SkyScan 1076

In vivo X-ray Microtomograph

Instruction Manual

© SkyScan 2002

Manufactured by
SkyScan n.v.
Vluchtenburgstraat 3,
2630 Aartselaar
Belgium

TABLE OF CONTENTS

1 INTRODUCTION IN X-RAY MICROSCOPY AND MICROTOMOGRAPHY

1.1 Introduction

Any conventional optical or electron microscopes allow visualising only two-dimensional images of a specimen surface or thin slices. However, in most cases a conclusion about original three-dimensional object structures cannot be made on the base of two-dimensional information.

One can obtain the three-dimensional information of object structures by cutting them into very thin slices, which can then be visualised in the light microscope and interpolate the two-dimensional information into a three-dimensional structure model. This method however is not only very cumbersome but also not very reliable since the object structure itself can be altered by the preparation technique and the distance between the slices is usually too coarse to avoid loss of 3-D information.

An x-ray (radiography) system produces two-dimensional shadow images of complete internal three-dimensional structures, but in a single two-dimensional shadow projection the depth information is completely mixed. Only an x-ray tomography system allows us to visualise and measure complete three-dimensional object structures without sample preparation or chemical fixation. Typically, the spatial resolution of conventional medical CT-scanners is in the range of 1-2.5 mm, which corresponds to 1-10 cubic mm voxel (volume element) size. Computerised x-ray microscopy and microtomography now gives possibilities to improve the spatial resolution by seven to eight orders in the volume terms. The system "SkyScan 1076" allows to reach a spatial resolution of 15 µm corresponding to near $3x10^{-6}$ cubic mm voxel size. As in the "macro" CT-scanners, the internal structure can be reconstructed and analysed fully non-destructively.

1.2 Basis principles of microtomogaphy

Any x-ray shadow image is corresponding to a two-dimensional projection from the three-dimensional object. In the simplest case, we can describe it as a parallel x-ray illumination. In this approximation, each point on the shadow image contains the integration of absorption information inside the three-dimensional object in the corresponding partial x-ray beam.
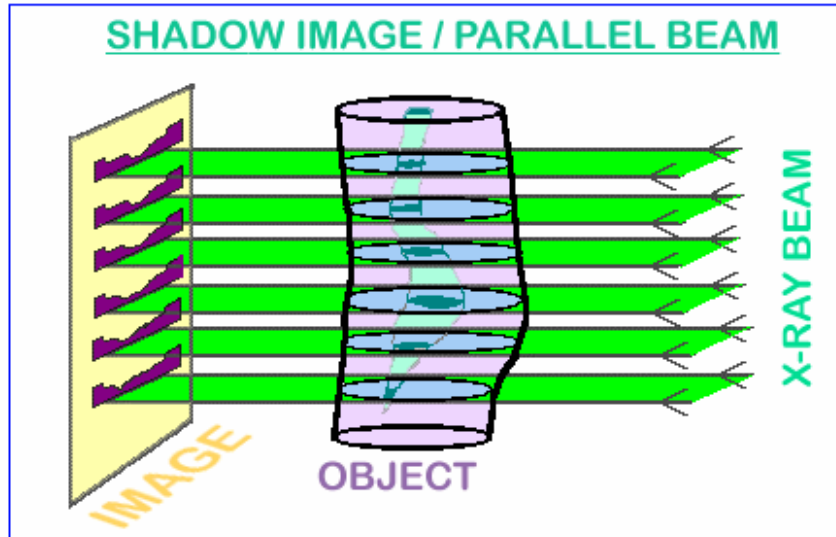
For parallel geometry one can divide the problem of a three-dimensional reconstruction from two-dimensional projections into the serial reconstruction of two-dimensional object slices from one-dimensional shadow lines. Let's show a possibility of this reconstruction on a simple example, an object with only one point with significant absorption in an unknown place. In the one-dimensional shadow line we will have a decreasing of intensity of the shadow of absorption in the object area, see figure on the next page. Now we can initialise in the computer memory an empty array of pixels (picture elements) corresponding to possible object displacement. Of course, one must be sure that all parts of the reconstructed object will be inside the field of view. Because we have the position of the shadow from the absorption points of the object, we can mark on the area of reconstruction in the computer memory all possible positions of absorption points inside the object as lines.

OBJECT   AREA OF RECONSTRUCTION
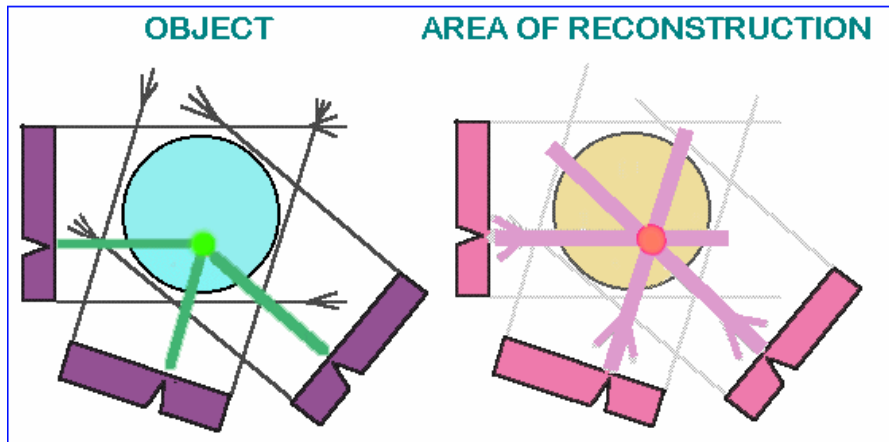
Now let's rotate our object and repeat this operation. In each new rotation position of the object, we will add to the area of reconstruction the lines of possible object positions corresponding to position of shadow. This operation is named "back-projection". After several rotations we can localise the position of the absorption point inside the area of reconstruction. In increasing the number of shadow projections from different views this localisation become more and more defined.

BACK PROJECTION: single point object

3 views   5 views   9 views   18 views
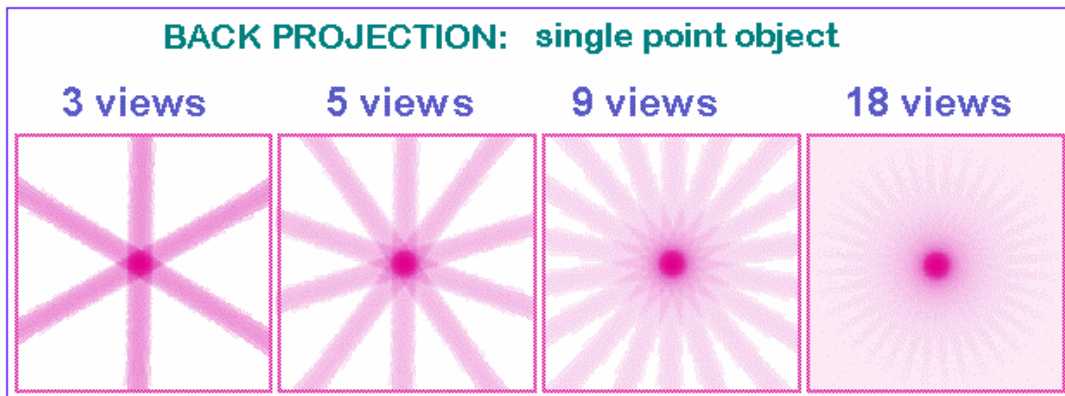
In the case of reconstruction from an infinite number of projections one can get an image with a good definition of the absorption area position inside the initial object. At the same time a blur area will accompany the pointer image because it is produced as a superposition of lines with all inclinations. Now we know what image will be produced from the pointer object and we can "pre-correct" the initial information in absorption lines to make the resulting image more corresponding to the real object. This correction ad some "negative absorption" outside the point of the object shadows to eliminate the positive blur in the back projection process (convolution).
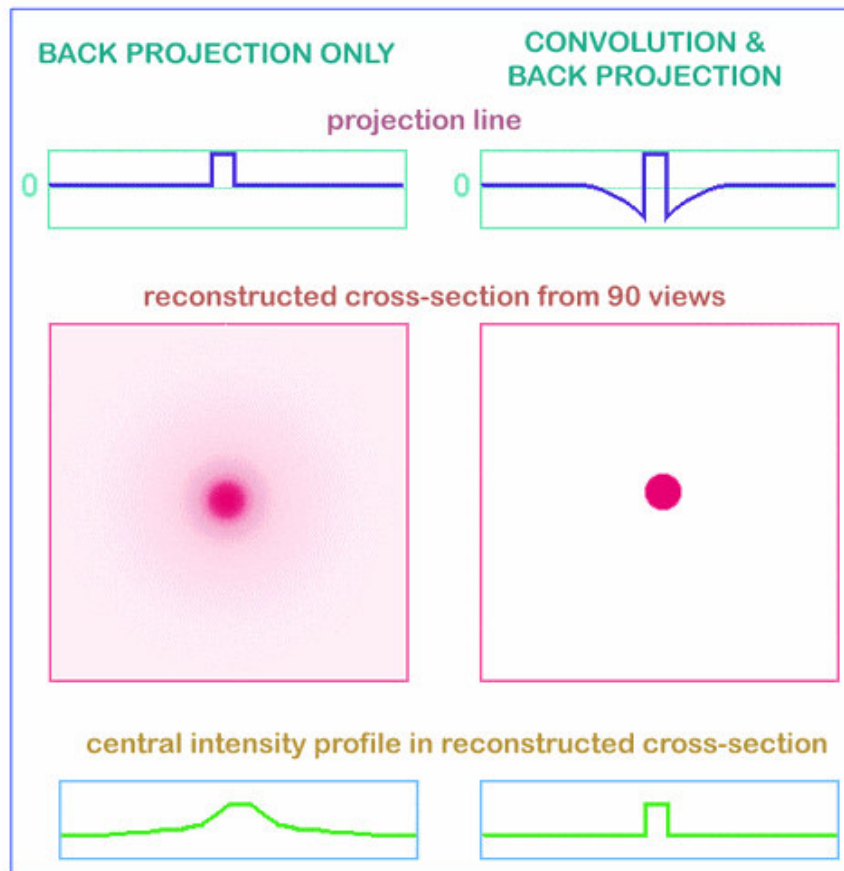
The same algorithm can produce the cross-section image not only from the single point object. Any real object can be represented as a big number of separate absorption voxels and linear absorption in any x-ray beam is corresponding to the sum of all absorption from all voxels inside this beam. By this way the two-dimensional cross-sections of the object can be reconstructed from the one-dimensional shadow lines in different views.

Unfortunately practically all x-ray sources cannot generate parallel beams. In a real case, one will use a pointer source and fan x-ray beam in the object area. For tomography reconstruction we can find the solution of this problem by the reordering of the shadow information. New pseudo-parallel beams can be constructed from the parts of several fan beams with different views and the same reconstruction method for fan-beam x-ray sources can be used.

FAN BEAM TO PARALLEL BEAM

PARALLEL BEAM APPROX.

ORIGINAL FAN BEAMS
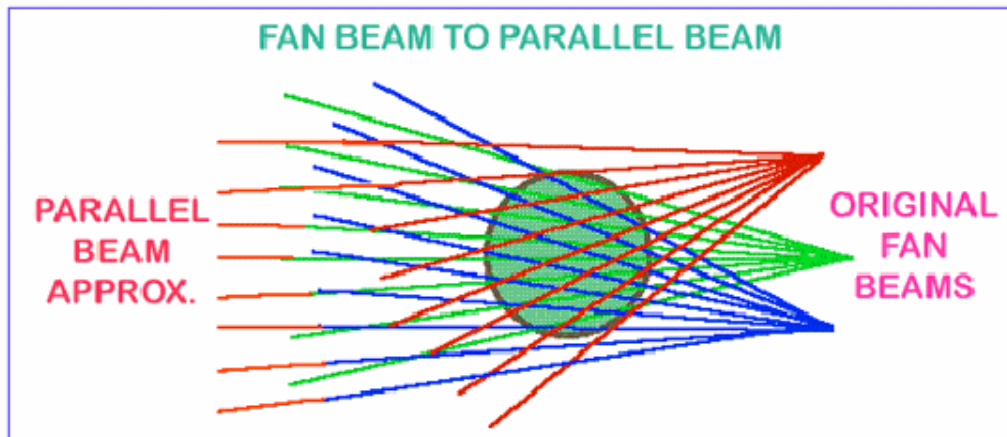
In the case of x-ray acquisition, the image contains information about the intensity reduction inside the three-dimensional object. Because the x-ray absorption is corresponding to exponential law, we can restore the linear absorption information from the shadow image by logarithmisation.

This operation is very non-linear and any noise in the small signal areas can produce significant errors in reconstruction. To eliminate these errors an averaging of initial data and results of logarithmisation can be used. On the another hand we can try to improve the signal to noise ratio in the shadow image to reach the most representative information.

One more effective way of noise reduction in the reconstruction process is a special selection of correction function for convolution before back projection. In the simplest case (described above) the correction function produces two "negative absorption" reactions around any signal or noise peak in the shadow line and this behaviour becomes very dangerous for noisy initial information.

Special selection of convolution function for correction with spectral limitation by "Hamming window" allows solving this problem. In x-ray microtomography an information from voxel with very small physical size should be detected and right choice of parameters for noise reduction becomes very important.

1.3 Reconstruction to image

1.3.1 Acquisition, creation of acquisition data

During the acquisition the source-detector pair will rotate over 180 degrees. At each position the shadow image or transmission image will be acquired.

Cone beam acquisitions saves all of these projection images as 16 bit TIF files. The data set after scanning consists of a set of images, all of them are normal transmission X-ray images. For each position over the 180 degrees rotation a full 16 bit shadow image will be stored on disk. The number of files after this acquisition is thus depending upon the rotation step selected. For a typical step of 0.7 degree, there will be 257 images plus a small number to start the re-sampling of the images for horizontal or fan compensation of the x-ray beam.

1.3.2 Start of the reconstruction

After the acquisition is finished we have to start the reconstruction. We will use the 16-bit TIF shadow images for the reconstruction. We will now generate from this, by using the reconstruction algorithm, a raw data reconstructed cross-section. This is not yet an image, it is a float point matrix holding absorption values in the reconstructed cross-section.

	1	2	3		n-2	n-1	n
1	0.022	0.024	0.013	… … … ….	0.910	0.990	0.950
2	0.023	0.026	0.012	… … … ….	0.870	0.900	0.890
3	0.028	0.027	0.019	… … … ….	0.800	0.810	0.780
4	0.026	0.210	0.020	… … … ….	0.820	0.830	0.840
⋮	⋮	⋮	⋮		⋮	⋮	⋮
n-5	0.030	0.031	0.034	… … … ….	0.710	0.720	0.740
n-4	0.031	0.034	0.390	… … … ….	0.700	0.730	0.760
n-3	0.034	0.035	0.042	… … … ….	0.730	0.790	0.780
n-2	0.036	0.036	0.041	… … … ….	0.740	0.780	0.770
n-1	0.040	0.037	0.040	… … … ….	0.750	0.770	0.720

The size of the matrix is like the number of pixels inside a cross-section or in a line on the CCD array (n is the number of pixels in a line of the shadow image or the CCD array).
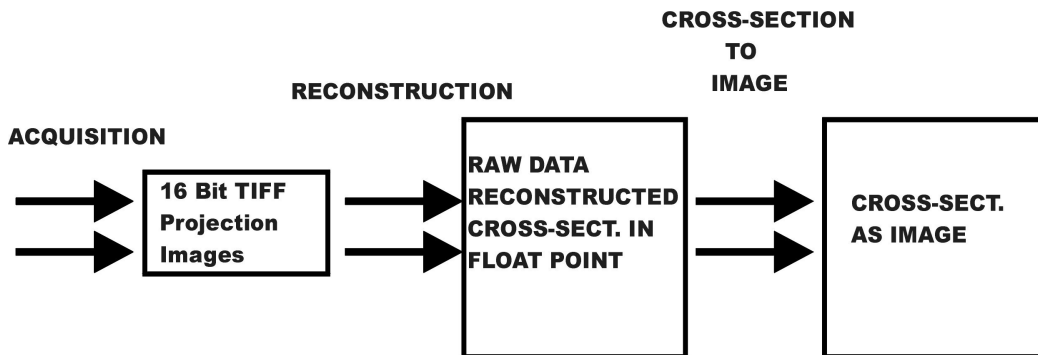
We can save the reconstructed cross-section as a float point matrix holding the attenuation values after the reconstruction or as explained in the next section transform it to an image with the 256 grey values (8bit).

1.3.3 Cross-section to image

After creating the raw data reconstructed cross-section, we have to generate an image. Any images uses 256 gray scales. Therefore, we have to find away to convert the 12 bits or more, depending upon the camera, information into a gray scale image.
Minimum and maximum values are selected. All values between these will be displayed as half tone. In a normal image, all attenuation values below the minimum will be white everything above the maximum will be displayed as black. Reconstructed array will be shown as a half-tone image of cross- section with linear conversion to 256-grades of gray inside selected density interval.
In the Skyscan systems, using a Windows environment, the final image generated can be exported to BMP, RAW 16Bit or TXT -files.

Following image summarizes all action and steps to generate the cross sectional data.

```
                                            CROSS-SECTION
                                                  TO
                        RECONSTRUCTION          IMAGE

ACQUISITION

            ┌──────────────┐    ┌──────────────┐    ┌──────────────┐
            │ 16 Bit TIFF  │    │ RAW DATA     │    │ CROSS-SECT.  │
  ──►       │ Projection   │ ──►│ RECONSTRUCTED│ ──►│ AS IMAGE     │
  ──►       │ Images       │ ──►│ CROSS-SECT. IN│ ──►│              │
            └──────────────┘    │ FLOAT POINT  │    │              │
                                └──────────────┘    └──────────────┘
```

2 SKYSCAN 1076 SYSTEM OVERVIEW

SPECIFICATIONS:

Maximum object size	68mm(D) x 200mm(L) for rats or 35mm(D) x 200mm(L) for mice, 17mm(L) per single scan
X-ray source	20-100kV, 10W, <5um spot size (@4W), >10000h estimated lifetime, air cooled sealed type, 4-positions automatic filter changer for energy selection
X-ray detector	10 Megapixel (4000x2300x12bit) cooled digital X-Ray camera with fibre-optic coupling to scintillator
Spatial resolution	User selectable pixel size 9µm / 18µm / 35µm (isotropic), 15µm low-contrast resolution (10% MTF)
Projection / cross-section Image size and formats	1000x520…8000x2000 pixels projection images (16-bit TIFF format) 1000x1000…8000x8000 pixels cross-section (BMP, RAW 16-bit, TXT formats, converter to JPEG)
X-ray loading to the animal	0.1-0.5 Gy per scan typical
Scanning system	source-detector pair rotation with 0.02 deg. min. step size, 50um object positioning accuracy with 400mm travel, 50mm camera positioning/alignment with 1um accuracy, <10 microns overall stability during scanning
Software package	scanner control, preview (35x200mm scan), acquisition for reconstruction, volumetric (cone-beam) reconstruction of one / several / all cross sections, ROI-reconstruction, local density measurements in HU, 3D-rendering, virtual manipulation with reconstructed object, morphological analysis in 2D and 3D
Reconstruction algorithm	Modified Feldkamp: multislice volumetric (cone-beam) reconstruction. Up to 2000 slices can be reconstructed after one scan. Full image mode, partial reconstruction mode, possibility for detail local reconstruction with object bigger than field of view.
Radiation safety	<1uSv/h average during full scan at 10cm from the instrument surface
Installation requirements	Power 100-130V/5A/50-60Hz or 200-240V/3A/50-60Hz, 18-28C temperature, <85% humidity, no condensation, vibrations 0.1…100Hz <40 microns
Size/Weight	Desk top instrument 750mm(H) x 650mm(D) x 2200mm(W), 150Kg + computer, monitor, keyboard, mouse

The "SkyScan-1076" is a high- resolution low-dose X-ray scanner for in-vivo 3D-reconstruction with spatial resolution of up to 15 microns inside the small laboratory animals (rats, mice, etc.). It consists of the combination of Micro-CT system and a computer with system control software and reconstruction software. This system allows reconstructing non-invasively any cross-section through the animal body with possibilities to convert the reconstructed dataset into a realistic 3D-image and calculate internal morphological parameters.

The equipment contains an X-ray microfocus tube with high-voltage power supply, a rotation stage with overall accuracy of <10µm, a translation stage, a two-dimensional X-ray CCD-camera connected to the frame-grabber and a Dual Intel Xeon computer with LCD monitor.
All subsystems of the X-Ray Microtomograph are inside a steel desktop case.
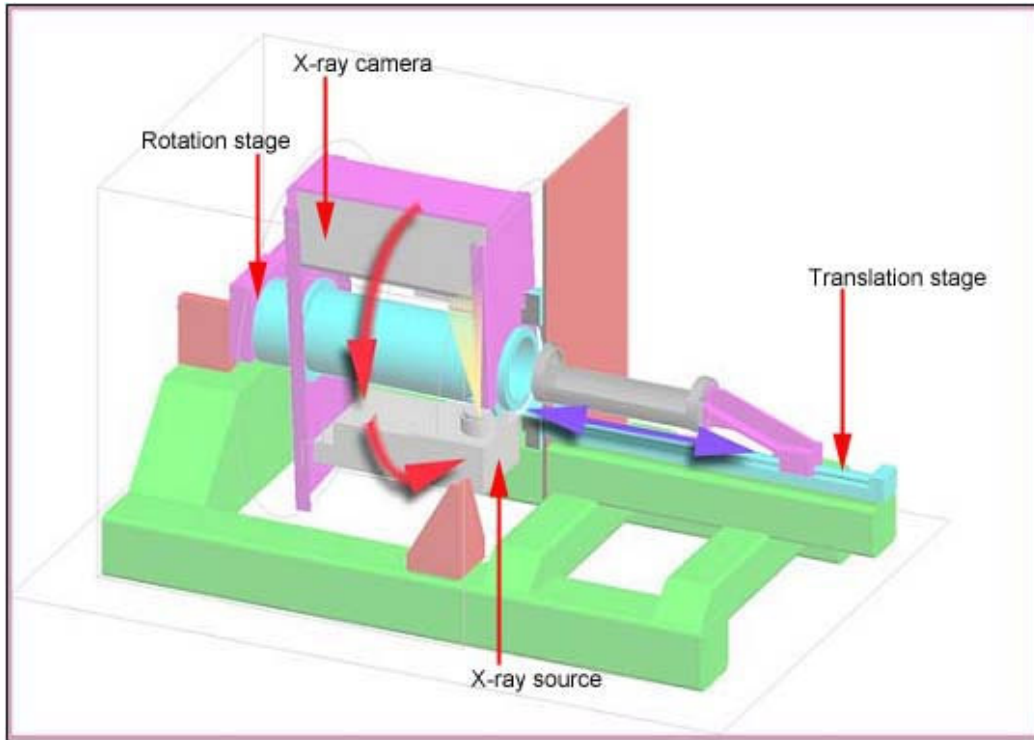
For "SkyScan-1076" the X-ray microfocus tube with 5 micron focal spot size operates at 20-100kV / 0-250µA. The special X-ray CCD-camera is based on 10 Megapixel (4000x2300 pixels) cooled CCD-sensor with fibre optic coupling to x-ray scintillator

The X-ray shadow projections are digitised as 1000x520 to 8000x2000 pixels with 4096 brightness gradations (12 bit). The reconstructed cross-sections have a 1000x1000 to 8000x8000 pixels (float point) format and 9 /18 /35µm pixel size in any place of the scanning area. The scanning area is 68mm x200mm or 35mm x 200mm (two carbon-composite beds supplied)

For the reconstruction one can use a volumetric (cone-beam) reconstruction of one / several / all cross-sections or a ROI-reconstruction. After the serial reconstruction, one can display the cross-sections onto the screen as well as construct a realistic 3D-image with possibilities to "rotate" and "cut" the object model. On this model, one can calculate the internal morphological parameters.

3 INSTALLATION PROCEDURE

In case of damage during transportation or any defect in scanner operation, please contact your local Skyscan distributor (see www.skyscan.be for distributors). Alternatively, send the scanner directly to Skyscan, after contacting first, at following mail address:

Skyscan, Vluchtenburgstraat 3, Aartselaar B-2630, BELGIUM.
Email: info@skyscan.be, Tel: +32 3 877 57 05, Fax: +32 3 877 57 69

Only trained people by Skyscan are authorized to do service and reparations on the scanner!

For lifting and carrying, remove the four black plastic caps and put the supplied steel bars through the holes. One can carry the system with four persons.
Use the steel bars as handgrips.
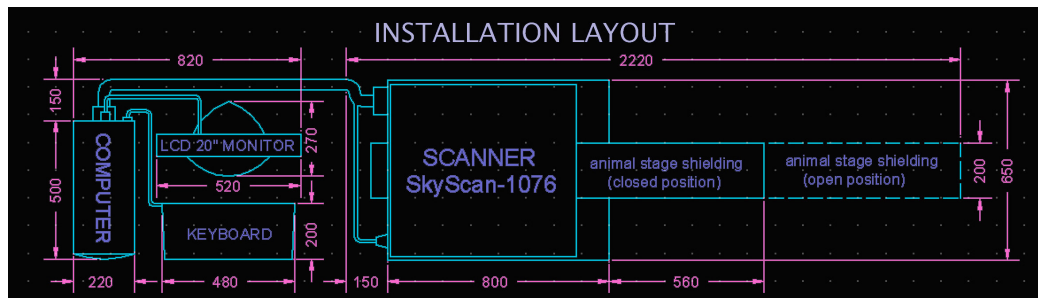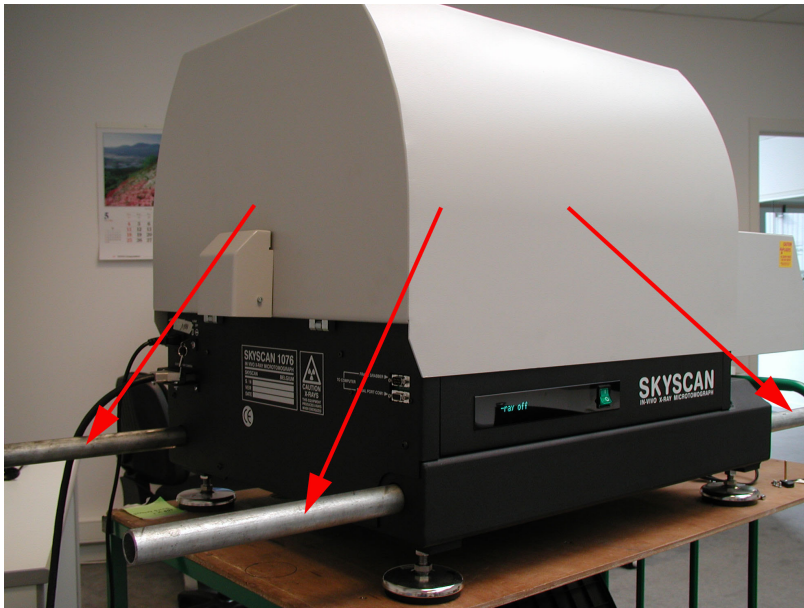
The table, on which the system will be put, should be strong enough to support a weight of 150 – 200 kg. The distance between the backside of the system and the wall should be at least 10 cm, necessary for the airflow.

3.1 Connections

To start the system one should make the connections shown in the figure below. All interconnections should be made before connection of the main power plug to 220-240 V AC or 100-130V AC.
Connect the scanner to the computer:

- Use a serial cable from the lowest serial connector on the system to the serial Port COM1 on the computer.
- Use second serial cable from the upper serial connector on the system to the analog frame grabber on the computer.
- Use video cable from digital x-ray camera connector on the system to the digital frame grabber on the computer.

The length of every signal cables is 2m. Use only the supplied signal cables.

The digital and analog frame-grabbers are pre-installed on the computer.

Interconnections for SkyScan1076:

Connections for computer:
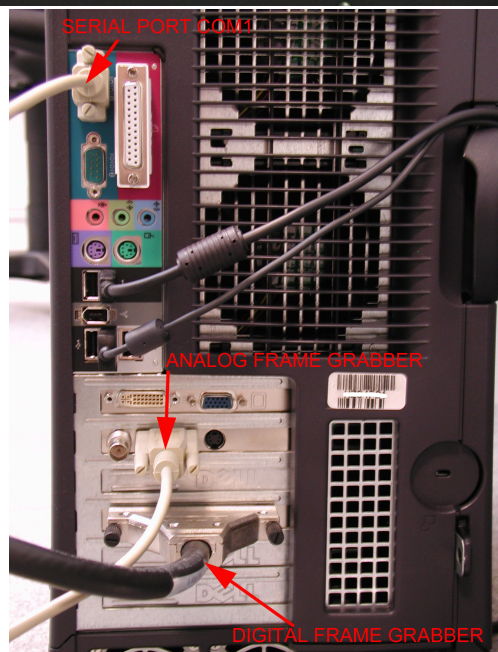
3.2 Power supply

Never connect 200-240V to a system meant for 100-130V and vice versa. Look to the label above the AC inlet (see picture on previous page).
The replaceable fuse is the same for both main voltages: type T rate 5A. Before replacement of the fuse, disconnect the power cable from the system.

After connection of power cable to the system and wall socket one can start system by power switch in the front panel.
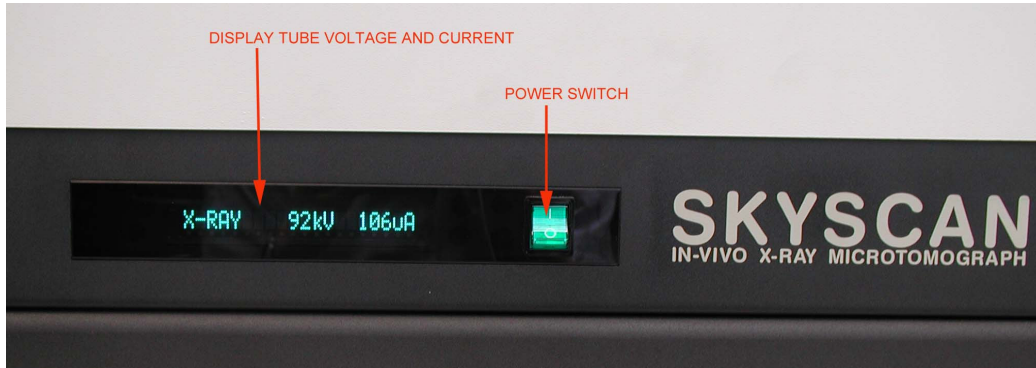
3.3 Starting up Skyscan1076

Switch the system on by the Power Switch.
Start the external computer also. The start procedure for computer can take couple of seconds with following start of "Windows 2000" or "Windows XP".

X-ray scanner should be always ON before starting control software in the external computer. Control and reconstruction software for Skyscan1076 system can be started from corresponding icon on the screen.

3.4 X-ray Key switch

A key switch for the x-ray source is located next to the AC inlet of the system. (See picture on previous page.)
For normal operations of scanner the switch "X-RAY" must be turned ON. You can keep the ON position of this switch continuously even when switching off and on the main power.
For security reasons the X-ray tube can be disabled or switched off by using the "X-RAY KEY SWITCH".

3.5 X-Ray Shutter

Normally the X-Ray shutter is always in the closed position. The X-ray shutter is open for acquiring one X-ray image or a scout scan and during scanning. The LED's indicate the X-Ray shutter position:

The X-ray shutter is open when the red LED lights up.

The X-ray shutter is closed when the green LED lights up.

4 SYSTEM CONTROL SOFTWARE

The software package for system control has the same operation principles as any other Windows-based program. One can select any functions by "mouse" from the menu / submenu in the top line and change modes and parameters by "mouse pressing" to the corresponding control areas or buttons. To start the Skyscan1076 control software double click on the corresponding icon:

After the program is started, an initialisation window is shown on the screen. During initialisation, the object-bed moves to the outside position. After initialisation, the shutter closes and the door become unlocked.
The start image on the screen contains the main menu bar and the control bar in the top of the screen.

4.1 Tube setting and start.

In the control bar, one can find the button to switch the tube ON or OFF:

To start X-ray tube push X-ray button:

Voltage and current will be set automatically corresponding to current pixel size and filter. Both values can be adjusted in "Options" -> "Scanning Modes" (see 5.3.4).
The Windows Taskbar in the bottom indicates the voltage and current settings of the X-ray tube. The X-ray voltage and current are also shown on the front panel display of the system.

In the case of first start or interrupt >8 hours after last usage of the x-ray source, a new information dialog will appear into the screen. It will be indication of aging (warming up) process for the soft start of the x-ray source. The aging allows keeping a very long tube lifetime. Typical time of an aging process is 15-17 minutes (for <1 month's pause in tube usage). In the case of big differences of voltage indications on the taskbar and in the front panel display of the system (>20% from maximum values) contact your supplier for service.
After aging, tube will be switched off automatically. One can switch it on in any time without extra delay. After tube start, the normal behaviour is an increasing of the acceleration voltage in the screen indicators and in the front panel display with following increasing of the current.

One can change the power settings of the tube by double clicking on the "x-ray generator on" indicator:

Following dialog box will be shown onto the screen:
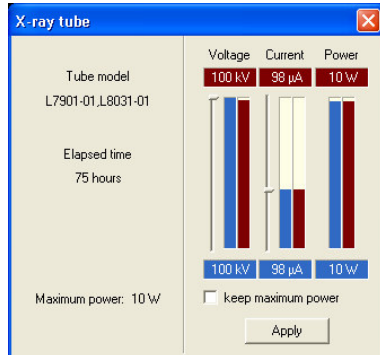
Left part of the dialog box shows information about the type of x-ray source, elapsed time (active emission) and maximum available power. Right part is used for indication of current values (red) and adjustment for new values (blue). New values can be applied by pressing to Apply button. The voltage reaches from 0 to 100 KV, the current from 0 to 250 µA.

The "Keep maximum power" option adjusts voltage from current or current from voltage to get maximum available power from the x-ray source.

For animal/object replacement press "Remove object" button in the control bar:

Object will be moved out of scanner. The X-Ray tube will switch off automatically and the door lock will become open.

The status bar in the bottom indicates the present position.

Now it's possible to open the door by hand.

4.2 Animal holders

There are two standard carbon-composite object beds supplied with the system.
The object bed with diameter 33 mm should be used for mice and the object bed with diameter 65 mm should be used for rats. Both have the same length of 400 mm and a maximum scanning length of 200 mm.

One can exchange the two beds easily.
First, remove the already installed object bed.
Start system and control software, after initialisation the shutter closes (green LED lights up). Open the door by hand. Close software and switch off the system.
Move the bed by hand so that the "PIN" is completely outside the system.

PIN
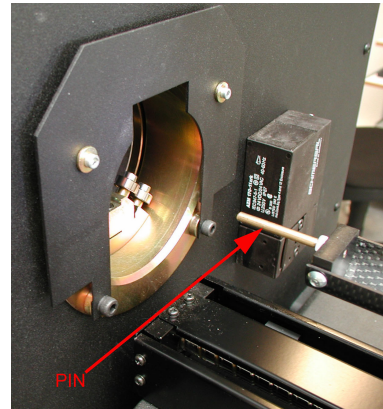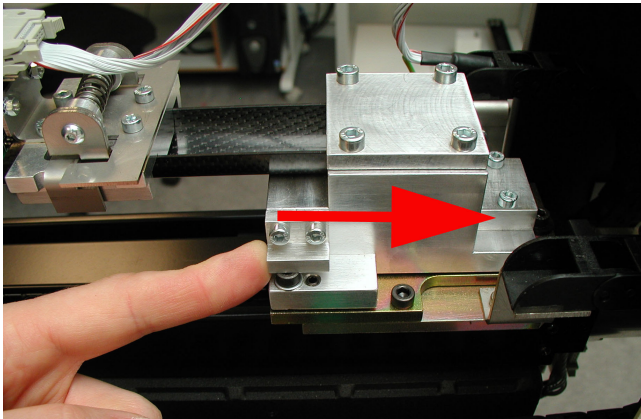
Remove the visual camera by pressing on the two sides of the fastening mechanism. Be careful that you don't damage the flat cable.
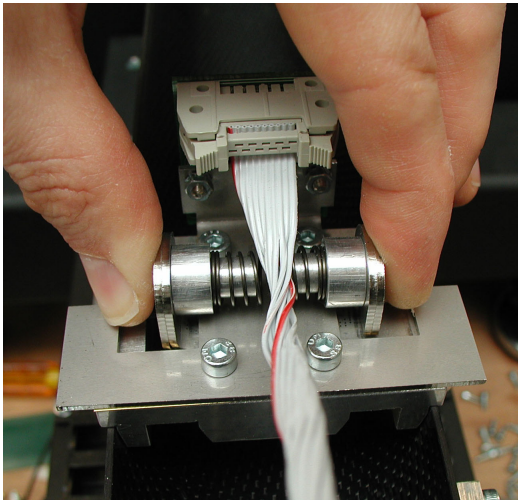
Put the visual camera on top of the door.
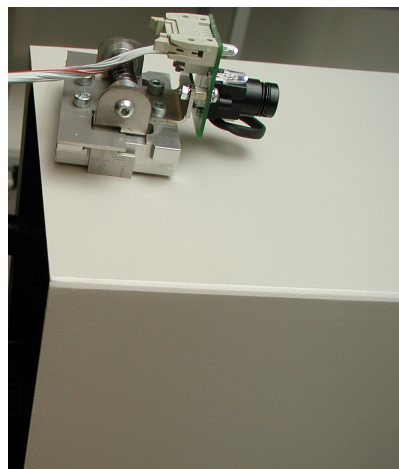
Remove the two screws on both sides of the bed.
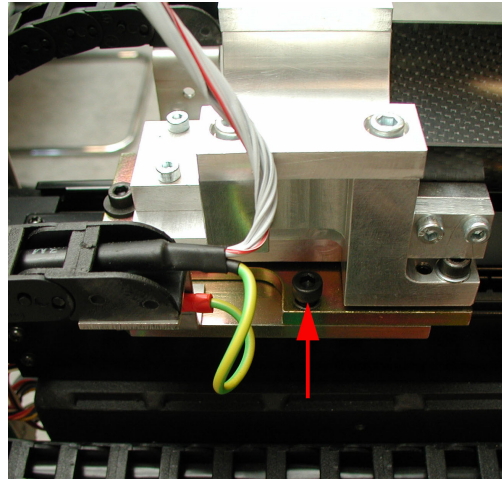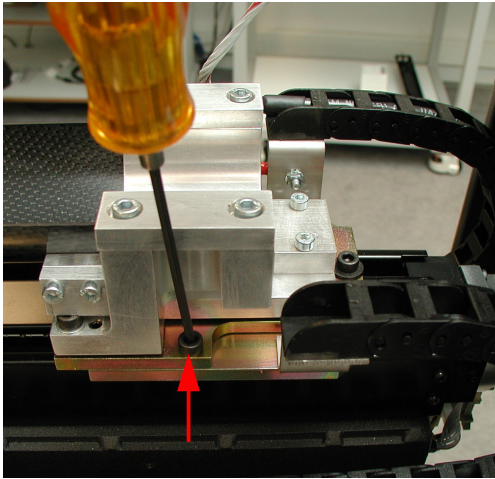
Now it is possible to remove the bed completely.

The installation of the small (33 mm) bed needs one extra support.
First, one has to install this extra support (see picture).
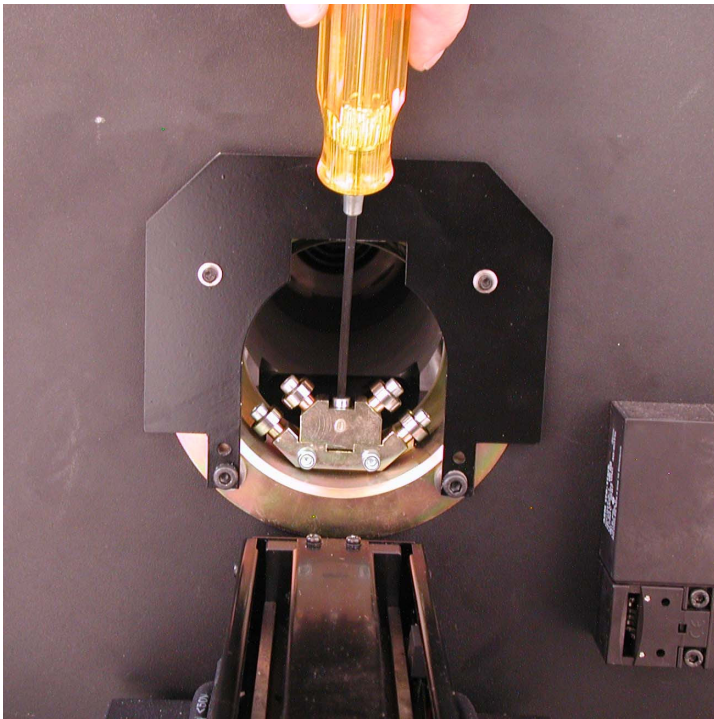
Remove this extra support for installation of big (65mm) bed.

Fasten the small bed with two screws on both sides and put the camera back on the small bed.
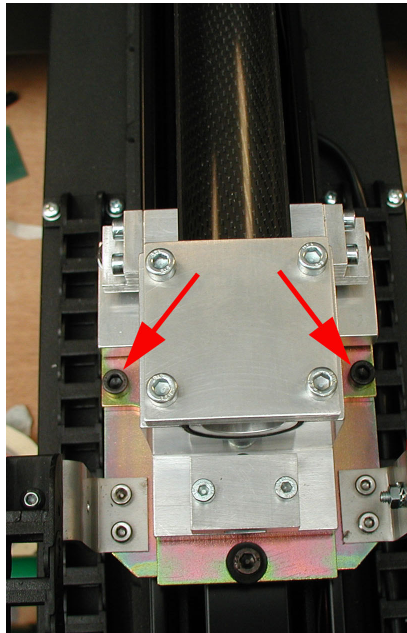
Close the door by hand and start system and software again.

4.3 Scout Scan and X-ray image

After installation of mouse or rat, close the door by hand.

Mouse on animal bed with physiological monitoring sensors:
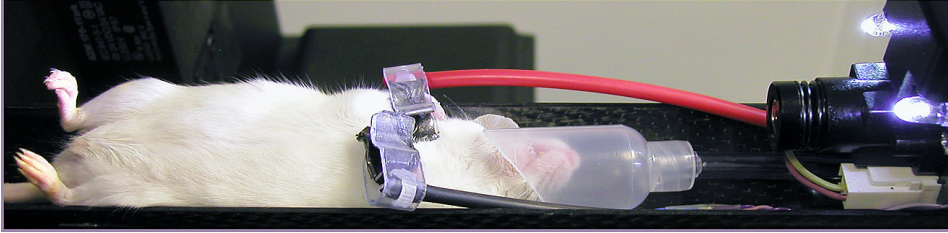
To start X-ray tube push X-ray button:

Voltage and current will be set automatically corresponding to current Filter and Pixel size. Now we can start the scout scan, it's a preview of the animal over the complete length of the scanning area.

          Start scout scan.

Scout image will be displayed in the left part of the screen. The scout scan can be stopped any time by corresponding dialog (Press Stop button) or program will try to create the image with maximum available length of the object bed. When scout scan will complete, one can select a place of the scanning in the scout view by double click to the left mouse button in the scout image. Object bed will be moved to corresponding position and one full-format x-ray image from this position will be acquired to the right part of the screen. The position of the bed is indicated in the status bar:

rotation: ready   position 97.750 mm
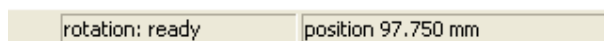
In the Options menu, one can select one of the three pre-installed filters or "no filter". If the filter position is changed, one can verify the X-ray image. In the control bar, one can find the button "Get X-ray image".

          Captures one image from X-ray camera

4.4 Start Scanning

If the position of the bed is correct and the preview image is good enough, one can start the acquisition.

Start scanning

Following dialog box will be displayed onto the screen:
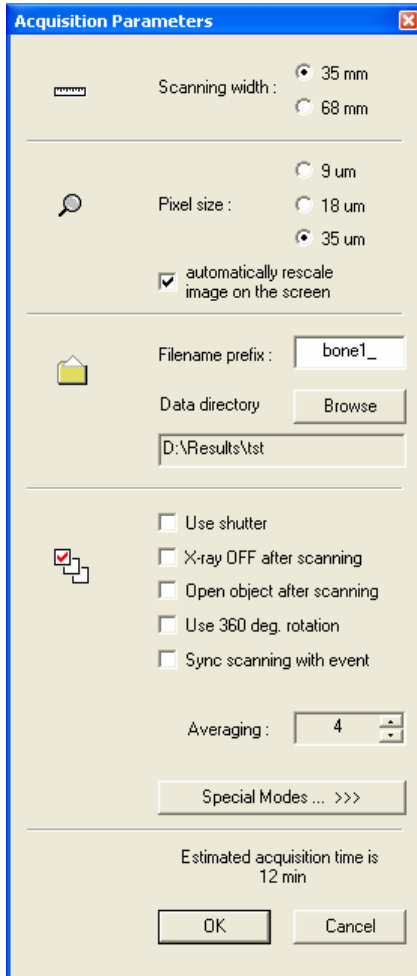
Scanning width should correspond to the size of scanning area (diameter of object bed).

Pixel size defines resolution for reconstructed images.

Filename prefix sets the front part of the filenames for acquired data and results of reconstruction. Use **Browse** button to select another data folder or to create new one.

For information that is more detailed see 5.2.8 or 5.3.1.

Set "Scanning width" corresponding to the used animal bed and select desired "Pixel size".
Now one can type a new Filename prefix for the dataset and select or create a new data directory.
After selection of the desired acquisition parameters (take into account the "Estimated acquisition time" in the bottom of the dialog window), press OK, to start the acquisition.

After scanning, unlock the door by , open it by hand and remove the object or select another scanning place by double clicking to the scout view.

4.5 Reconstruction

One can start the external Cone-beam reconstruction program from the Toolbar:

    Starts reconstruction program

Alternatively, one can start the Cone-beam program from the shortcut on the desktop:

Following dialog box allows choosing data set for sending to reconstruction.
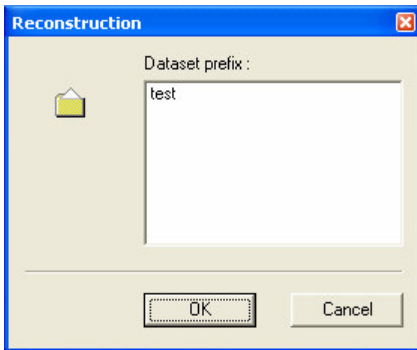
Click on the dataset prefix you want to reconstruct and press OK.

Start single reconstruction (choose fastest). After reconstruction of one crossection, set Crossection to Image values (Auto or other values)

Options -> Reconstruction : choose value for Beam hardening correction, smoothing… and check it. Press button and start single. Set Crossection to Image values again.
To invert images to negative one use and choose Negative.
Press button and choose Top, Bottom and Step (normally 2-10). Start All.

After reconstruction show results by as section-by-section movie.
Use for on-line help on the reconstruction program.

4.6 3D-Reconstruction

To convert the reconstructed dataset (bitmap files) into a 3D-image and to calculate the internal morphological parameters one can use the external program "3D-Creator".
One can start the program from the shortcut on the desktop:

Alternatively, one can start the 3D-creator from the 3D rendering command inside the cone-beam reconstruction program:

All details of using this program can be found in the special Manual for "3D-Creator" and "Format Converter" programs.

In the case of big number of complicated 1024x1024 cross sections we recommend to reduce image format in dataset till 512x512 or 256x256. It can be done by external program named "Format Converter" or it starts automatically from the 3D rendering command inside the cone-beam reconstruction program. One should select the original dataset and name for new one with reduced format as well as a reduction ratio (1/2, ¼…). New dataset with reduced format can be used for the creation of realistic 3D-images by "3D-Creator" program.

5 MENU AND SUBMENU FUNCTIONS

5.1 General layout of the main menu

The top menu bar in the screen gives access to all system functions. Let's consider all menu / submenu possibilities in more details.

Commands

Actions menu
Options menu
Help menu

It is a standard Windows main menu and operates thus in exactly the same way.
Selecting items from the menu, running through and opening of the sub-menus and applications is done as any standard Windows program.

5.2 The Actions menu

Actions	Options	Help		
Open Image ...			Ctrl+O	-> Opens an existing image file.
Save Image ...			Ctrl+S	-> Saves the image to disk
Print Image			▶	-> Displays submenu
Delete Dataset			▶	-> Displays submenu
Grab X-Ray Image				-> Acquires one image from X-ray camera
Show Visual Image				-> Starts external program for animal imaging and physiological monitoring
Start Scout Scan				-> Starts scout scan
Start Scanning for Reconstruction ...				-> Starts scanning
Remove Object				-> Removes object from scanner
Start Reconstruction ...				-> Starts reconstruction program
Set Object Position ...				-> Sets the position of the bed by numerical value
Set View Angle ...				-> Sets the rotational position of the source-detector pair around the object
Exit				-> Closes the control program

5.2.1 Open Image command

Use this command to open an existing image file.
Following "Open" dialog will appear onto the screen.
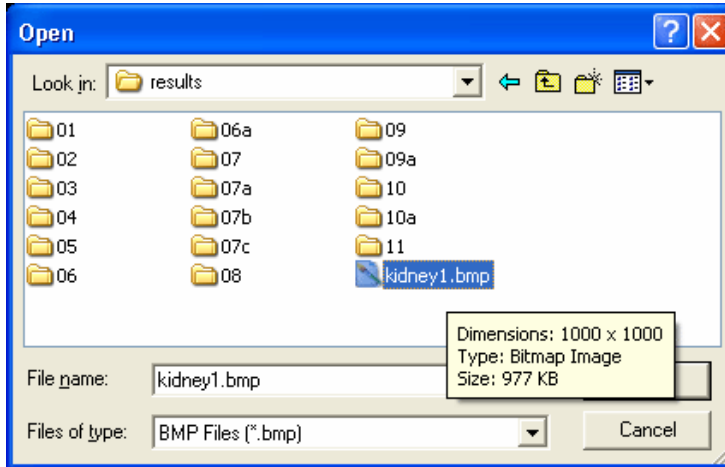
The following options allow you to specify which file to open:

File Name

Type or select the filename you want to open. This box lists files with the
extension you select in the List Files of Type box. You can open the file by
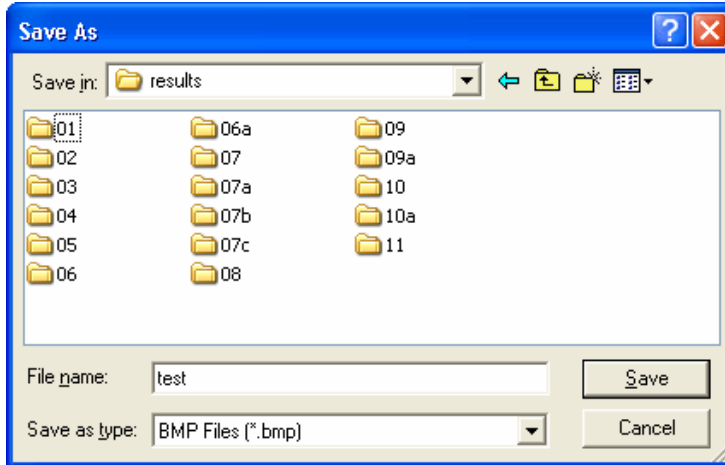clicking twice to corresponding filename in the table or list.

List Files of Type

Select the type of file you want to open BMP-files or TIFF-files

Shortcuts

Toolbar:
Keys: CTRL+O

5.2.2 Save Image command

Use this command to save the image to the disk. The program will display the following "Save" dialog box:

The following options allow you to specify the name and type of the file you're about to save:

File Name

Type a new filename to save a document with a different name. You can select existing filename to replace existing image. Program will ask you a confirmation to delete existing file

Save as type

Select the type of file you want to save: 8-bit BMP-file or 16-bit TIFF-file

Shortcuts

Toolbar:
Keys: CTRL+S

_____

5.2.3 Print image command and submenu

The Print submenu offers the following commands:

Print... Ctrl+P	-> Prints a document
Print Preview	-> Displays the image as it would appear when printed
Print Setup...	-> Selects a printer and a printer connection
Page Layout...	-> Sets the print layout

Print command

Use this command to print a document. This command presents a Print dialog box, where you may specify the range of pages to be printed, the number of copies, the destination printer, and other printer setup options.

Shortcut

Toolbar:

Print Preview command

Use this command to display the image as it would appear when printed. When you choose this command, the main window will be replaced with a print preview window in which one or two pages will be displayed in their printed format. The print preview toolbar offers you options to view either one or two pages at a time; move back and forth through the document; zoom in and out of pages; and initiate a print job.

Print Setup command

Use this command to select a printer and a printer connection. This command presents a Print Setup dialog box, where you specify the printer and its connection.

Print Layout

Use this command to set print layout. Following dialog window will be displayed:
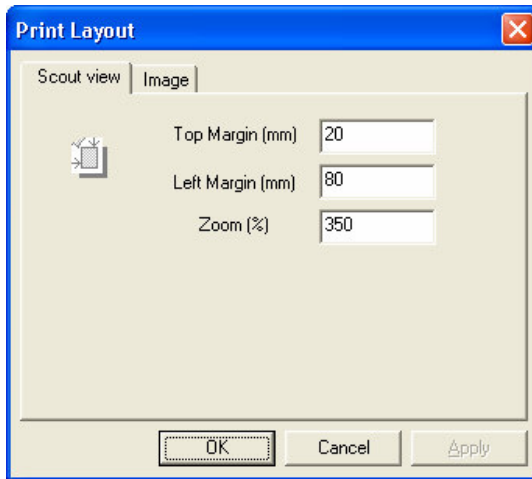
Print layout for scout view contains adjustments for printing position (margins) on the page and image scale as a zoom from original (corresponding to object size) format.
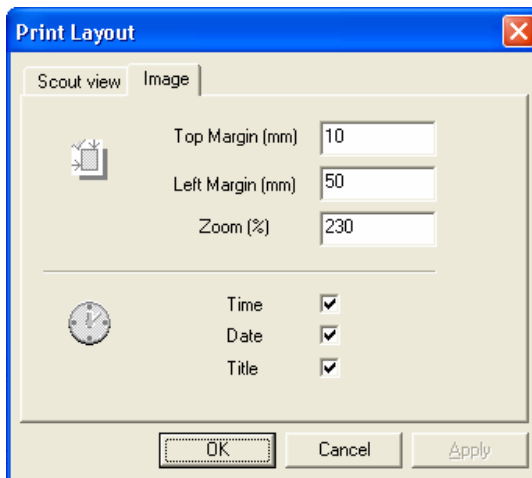
Print layout for image contains the same adjustments as above plus possibilities to include in the printing form some additional information, such as time, date and title of the image
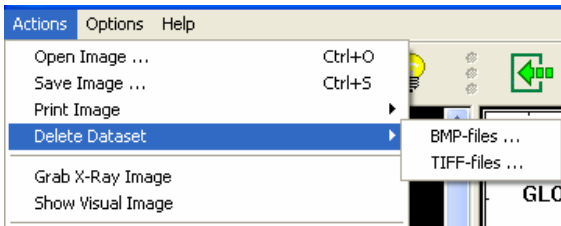
5.2.4 Delete Dataset and submenu

Delete Dataset -> BMP-files
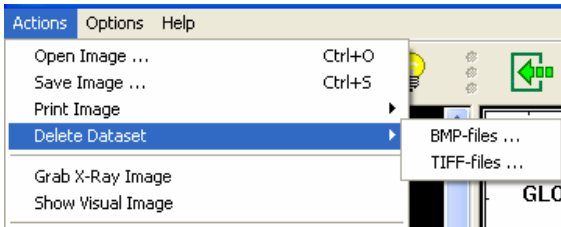
Use this command to remove full set of reconstructed results with selected prefix from the disk.

Delete Dataset -> TIFF-files

Use this command to remove full set of acquired projection images with selected prefix from the disk.

5.2.5 Grab X-Ray Image

Use this command to acquire one image from X-ray camera. One can use "Zoom In" and "Zoom Out" options in the context menu, activated by right mouse button, to adjust necessary image size on the screen after acquisition.

Shortcut

Toolbar:
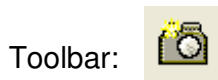
The "Grab X-Ray Image" function is also accessible from additional menu by clicking to the right mouse button in the X-Ray image on the screen

5.2.6 Show Visual Image and physiological monitoring

This command will start the external program for animal imaging and physiological monitoring. Visual image of the object can be shown in one of three sizes.
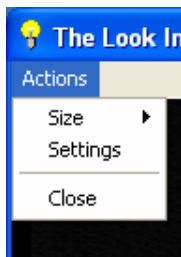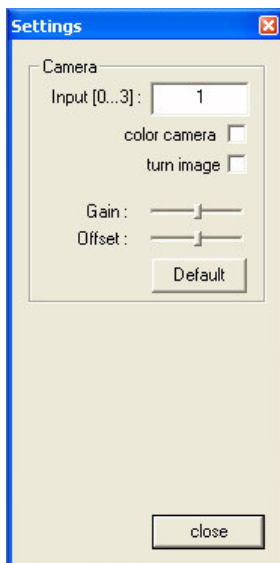
Shortcuts

Toolbar:

Icon on desktop:

5.2.6.1 The Actions menu
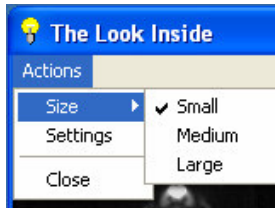
-> Use this command to select the size of the visual image shown on the screen.

-> Use this command to select the settings for the visual camera

-> Changes the camera Input (for skyscan1076 choose 1)

-> Change to color image if color camera is available

-> Turns the image upside down

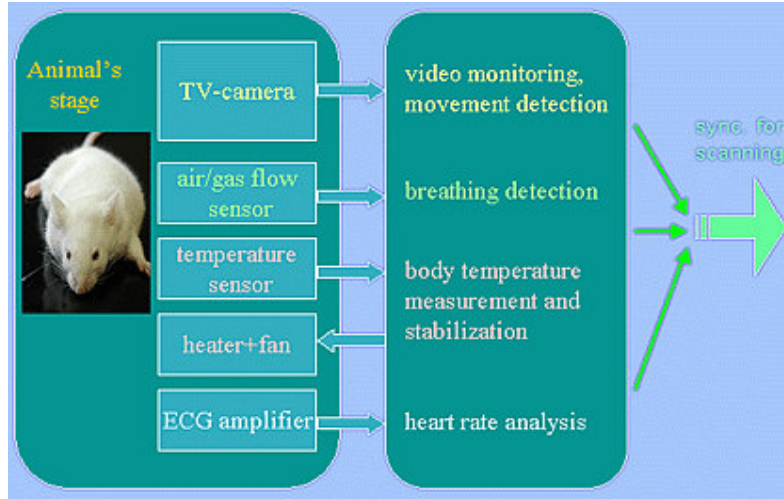-> Adjusts the Gain (contrast) of the visual image

-> Adjusts the Offset (brightness) of the visual image

-> Selects default settings for Gain (=1) and Offset (=0)

-> Closes the "Settings" window

5.2.6.2 Physiological monitoring subsystem

Subsystem for physiological monitoring of the small laboratory animals during scanning contains following parts:


The TV-camera in the animal bed produces real-time visual image. Operator can see the animal on the computer screen in any time including x-ray scanning. Special software detects local movement of the animal body. This information can be used for scanning synchronization with animal's breathing.

Air / gas flow sensor can be connected to the mask in the animal bed for direct breathing detection. The breathing sensor reacts to the changes of the direction in the air or gas flow. Typical sensitivity is 0.002 mBar differential pressure or 1sccm (1 cm3 of gas per minute) airflow. Sensor can be connected in between of the gas anaesthesia system and the pipe to the animal's mask.

Temperature sensor on the animal bed measures the body temperature with accuracy of 0.2-0.3C in the range of 18C...40C. Physiological monitoring software shows this information on the screen as a curve and numerical value. Operator can adjust a specific temperature for switching of animal heating by airflow in the case of measured temperature decreases under this value.

ECG-amplifier connected to the animal by special non-metallic wires and electrodes. It makes all connections practically invisible in x-ray image. Heart pulses and numerical value of the heart rate are indicated onto the screen.

**Screen copy of the Skyscan-1076 software during scanning with Physiological Monitoring.**
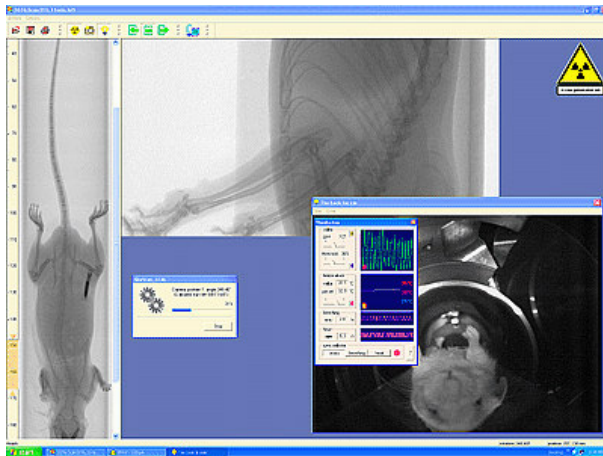
Left part: scout view (up to 200mm) with selection of the scanning position.
Central part: real-time x-ray image and progress indicator;
Right bottom part: physiological monitoring and real-time visual image of the animal.

5.2.6.3 Physiological Monitoring window onto the screen

Physiological monitoring information will be shown in additional window in the top of the visual image. Operator can switch ON the Physiological monitoring window by typing "PM" on the keyboard (capital letters: "shift" + pm) during operation of visual camera. To close Physiological monitoring window press: ⊠
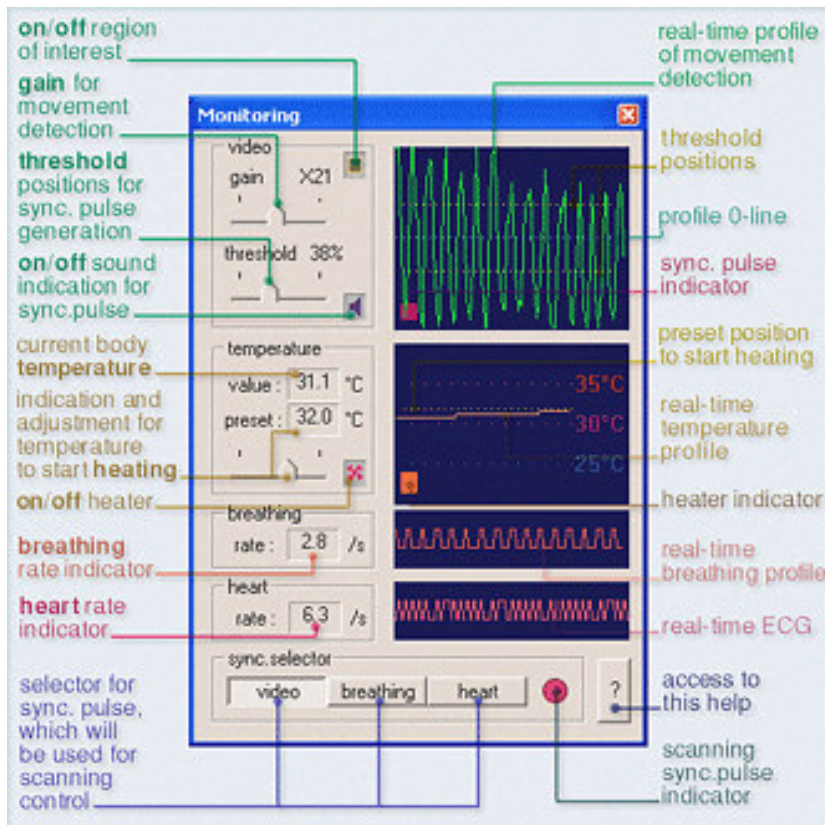
In the Actions Menu of the "LookIn" program additional settings for temperature will appear during Physiological monitoring:
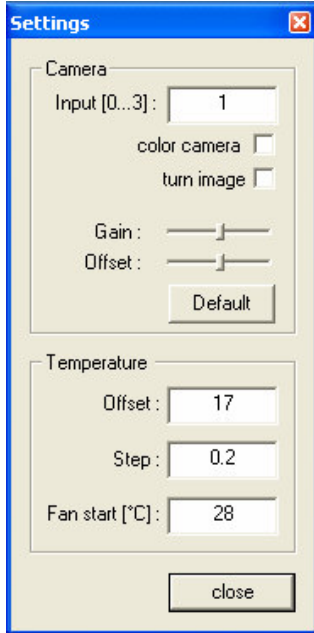
-> Add this number for calibration of temperature sensor.

-> Multiply with this number for calibration of temperature sensor.

-> Selects the minimum measured temperature value from which the fan/heating should start.

Adjustments (calibration) of "Offset" and "Step are normally only necessary after replacement of the temperature sensor.

Adjustment of "Fan start" is only necessary if the room temperature is higher than 28°C.

5.2.7 Start Scout Scan

Use this command to start scout scan. Scout image will be displayed in the left part of the screen. The scout scan can be stopped in any time by corresponding dialog or program will try to create the image with maximum available length of the object bed. When scout scan will complete, one can select a place of the scanning in the scout view by double click to the left mouse button. Object bed will be moved to corresponding position and one full-format x-ray image from this position will be acquired to the right part of the screen.
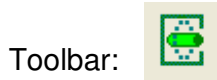
Shortcut
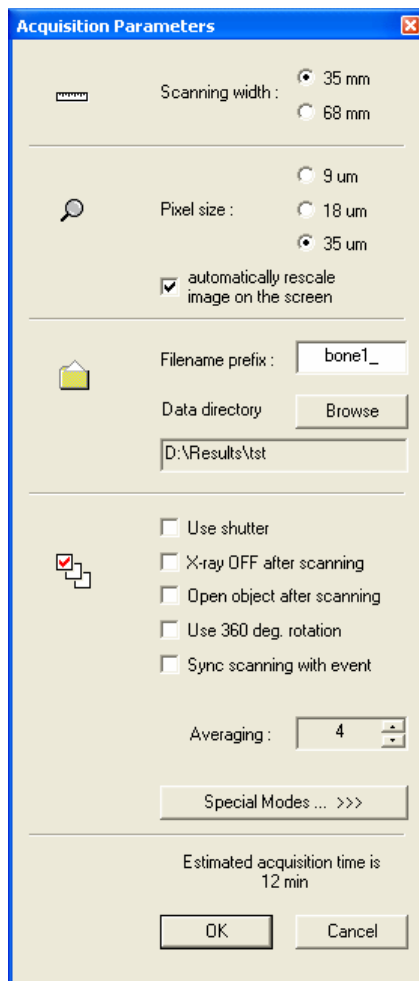
Toolbar:

5.2.8 Start Scanning for Reconstruction

Use this command to start scanning to acquire data for following reconstruction. The program displays the Acquisition Parameters dialog box to adjust all necessary settings.

Shortcut

Toolbar:

Acquisition Parameters dialog box

Use this command to set the acquisition parameters. Following dialog box will be displayed onto the screen:

Scanning width should correspond to the size of scanning area (diameter of object bed). Sometimes possible to scan the central part of the big size object with small Scan width (central part will be scanned) -> Use "Object size > Field of view" option in the reconstruction program in this case.

Pixel size defines resolution for reconstructed images. Big pixel size allows accelerate acquisition and reconstruction process against spatial resolution. Scanning with small pixel size improve spatial resolution, but may take significant scanning and reconstruction time and required a lot of disk space.

Filename prefix sets the front part of the filenames for acquired data and results of reconstruction. Use **Browse** button to select another data folder or to create new one.

Use shutter option increase acquisition time by 50%, but reduce x-ray dose to the animal near twice.

X-ray OFF after scanning allows automating switching off x-ray source in the end of the scanning cycle.
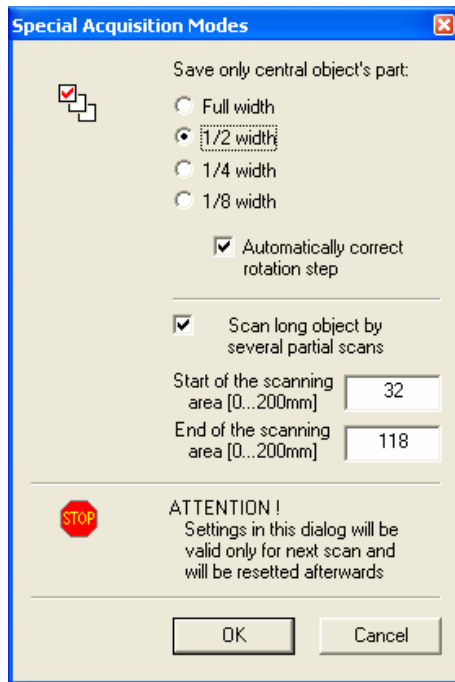
Open object after scanning remove object bed from the scanner in the end of scanning cycle and open the lock for access to the animal.

Use 360deg. rotation create data sets with improved reconstruction quality in the top and in the bottom of the field of view, but scanning time will increased by near twice.

Sync scanning with event allows synchronize all shots in the acquisition with external event, selected in Physiological Monitoring subsystem (breathing, local body movement, etc.). In the case of selection for this option one can adjust in the following slider necessary delay of the acquisition start from the sync pulse.

Averaging of several images in each rotation position reduce noise, but takes more time for scanning.

Special modes button open next dialog window with parameters settings for nonstandard scanning modes.

Save only central part option allow acquiring projection images for reconstruction of specific central part of the object, which can be 1/2, 1/4, 1/8 of the selected scanning width. It can reduce necessary disk space for scanning of small objects located in the central part of the specimen bed. Use "Object size > field of view" during reconstruction to avoid influence of the object bed and surrounded specimen parts.

Automatically correct rotation step option can reduce acquisition time by increasing of scanning angular steps according to reduction in the scanning area.

Scan long object by several partial scans mode allows automatic combining several partial scans along the object into one long reconstructed dataset. Operator can select start and end position of the long scanning area according to the ruler aside of the scout view. During scanning several partial datasets will be created. In the start of reconstruction corresponding dialog box will allow making choice to reconstruct one partial dataset or automatically reconstruct and connect all available datasets inside the long scan. We recommend to reconstruct one cross section from one partial dataset to set all parameters, then – close reconstruction program and select reconstruction again with choice of reconstruction for all available datasets inside the long scan. In this case, all partial datasets will be reconstructed with the settings, taken from the first reconstruction you made.

5.2.9 Remove Object

Use this command to remove object from the scanner. Object will be moved out off scanner and door lock will be open.

Shortcut

Toolbar:

5.2.10 Start Reconstruction

Use this command to start reconstruction program. Following dialog allows choosing data set for sending to reconstruction.

Shortcut

Toolbar:

5.2.11 Set Object Position

Use this command to set the position of the object bed inside scanning area by numerical value. Typical range of acceptable positions is 0…200mm. The "0-position" corresponds to the scanning close to the front end of the object bed in the field of view. Negative positions are corresponding to the object outside the scanner, Positive positions corresponding to the object inside the scanner.

Following dialog will be shown to select necessary object position:
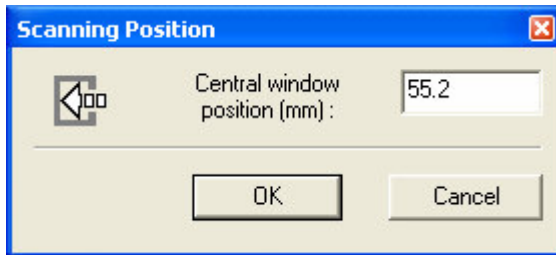
5.2.12 Set View Angle

Use this command to set the rotational position of the source-detector pair around the object. Typical range of acceptable positions is 0…360 degrees.

Following dialog will be shown to select necessary viewing angle:

5.2.13 Exit command

Use this command to close the control program for SkyScan-1076 Micro-CT instrument. You can also use the Exit command on the Actions menu.

Shortcuts

Mouse:
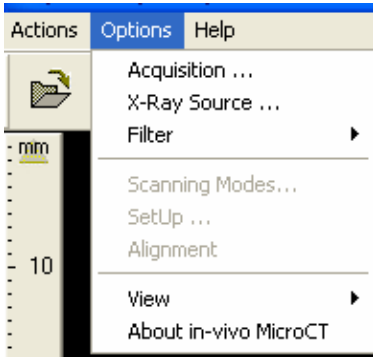
Double-clicking to the left top corner of the program window is the same as choosing the Exit command.
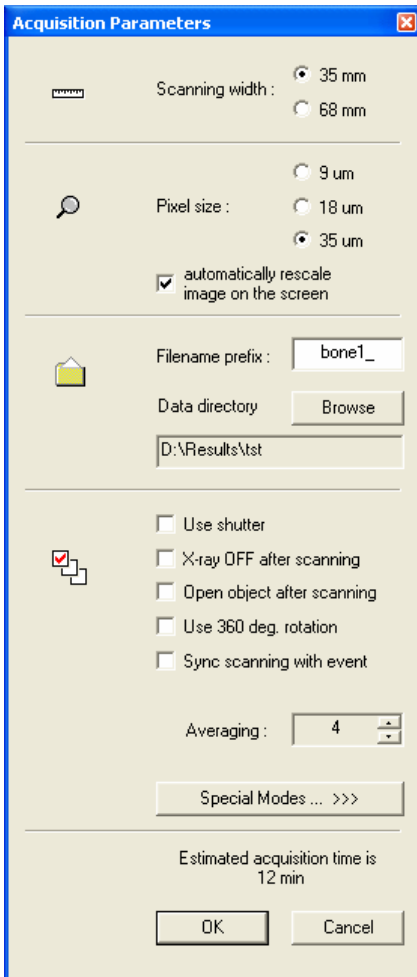

Keys: ALT+F4

5.3 The Options menu

-> Sets the acquisition parameters

-> Adjusts the settings for the X-Ray source

-> Selects corresponding physical filter

-> Access to the settings for all possible acquisition modes

-> Modifies geometrical settings of the scanner.

-> Starts automatic alignment procedure of the scanner

-> Displays submenu

-> Displays the copyright notice and version number of the control software

5.3.1 Acquisition

Use this command to set the acquisition parameters. Following dialog box will be displayed onto the screen:

Scanning width should correspond to the size of scanning area (diameter of object bed). Sometimes possible to scan the central part of the big size object with small Scan width (central part will be scanned) -> Use "Object size > Field of view" option in the reconstruction program in this case.

Pixel size defines resolution for reconstructed images. Big pixel size allows accelerate acquisition and reconstruction process against spatial resolution. Scanning with small pixel size improve spatial resolution, but may take significant scanning and reconstruction time and required a lot of disk space.

Filename prefix sets the front part of the filenames for acquired data and results of reconstruction. Use **Browse** button to select another data folder or to create new one.

Use shutter option increase acquisition time by 50%, but reduce x-ray dose to the animal near twice.

X-ray OFF after scanning allows automating switching off x-ray source in the end of the scanning cycle.

Open object after scanning remove object bed from the scanner in the end of scanning cycle and open the lock for access to the animal.
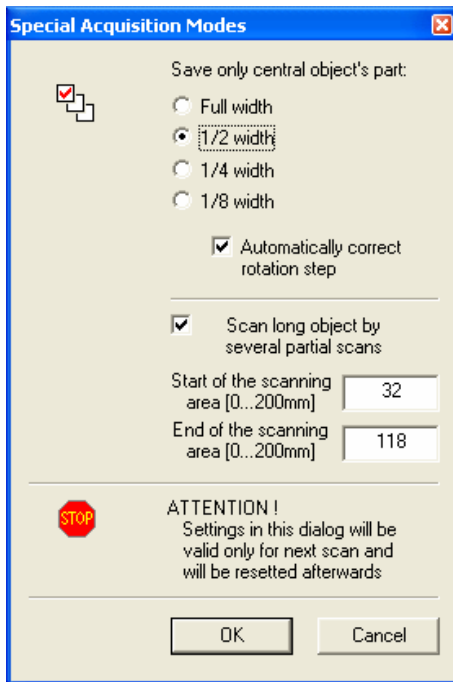
Use 360deg. rotation create data sets with improved reconstruction quality in the top and in the bottom of the field of view, but scanning time will increased by near twice.

Sync scanning with event allows synchronize all shots in the acquisition with external event, selected in Physiological Monitoring subsystem (breathing, local body movement, etc.). In the case of selection for this option one can adjust in the following slider necessary delay of the acquisition start from the sync pulse.

Averaging of several images in each rotation position reduce noise, but takes more time for scanning.

Special modes button open next dialog window with parameters settings for nonstandard scanning modes.

Save only central part option allow acquiring projection images for reconstruction of specific central part of the object, which can be 1/2, 1/4, 1/8 of the selected scanning width. It can reduce necessary disk space for scanning of small objects located in the central part of the specimen bed. Use "Object size > field of view" during reconstruction to avoid influence of the object bed and surrounded specimen parts.

Automatically correct rotation step option can reduce acquisition time by increasing of scanning angular steps according to reduction in the scanning area.

Scan long object by several partial scans mode allows automatic combining several partial scans along the object into one long reconstructed dataset. Operator can select start and end position of the long scanning area according to the ruler aside of the scout view. During scanning several partial datasets will be created. In the start of reconstruction corresponding dialog box will allow making choice to reconstruct one partial dataset or automatically reconstruct and connect all available datasets inside the long scan. We recommend to reconstruct one cross section from one partial dataset to set all parameters, then – close reconstruction program and select reconstruction again with choice of reconstruction for all available datasets inside the long scan. In this case, all partial datasets will be reconstructed with the settings, taken from the first reconstruction you made.

5.3.2 X-Ray Source

Use this command to adjust settings for x-ray source. Following dialog box will be shown onto the screen:
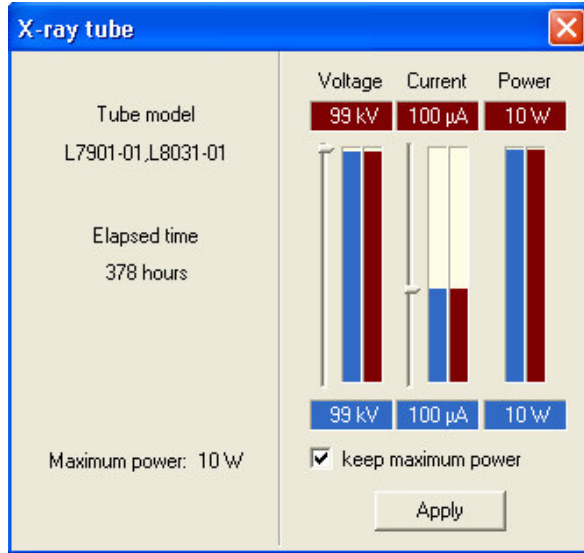

Left part of the dialog box shows information about the type of x-ray source, elapsed time (active emission) and maximum available power.

Right part is used for indication of current values (red) and adjustment for new values (blue). New values can be applied by pressing to Apply button.

Keep "Maximum power" option adjusts voltage from current or current from voltage to get maximum available power from the x-ray source.

5.3.3 Filter

Use this command to select corresponding physical filter in the X-ray beam. Acquisition parameters, camera settings and all correction maps will be adjusted corresponding to new selected filter.
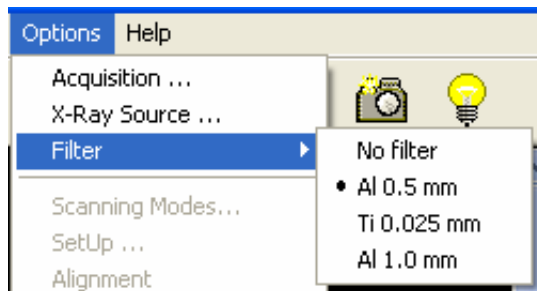
5.3.4 Scanning Modes

This command is only accessible by pressing "Ctrl"+"Alt"+"Shift"+"s". Pressing the password combination again will close access to this command.
Use this command for access to the settings for all possible acquisition modes:
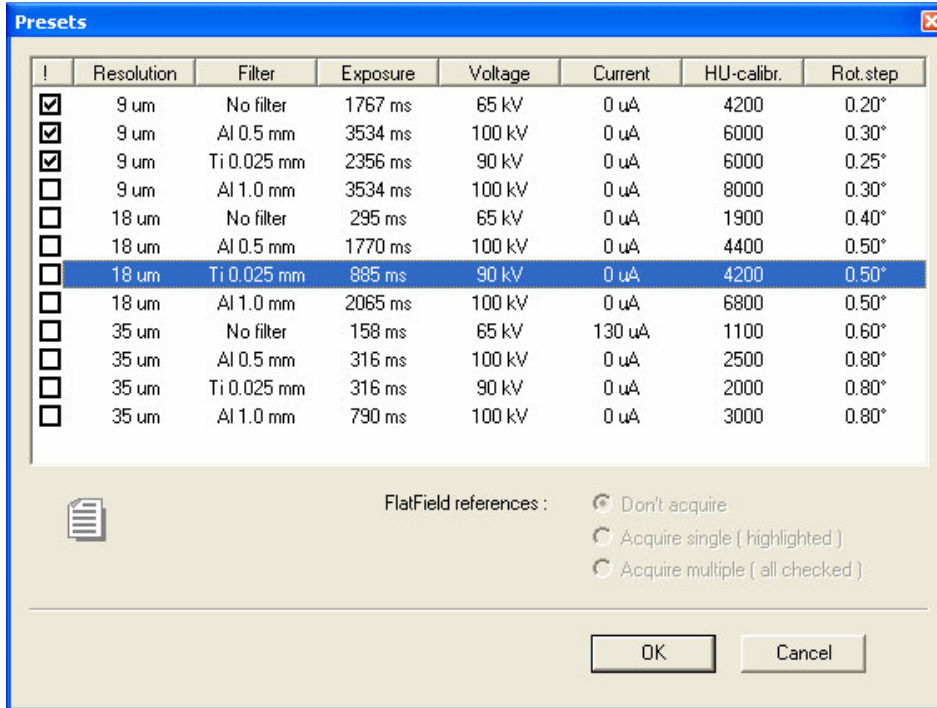
Top part of the dialog window contains a table with 12 acquisition modes for 4 available filters and 3 possible pixel sizes. For sorting according to specific parameter click twice to the corresponding column title.
To modify any particular scanning mode click twice to the corresponding line in the table. Following dialog will allow modification of all listed parameters for selected mode.
One of the scanning modes can be selected (highlighted) to update flat-field correction maps after modification in acquisition parameters. In the case of modifications in several modes, use check boxes in the front of corresponding lines for indication of the modes to update.
Flat-field references can be acquired for one or several selected modes by choosing corresponding option in the bottom of dialog window.

5.3.5 Set-up

This command is only accessible by pressing "Ctrl"+"Alt"+"Shift"+"s". Pressing the password combination again will close access to this command.

Use this command to modify geometrical settings of the scanner.

ATTENTION!
Wrong values in one or several parameters can cause the scanner damages, image distortions or incorrect reconstruction results.

Pixel size adjusts the size of the image pixel in the object plane for the maximum camera resolution.
Optical axis shows position of the scanning line (0…2000) where x-ray beam from the source hit the camera with exactly 90 degrees angle.
Horizontal overlap (in number of pixels) adjusts overlap of two images in the case of the scanning with biggest object size. In the overlapped area two images from the different camera positions will be combined by averaging with linear weight function.
Vertical overlap (in number of lines) adjusts image overlaps during scout scan. Image rotation (in degrees) defines the rotation angle of the two partial images in the scanning with biggest field of view for creation of one continuous image.
Source-Detector distance defines the distance in mm from the emission point inside the x-ray source to the scintillator layer inside the camera.
Source-Object distance is a distance in mm from the emission point inside the x-ray source to the rotation axes of the source-detector pair.
**Flat-field references for current mode allows** acquire, load or clean the reference maps for flat-field correction for the current camera mode in the central camera position. Acquired reference fields will be immediately automatically saved to the disk.

5.3.6 Alignment

This command is only accessible by pressing "Ctrl"+"Alt"+"Shift"+"s". Pressing the password combination again will close access to this command.

Use this command to start automatic alignment procedure of the scanner.

5.3.7 View
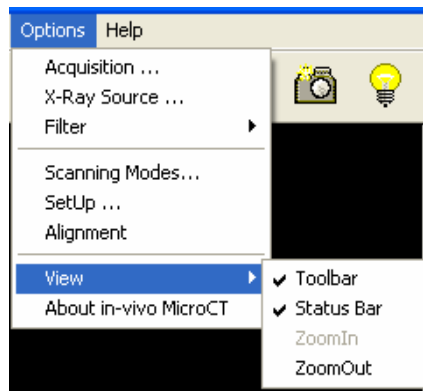
-> Shows or hides the Toolbar
-> Shows or hides the Status bar
-> Use this command to zoom in X-Ray image
-> Use this command to zoom out X-Ray image

5.3.7.1 Toolbar command

Use this command to display and hide the Toolbar, which includes buttons for some of the most common commands in the control program for Skyscan-1076 micro-CT scanner. A check mark appears next to the menu item when the Toolbar is displayed.

Toolbar:
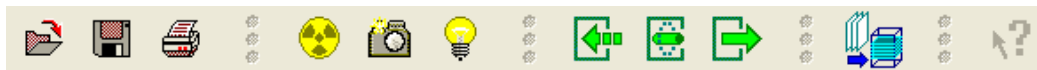
The toolbar is displayed across the top of the application window, below the menu bar. The toolbar provides quick mouse access to many tools used for scanner control and data access.

Click	To
	Open an image.
	Save the active image.
	Print the active image.
	Start or stop the x-ray source.
	Acquire x-ray image.
	Access to visual and physiological monitoring program.
	Start scout scanning.
	Start data acquisition for reconstruction.
	Remove the object from the scanner and open specimen chamber.
	Start reconstruction program.
	Access to context help.

5.3.7.2 Status Bar command

Use this command to display and hide the Status Bar, which shows the position of the object and source-detector pair in the current moment. A check mark appears next to the menu item when the Status Bar is displayed.

Status Bar:

rotation:12.90°	position 25.500 mm	
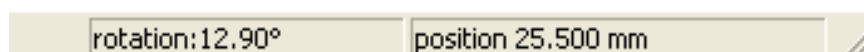
Status Bar shows the position of the object inside the scanner in mm and source-detector pair angular position in degrees in the current moment.
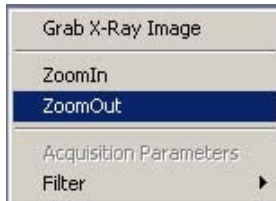
5.3.7.3. Zoom In

Use this command to zoom in image from X-ray camera onto the screen.

5.3.7.4 Zoom Out

Use this command to zoom out image from X-ray camera onto the screen.

The Zoom In or Zoom Out functions are also accessible from additional menu by clicking to the right mouse button in the X-Ray image on the screen.

5.3.8 About in-vivo Micro-CT

Use this command to display the copyright notice and version number of your copy of the control software for the in-vivo micro-CT scanner Skyscan-1076.

5.4 Help

The Help menu offers the access to the help-information available on this software package.