
- If the camera has a shutter, start an acquisition with long exposure time via the software. This opens the camera shutter at the bottom and allows you to remove dust particles.

Caution: Do not damage the shutter by touching it when it's closed!

• Clean the camera using pressurized air spray.

#### Installing the camera

 $\circ$   $\;$  Install the camera with both screws to the transport unit. The port must face the camera lift.

## 8.1.4 Cleaning the Lens

The lens can be cleaned when it is removed from the instrument. To do so, carefully unscrew the lens from the C-Mount thread. Hold the lens with both hands since it could be damaged when falling down on the bottom plate when the thread is completely unscrewed. It should only be cleaned using pressurized air spray or cleaning agents available in photo shops!

To reinstall the lens hold it with both hands and carefully screw it to the C-Mount. Make sure the F-stop is set to 0,95 and locked to avoid loss of light.

# 8.2 Darkbox

# 8.2.1 Replacing the Air Filter

The air filter should either be cleaned or replaced, depending on the level of contamination. Remove the black ventilator cover on the rear of the instrument and clean the filter (blow it out or wash it) or replace it.

# 8.2.2 Replacing the Fuse

The mains fuses are located on the rear panel of the *NightSHADE evo*. The fuse holder and the mains connection socket are one unit.

### Proceed as follows:

- Pull the power cord.
- Insert a screwdriver from below into the opening of the fuse inset.
- Pull out the fuse inset with the screwdriver like a drawer.
- Replace the fuse.
- Insert the fuse holder again until it clicks into place.

Use only certified fuses (original spare part) that match the value and the line voltage indicated on the device. (250V T 6,3AL)

# 8.2.3 Setting the Door Sensor

In case the sensor on the door frame should be displaced so that there is no contact although the door is closed, you can easily re-adjust the sensor as follows:

- Open the locking screw at the bottom of the sensor using a 1.5mm Allen key.
- Push the sensor out inserting the Allen key at the rear side of the sensor housing, so much that it protrudes by about 1 cm. Do *not* tighten the locking screw.
- Carefully close the door and lock it so that the sensor is pushed through the door into its correct final position.
- Open the door carefully and tighten the locking screw.



# 8.2.4 Replacing the halogen lamp

The halogen lamp is located close to the excitation filters. It is protected by a black cover plate.



To replace the lamp proceed as follows. Make sure that the same type of lamp is used to guarantee proper light output.



Be careful when touching the lamp since it could be hot when the lamp was in use before.

- Switch off the instrument and unplug the mains connector. If the lamp was in use before allow some time to cool down the lamp.
- Remove the screw that closes the flap to open the top flap to get access to the lamp module.
- Pull out the black protection cover to get access to the lamp.
- Remove the lamp from the snap in holder.
- Unplug the lamp socket and plug in the replacement lamp. Do not touch the reflector of the lamp to avoid damage of the lamp.
- Place the lamp back into the snap in holder. Make sure the lamp is fitted properly in the holder (round edge fit with lamp reflector).
- Insert the protection cover. Make sure the cover is properly fixed and both pins are securely fastened in the mounting holes.
- Close the top flap and fix the screw.

# 9 Technical Data

Camera	Peltier/air-cooled CCD camera C-Mount adapter
Resolution	1024 x 1024 Pixel x 13µ
Quantum efficiency (max.)	>90%.
Noise	Readout: $\Sigma$ 7 e <sup>-</sup> RMS
Exposure time	from milliseconds to hours
Binning of pixels	Variable in X- and Y-direction to increase the signal/noise ratio
Temperature	Ambient temperature during operation: 10° to 28° C Cooling temperature -20200°C Ambient temperature during storage: -20° to 70° C No condensation
Lens	standard: 25mm f = 0,95 macro: 50mm f = 0,95
Interface	USB 3 (B-type connector)
Illumination	internal illumination can be adjusted via the software optional white light fluorescence illumination (400-800 nm)
Max. sample size	266 mm x 266 mm
Min. sample size	130 x 130 mm with standard lens (65 x 65mm with macro lens)
Dimensions	Acquisition device:600 mm x 900 mm x 400 mm (WxHxD) Camera head: 110 mm x 175 mm x 110 mm Sample chamber (inside): 400 mm x 380 mm x 450 mm
Weight	approx. 40 kg (depending on version)
Operating voltage	AC 100-240 V (+/-10%); 50-60Hz
Power rating	600VA, (max. 200VA through internal power socket)
Fuse	250V T 6,3AL
Computer	Minimum requirements: Standard PC with 3 GHz Pentium processor (MultiCore recommended) 4 GB RAM. Free USB2 port Graphics card with 1280x1024 pixels resolution 32 bit colors, DirectX 9c Sup port 17" LCD monitor Windows 10 (32 or 64 bit)

# 10 Appendix

- **10.1** Accessories
- 10.1.1 LED Panels



The LED panels are used to simulate environmental conditions like day / night changes while the sample is placed inside the dark box and no measurement is running. The LED panels are available in 2 different versions with the following wvelenght configuration:

- Green panel (ID 56589-01)
  - Blue (470nm)
  - o Green (520nm)
  - Red (660nm)
  - Far red (730nm)
- White panel (ID 56589-10)
  - Blue (470nm)
  - White (cold white)
  - o Red (660nm)
  - Far red (730nm)



Make sure to wear standard safety glasses when using the LED panels due to the possible high intensity of the light source (CE EN 166, EN 172). These safety glasses ensure that bright, intense light intensity will not harm the beholders eyes.

The panels are coded for the left and the right hand side. They are fixed on the upper left and right side above the sample using the hooked fixing.

The spectra of the single LED channels are shown in the diagram below





The electrical and cooling liquid tubing connections inside of the dark box are shown in the image below:



Connect the LED panels only while the instrument is powered off to prevent electrical damage of the LED panel.



The control cable of each panel is plugged into the socket at the corresponding side of the connection box (see 5.3.3).

Water cooling connects to the water supply bar located at the rear side of the dark box. The plug at the lower side of the supply bar is a bypass plug (instead of the temperature controlled sample table) and must be present to ensure a flow of cooling water.

External connectors for the cooling liquid are located at the rear side of the miniChiller and the rear panel of the NightSHADE evo:



Tubing connector at the miniChiller and at the rear panel of the NightSHADE evo

When the miniChiller or any other device using liquids to cool the LED panel is connected to the NightSHADE evo it is not allowed to operate 100/230V devices such as transilluminators etc. inside the dark room due to electrical safety regulations.

The preset temperature of the cooling water in the miniChiller must not be set below ambient temperature since this could lead to condensation on the LED panel.

The "Artificial daylight" control must be enabled in the indiGO service backend in order to control the intensity of the LED panels.

Sample Changer

To operate the cooling device please refer to the dedicated manual of the minChiller.



## 10.1.2 Temperature Controlled Measurement Table

The temperature controlled measurement table is used for measurements which require a temperature controlled environment.

If you are working with the heated macro table, plug the connector into the 9-pole Sub-Dconnector on the power box at the center top inside the darkbox (see 5.3.3.) The device must be switched off, since after the NightSHADE evo is switched on the heating system is detected and may get initialized.

If the heating system is plugged in during ongoing operation, it will work only after the device has booted up again. This is done by clicking on . The state and selecting **Boot Device** from the **Reference** dropdown list.

Reference 🔻		System
۲	Boot Device	

After approx. 5 to 10 minutes the heated measurement table has reached its working temperature (preset 37°C). The current temperature is displayed in the bottom status bar of the indiGO user interface **1** • 228°C 9.



The default temperature can be defined in the configuration backend of the software accessible through the instrument controller. Right click on the controller icon and log-in with a valid user account information selecting Setup from the context menu.

In the **<Presets> <Heater>** section the default temperature can be set. When the checkbox at the bottom is checked indiGO will set the default temperature at system initialization.

Instrument Controller Setup		? <mark>×</mark>
Instrument     Presets     Bresets     Calbration     Video Codecs     Quick Check     State     Advanced     About     Filter Management     User Management	Presets Heater	Heater Default Temperature         Temperature [?]: 37         Hote         The Heater is autodetected by the system.         A default temperature could be set for the heater. The temperature will be set if a heater is connected.
		☑ Set default heater temperature on init.
Instrument is ready		Close

# 10.1.3 Temperature controlled tray with anti condensation



The temperature controlled tray can be placed inside the NightSHADE evo to allow a temperature controlled base plate when using petri dishes. The temperature is regulated by the water cycle used by the LED panels as well. Since the miniChiller can only cool down liquids it might be necessary to use the miniStat 125 or the miniChiller Plus when temperatures above or around ambient are required. The tube connections from chiller to NightSHADE evo are described in chapter 10.1.1.

A water cycle flows through the base plate of the tray which stabilizes the temperature. To avoid condensation of the petri dish lids an airflow is established over the dishes in a circular flow direction. This temperature should be about 3-5 degrees above the table temperature and can be set using indiGO (see 10.1.2).



The temperature controlled tray must be connected to the water cycle using the lower two connectors at the water supply bar. When the tray is not used, both outlets must be shorted (bypass) as shown below. If this is not done, the LED panels are not cooled properly due to an interruption in the water cycle. This could lead to an error state of the LED panels due to overtemperature.



The temperature table itself is positioned inside the darkbox by the 4 feet. Make sure the 4 feet fit inside the corresponding wells located at the base plate.



Avoid temperatures who are significantly below ambient as this could lead to condensation.

10.1.4 Turntable





The turntable is used together with the sideview option. Using the turntable up to 6 samples can be measured in a automatic sequence. 3 different sample holders can be selected:

- 100mm square petri dishes
- 130mm square petri dishes
- DeWit culture tubes

Installation:

- Turn off the NightSHADE evo
- Place the turn table on the base plate that the rubber feet of the table will fit the positioning holes and markers on the positioning plate. The orientation of the turn table can be seen on the image below. Depending on the sample size the turn table can be lifted up to change the image center. In addition the positioning plate offers 2 different positions to adjust the sample size.

When using the 130mm petri dishes the turn table must be placed at the outer right position to ensure the whole plate can be imaged using the standard lens. The 100mm petri dishes can be completely measured at the left position of the position

plate. In this case the adjustable feet of the turn table must be pulled out to elevate the turn table (bajonet lock) to place the sample center in the center of the optical axis.



- Connect the 9 pin cable supplied with the turn table to the connector at the connecting panel at the upper position inside the dark box. Make sure the cable is not blocking the table or gets sqeezed by the retractable base plate. Remove the dummy plug and keep it in a save place. When not using the 9 pin connector, the dummy plug must be present
- Switch on the NightSHADE evo to initialize the turn table

Before removing the turntable make sure, the instrument is switched off. The sample changer feature must be activated in the indiGO service backend in order to control the turn table

Sample Changer

The position of the turn table is defined by the number of the sampleID. 001 represents the sample position 1, 006 moves the turntable to position 6. 007 will use position 1 again.

If further movement of the table is required a different position can be selected using a control sequence (see. Chapter 4.2.4)



# 10.1.5 Test Light Source

The Test Light Source is used to calibrate and test the System.



It contains a low light source which will glow for about 15 min. without external supply. To charge the unit, use the included mains adapter.

Charging is indicated by a red LED on the test plate. As soon as the plate is fully charged, the green LED will light. It takes about 1-2 minutes to charge the plate.

Now the plate is ready to be used in the NightSHADE evo. The spot with the white reflector (microplate position E9) emits light with an energy of approx 20nW (see individual certificate). This spot can be used to check the NightSHADE evo calibration. The measuring time should not exceed 1 second since the camera could be saturated otherwise.

Position E6 gives a lower signal intensity. This position can be used to check the detection limit with additional neutral density filters on top of the light source.

A special center plate is optionally available to place the E9 light spot at the center of the image.

The required light position is controlled though the sliding switch at the side of the plate (position E9 -> overl.; E6 -> 96). When the plate is not in use put the slider switch to position "OFF".

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