

CHAPTER 7 ROUTINE MODE AND TOOLS

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ROUTINEMODE AND TOOLS
Contents

Carl Zeiss

LSM 510
LSM 510 META

7 ROUTINE MODE AND TOOLS

7.1 Routine Mode

7.1.1 General

The Routine Mode of the LSM 510 software permits the fast and easy acquisition of scanning images by using time-tested **Standard Examination Methods** or by **User Defined Examination Methods**.


Standard Examination Methods are included in the LSM 510 software package and must only be activated once during the first application of the routine mode.

User Defined Examination Methods are methods which were already created and optimized in the Expert Mode. If **User Defined Examination Methods** are also to be used in the Routine Mode, they must be exported in the Routine Mode first. Apart from **User Defined Examination Methods**, stacks and time series can also be exported to the routine mode in addition to simple frames.

Images scanned in the Routine Mode are displayed in the same image windows as in the Expert mode. The tools for image processing and the image display functions are identical.

7.1.2 Starting the Routine Mode

- Start the LSM program (see chapter 4).

 If you want to change from the Expert Mode to the Routine Mode and vice versa, close all the windows first.

- Activate the **Scan New Images** button in the LSM 510 Switchboard menu.
- Click on the **Start Routine Mode** button.
 - The **Routine Mode** window will appear.



Fig. 7-1 LSM 510 Switchboard menu

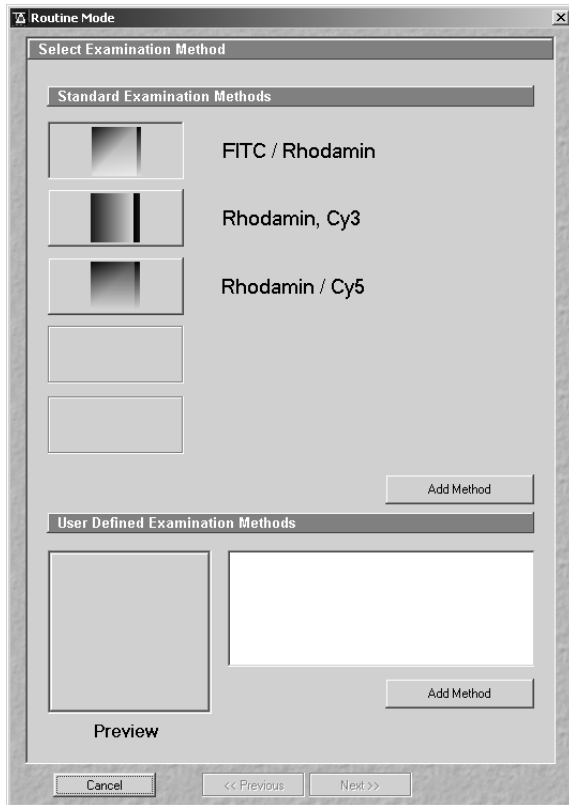



Fig. 7-2 Routine Mode - Select Examination Method window (first application)

In addition to the three standard examination methods contained in the LSM 510 software package, any required number of further examination methods can be exported from the Expert Mode to the Routine Mode.

 The three standard examination modes are only available if channel 1 is a Non-META PMT channel.

The maximum of five, freely selectable methods can be assigned directly to the **Standard Examination Methods** buttons.

- Click on the **Add method** button.
 - The **Select Method To Add To Standard** window appears on the screen.
- Use the checkboxes in the **Methods** frame to select / deselect a table.
- Click on the **Select All** button to select all current methods.
- Click on the **Clear** button to deselect all current methods.
- Exit the **Select Method To Add To Standard** by clicking on the **Close** button.

All the other methods required must be activated in the **User-Defined Examination Methods** selection box.

7.1.2.1 Activate User-Defined Examination Methods in the Routine Mode

Requirements:

The examination method to be imported has been exported from the Expert Mode to the Routine Mode using the **Export RM** function in the **Options** menu (see chapter 4).

- Start the Routine Mode.
- Click on the **Add method** button.
 - The **Select Method To Add To List** window appears on the screen.

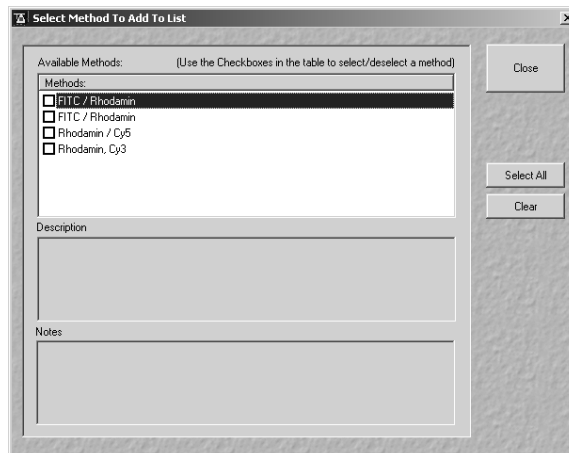


Fig. 7-3 Select Method To Add window

The method taken over from the Expert Mode appears, with the entered name, in the **Methods** panel of the **Select Method To Add To List** window, though without an icon. The method can now be activated as **Standard** or **User-Defined Examination Method**.

- Click on the **Add Method** button.
 - The **Select Method To Add To List** window appears on the screen.
- Use the checkboxes in the **Methods** frame to select / deselect a table.
- Click on the **Select All** button to select all current methods.
- Click on the **Clear** button to deselect all current methods.
- Exit the **Select Method To Add To List** window by clicking on **Close**.
 - The **Routine** window appears on the screen again. The method appears in the list of **User Defined Examination Methods**.
 - An overview image (Preview) for the selected method and the name of the scan mode used are displayed in the **User Defined Examination Methods** panel.

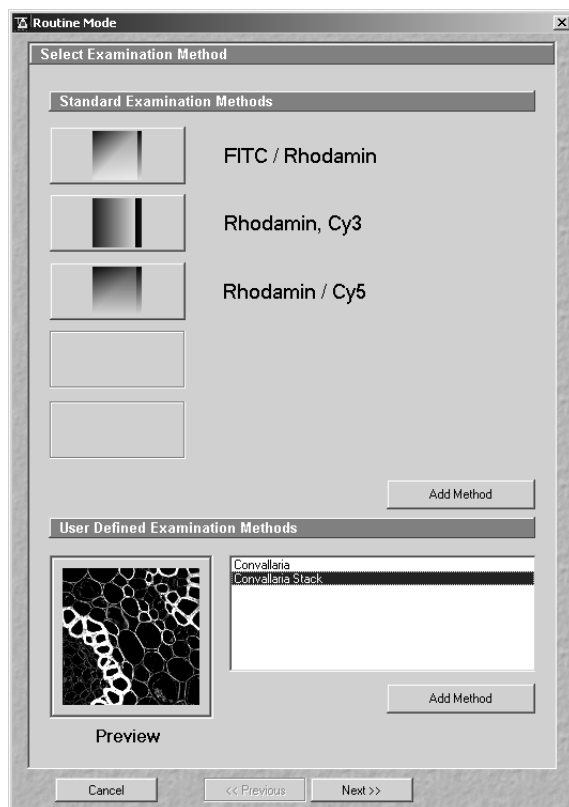


Fig. 7-4 Routine Mode - Select Examination Method window (user defined method activated)

7.1.3 Application of Examination Methods in the Routine Mode

The application of the Routine Mode is menu-controlled. The windows required for the creation of an image are limited to a minimum, and are opened one after the other using the button bar at the bottom of each window.

Lines, frames, stacks, time series and a combination of stacks and time series (4D: XYZT) can be recorded.

Additional windows will be displayed for the creation of stacks and time series. Lambda Stacks cannot be created in the Routine Mode.

The selected scan mode is displayed in all windows (except the Microscope Setup window) for checking purposes. It is possible to change to the next or previous window.

The buttons at the bottom of the relevant windows have the following functions:

Cancel button	Cancels the procedure, the Routine mode is finished.
<< Previous button	Activation of the previous window.
Next >> button	Activation of the next window.
Finish button	Ends the Routine mode; this button is active only in the Image Acquisition window.

7.1.3.1 Acquisition of a Frame

(1) Selection of the examination method

(a) Standard Examination Methods

- Click on the button of the required standard examination method, e.g. **FITC / Rhodamine**.

In case the current system has a META detector in channel 1 one has to define a method first for the routine mode.

- The **Routine Mode - Microscope Setup** window will appear.

(b) User Defined Examination Methods

- Click on the name of the examination method in the selection box of the **User-Defined Examination Methods** panel, e.g.: **Convallaria**.
- Then click on the **Next** button.
 - The **Routine Mode - Microscope Setup** window will appear.

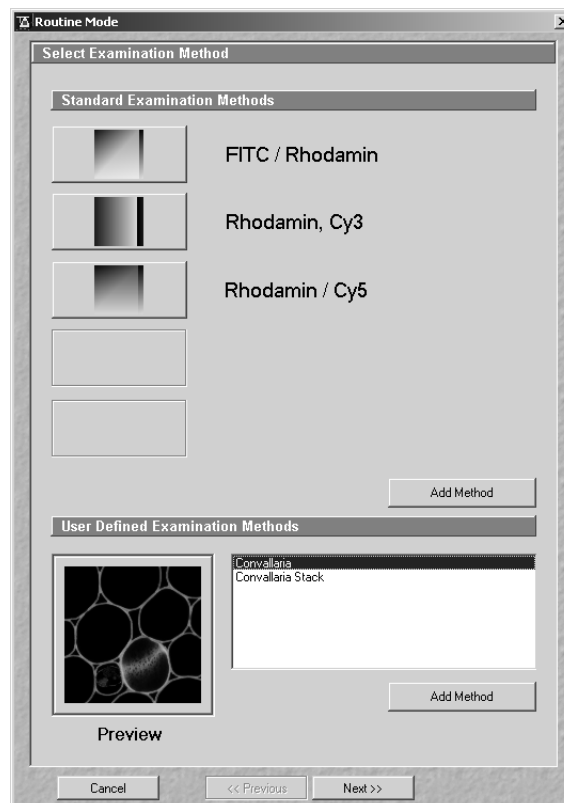


Fig. 7-5 Select Examination Method window

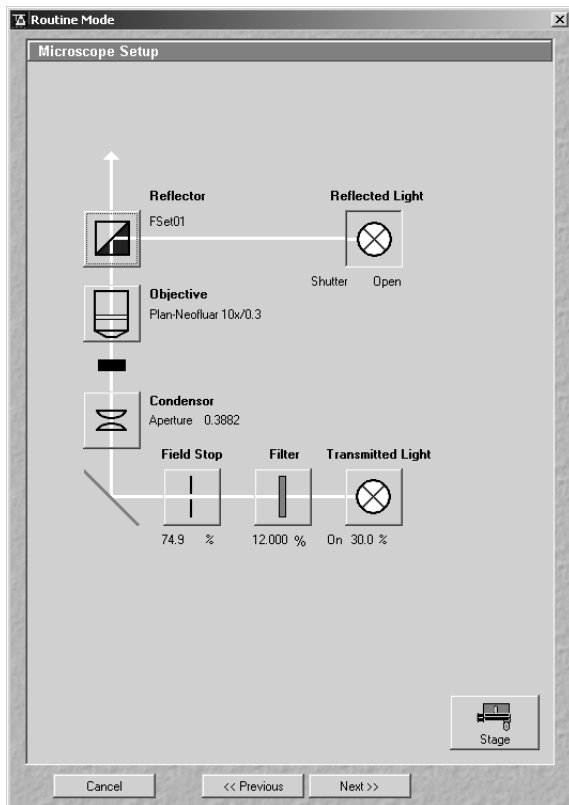


Fig. 7-6 Microscope Setup window

(2) Routine Mode - Microscope Setup window

- Prepare your specimen for examination in the same way as in the Expert mode, (see chapter 4). As in the Expert mode, different windows are available for the Axioplan 2 MOT and the Axiovert 100 M.
- Set the microscope parameters for transmitted light or reflected light by clicking on the appropriate graph (see chapter 4).
- Clicking on the **Stage** button enables you to open the **Stage and Focus Control** window and to perform motorized focusing and specimen positioning.

The **Stage and Focus Control** window is identical to that of the Expert Mode and is handled in the same way (see chapter 4).

The **Stage** button is also available in the **Image Setup**, **Image Stack Setup** and **Time Setup** windows.

- When you have completed your microscope settings, click on the **Close** button to close the **Stage and Focus Control** window.
- Click on the **Next** button.
 - The **Routine Mode - Image Setup** window will appear.

(3) Routine Mode - Image Setup window

When the **Image Setup** window is opened, all the parameters of the selected examination method are set automatically:

- **Image Size**
- **Noise Reduction** corresponds to **Speed**
- **Integration Time** corresponds to **Pixel Time**
- **Data Depth; Scan Direction** (bi-directional setting is taken over, but is not useful and will not be displayed)
- **Zoom, Rotation** and **Offset** are exported with the values **1, 0** and **0**
- **Beam Path** configuration (also Multitracks)
- **Contrast** corresponds to **Detector Gain**
- **Brightness** corresponds to **Ampl. Offset**
- **Optical Slice Thickness**
- **Excitation**
- **Average**

The parameters for Ratio and Bleaching cannot be exported to the Routine Mode.

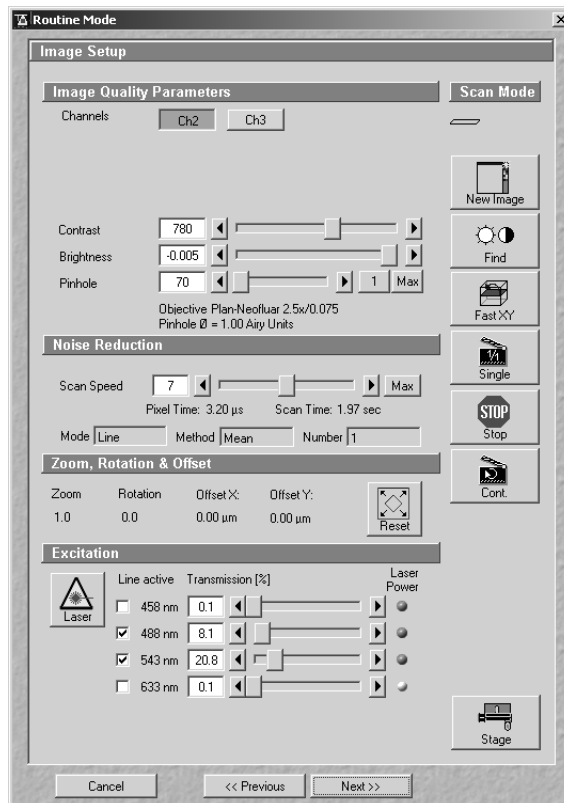


Fig. 7-7 Image Setup window

(a) Image Quality Parameters panel

The setting possibilities of the **Image Quality Parameters** panel permit the image quality to be optimized online (continuous scan procedure).

Optimization is performed track by track (if several tracks are used) and separately for each detector (channel), as in the Expert mode.

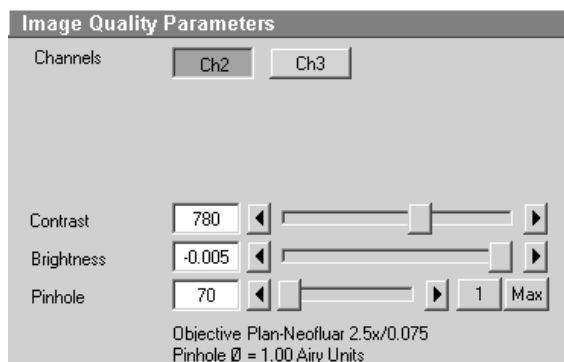


Fig. 7-8 Image Quality Parameters panel

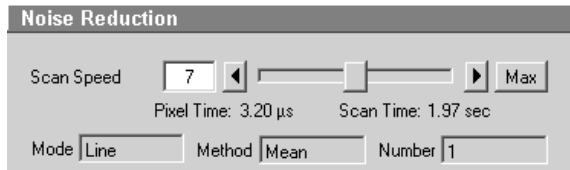


Fig. 7-9 Noise Reduction panel

(b) Noise Reduction panel

In the **Noise Reduction** panel, the scan speed and, depending on it, the scan time can be influenced.

The longer the scan time, the better the noise suppression.

- Use the **Scan Speed** slider to set the slowest acceptable **Scan Time**.

Average Mode, Method and Number are taken over from the Expert Mode and displayed, but they cannot be edited.



Fig. 7-10 Zoom & Orientation panel

(c) Zoom, Orientation & Offset panel

The set zoom, rotation and offset values are displayed in the **Zoom & Rotation** panel.

If the Crop function is used in the Image window, it can be deactivated again using the **Reset** button.

- Click on the **Reset** button to deactivate the **Crop** function.

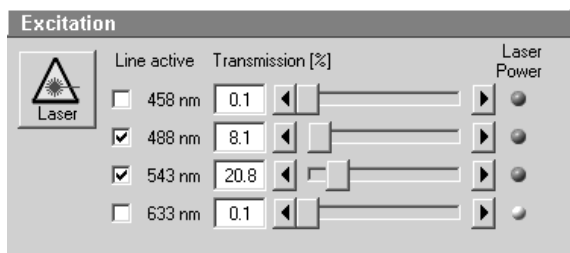


Fig. 7-11 Excitation panel

(d) Excitation panel

In the **Excitation** panel, the wavelength used (**Line Active** check box), the relevant transmission value (**Transmission** slider with input box) and the on / off laser status are displayed.

The laser status display indicates whether the laser has been switched on for the activated wavelength (green - on; gray - off).

If required, further wavelengths can be activated via the **Line Active** check box.

The laser setting is performed in the same way as in the Expert Mode (see chapter 4).

If the laser required for the activated wavelength is not switched on, the relevant message will be provided on the screen in a special window.

- In such a case, open the **Laser Control** window by clicking on the **Laser** button and set the relevant laser to **On**.

(e) Function buttons in the Routine Mode - Image Setup window

The selected scan mode is displayed at the top right of the **Image Setup** window for checking purposes.

The buttons arranged below have the following functions:

New Image button	Opens a new image window
Find button	Triggers single scan with automatic optimization of image brightness and contrast.
Fast XY button	Triggers continuous scan with maximum speed. This function should be used to a limited extent and only for a short period of time.
Single Scan button	Triggers single scan.
Stop Scan button	Stops the current scan procedure.
Contin. Scan button	Triggers continuous scan.
Stage button	Opens the Stage and Focus Control window.

(f) Start scanning

- Click on **Single Scan** or **Contin. Scan** to trigger the scanning procedure.
- If required, optimize the default parameters in the **Excitation, Zoom, Orientation & Offset, Image Quality Parameters** and **Noise Reduction** panels (using continuous scan).
- Click on **Stop Scan**.
- Click on **Next**.
 - The **Routine Mode - Image Save, Print & Export** window appears.

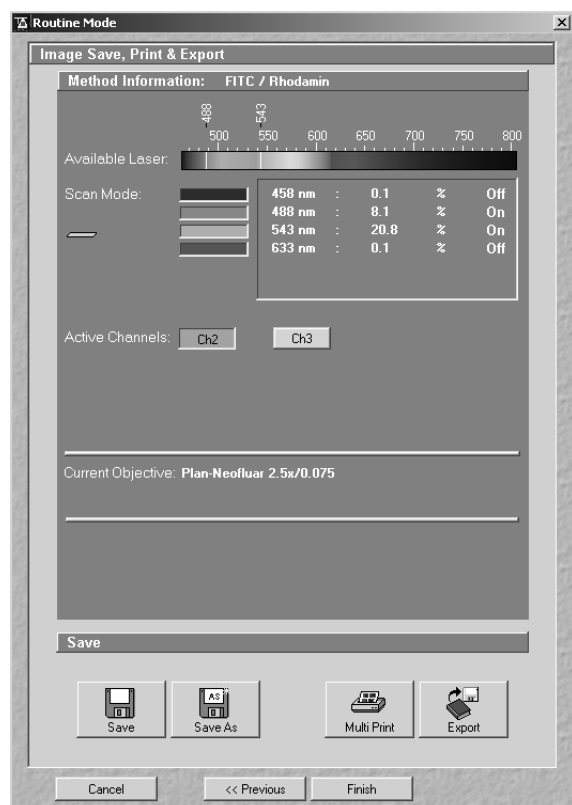


Fig. 7-12 Image Save, Print & Export window

(4) Routine Mode - Image Save, Print & Export window

The **Routine Mode - Image Save, Print & Export** window is used for data backup:

The set parameters are displayed in the **Information For Method: ...** panel.

The buttons **Save**, **Save As**, **Multi Print** and **Export** provided in the **Save** panel permit you to store, print or export scanned images. The procedure is identical to that in the Expert Mode.

- Click on **Save** or **Save As** to store the acquired image.
- Click on **Finish** to exit the Routine Mode and return to the LSM 510 Switchboard menu.

7.1.3.2 Acquisition of a Z Stack

For the acquisition of a Z Stack, the **Routine Mode - Image Stack Setup** window showing the Z parameters is additionally displayed. The parameters correspond to those of the imported Examination Method and, if necessary, have to be matched to the specimen under examination.

- Load the required stack examination method from the **Routine Mode - Select Examination Method** window.
- Open the **Routine Mode - Microscope Setup** and **Routine Mode - Image Setup** windows one after the other by clicking on **Next**. Check the settings and change them, if required (see section 7.1.3.1, page 7-7).
- Click on the **Next** button.
 - The **Routine Mode - Image Stack Setup** window becomes visible.

- You can take over the offered parameters by clicking on the **Start** button.
 - The image stack will be recorded.
- The scan in progress can be stopped at any time by clicking on the **Stop** button.

If Z-settings must be changed, proceed as described in the following.

(1) Routine Mode - Image Stack Setup window

The buttons arranged on the right in the **Image Stack Setup** window have the following functions:

New Image button	Opens a new image window.
Start button	Triggers scanning of a stack.
Stop Scan button	Stops the current scan procedure.
XYscan button	Triggers single XY-scan.
XYcont button	Triggers continuous XY-scan.
Stage button	Opens the Stage and Focus Control window.

The definition of the stack parameters in the **Image Stack Setup** window is made in accordance with the exported Examination Method. If the exported method was created using the HRZ 200 fine focusing stage, the Z-parameters are defined via the **Line Sel / Range** function of the **Hyperfine Z Sectioning** panel.

Otherwise, use the **Mark First/Last** function for the definition of the Z-parameters.

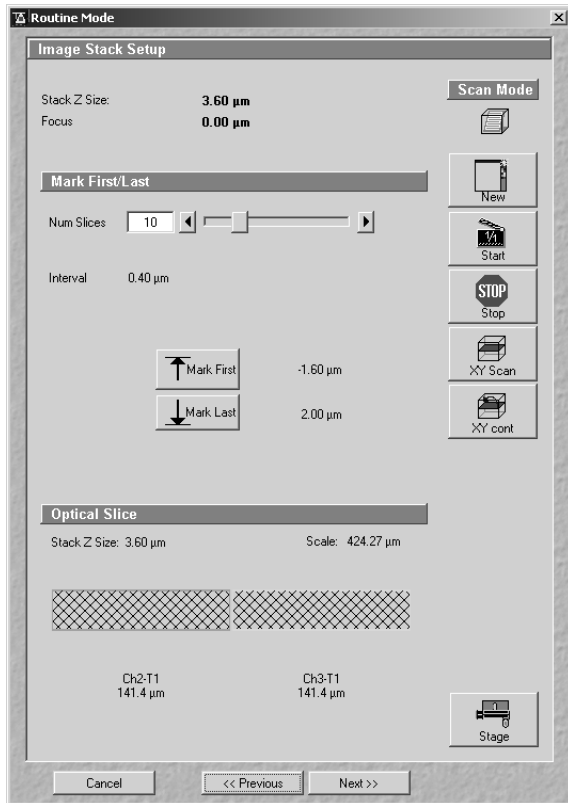


Fig. 7-13 Image Stack Setup window (Mark First/Last)

displays the following information:

Black:

Stack Z Size (μm) = intervals x (number of slices - 1)

Optimal Interval = depending on the objective used and the pinhole diameter setting

Red and other colors:

Presentation of the actual data set by the operator helps to optimize stack creation.

(a) Mark First/Last function

The parameters of the Z Stack to be created are also displayed in the window and can be changed online.

Stack Z Size: Optical indication, i.e. the optical beam moves over this (displayed) Z-thickness (online). The stage (nosepiece) is moved in such a way that the stack size, dependent on the refractive index, is achieved optically.

Focus Position: Mechanical indication, i.e. if the refractive index changes, the value of the focus position also changes in relation to the "0" value (online).

Z Sectioning: Setting of **Number of Slices**, **Interval** and **Current Slice** via sliders / arrow buttons.

Mark First/Last: Determination of the Z-value for the first and last XY-image of the stack, combined with manual focusing or **Stage** control.

Optical Slice: The **Optical Slice** window

For redefining the parameters of a Z Stack, use the setting functions of the **Mark First/Last** panel.

The optimum stack size (definition of the upper and lower limits of the stack) is determined by focus adjustment during a continuous scan.

- Click on the **XYcont** button.
 - A continuous XY-scan of the set focus position will be performed.
- Use the manual focusing drive or the **Stage and Focus Control** window to focus on the upper position of the specimen area where the Z Stack is to start.
- Click on the **Mark First** button to set the upper position of the Z Stack.
- Then focus on the lower specimen area where the recording of the Z Stack is to end.
- Click on the **Mark Last** button to set this lower position.
- The **Num Slices** slider enables you to set the number of slices. The limits of the Z Stack remain constant; the interval is matched accordingly.

The changes made are visualized online in the graphic display of the **Optical Slice** panel.

- Click on the **Start** button to start the recording of the Z Stack.

In case the upper and lower limits of the stack have been switched round, automatic matching will be performed by the software, since the stage of the Axioplan 2 imaging MOT always moves from bottom to top and the nosepiece of the Axiovert 200 M always moves from top to bottom.

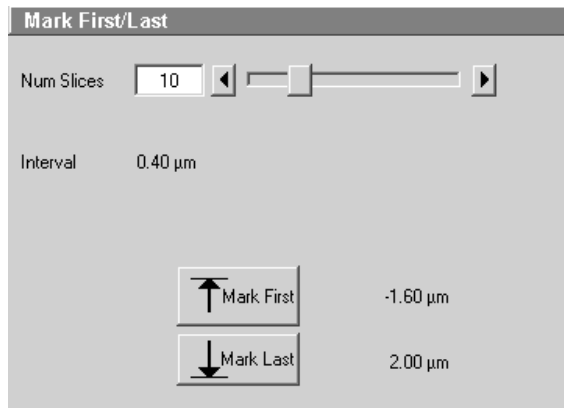


Fig. 7-14 Mark First/Last panel

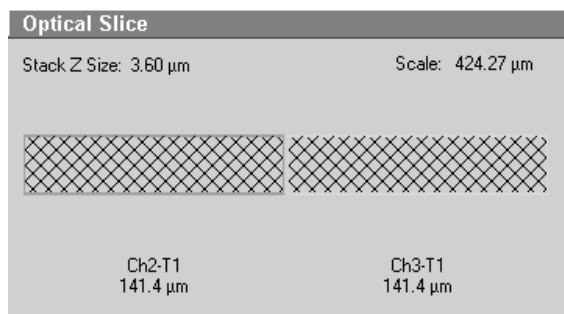


Fig. 7-15 Optical Slice panel

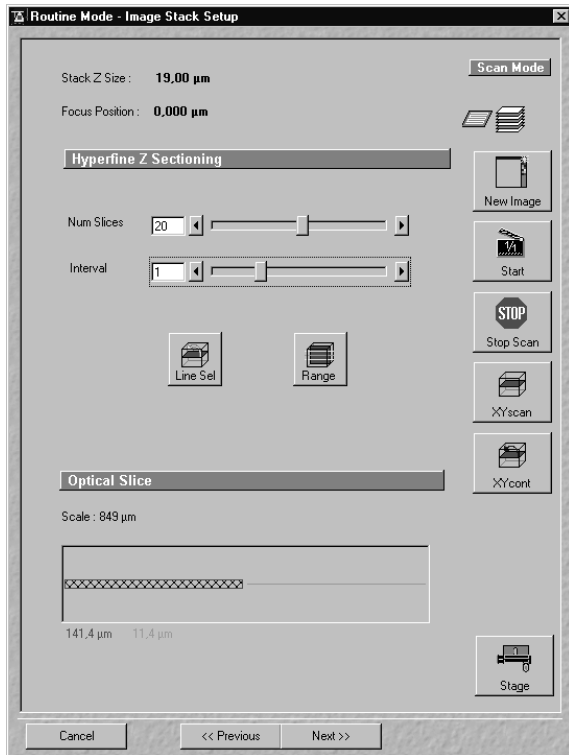


Fig. 7-16 Image Stack Setup window (Line Sel / Range)

For the definition of the stack limits, proceed as follows:

- Click on the **Line Sel** button.
 - An XY-scan of the current slice is performed. The cutline is displayed in the image center. The **Line** toolbar is displayed on the right edge of the Image window.

Use the **Line** toolbar to define the position, shape, width and color of the cutline in the Image window.

(b) Line Sel / Range function with HRZ 200

Activation of this function is only possible if the HRZ 200 fine focusing stage has been connected.

The HRZ 200 can be controlled via software (see **Stage** - Expert Mode, chapter 4).

The accuracy of the HRZ 200 regarding the step width in the Z-direction lies in the range of 10 nm.

The HRZ 200 allows stacks to be produced considerably quicker than via the focus.






The focus position remains unchanged.

The optimum stack size is determined with the help of the **Line Sel** and **Range** functions:

- Use the slider or the arrow keys to set the number of slices for the Z Stack.
- Use the slider or the arrow keys to set the size of the interval.

Num Slices and **Interval** can be varied independently of each other within the HRZ 200 work range of $\pm 100 \mu\text{m}$.

The following function buttons are available in the **Line** toolbar:

Function	Description
Arrow selection button 	Activates the mouse pointer for the selection, positioning and size modification of the cutline in the Image window. Size modification: Click on the drag point and keep the mouse button pressed. Drag the point and release the mouse button. Shifting: Click on the line and keep the mouse button pressed. Shift the complete line and release the mouse button.
Line arrow button 	Generation of a straight cutline in any direction in the Image window.
Opened free shape curve button 	Generation of an open, free shape curve (spline) in the Image window. The first click sets the starting point, each further click adds a line segment. A click with the right mouse button ends the process.
Line button 	Selecting the line width of the cutline.
Color button 	Selecting the color of the cutline.

- Click on the **Line arrow** button or the **Opened free shape curve** button in the **Line** toolbar.
- Define a straight line or a free shape curve (spline) as the cutline for the XZ scan.

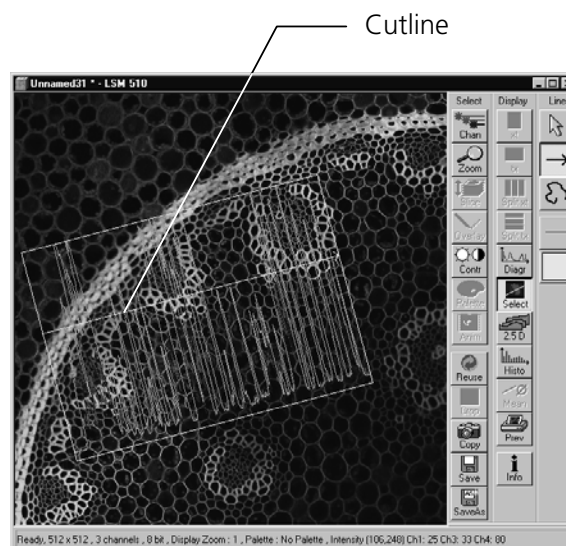


Fig. 7-17 Image window (Line Sel)

- Then click on the **Range** button.
 - The XZ-scan will be performed and displayed in the image window. At the same time, the position of the current slice is shown with a green line and the positions of the first and last slice with two red lines.

Red upper Z range limit Green cutline (current slice)

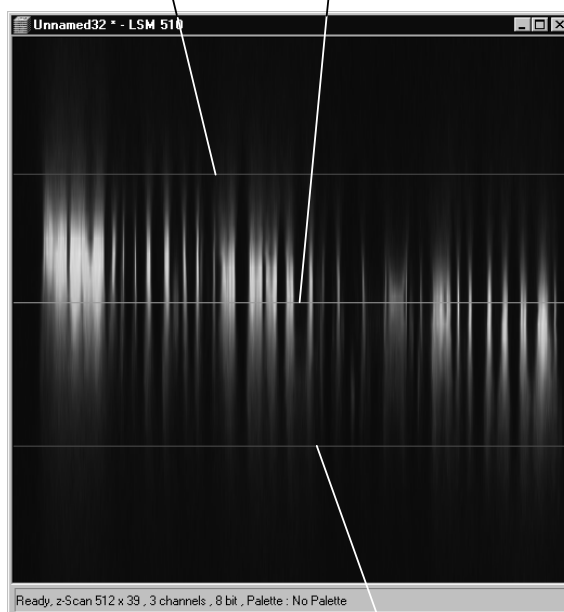



Fig. 7-18 Image window Red lower Z range limit

- Moving the green line (current slice) enables you to change the current focus position (moving the stage or nosepiece in the process). If the green line (Current Slice) is shifted after the creation of **Range**, the focus position will change (the HRZ 200 remains in the center position).
- Shifting one of the red lines enables you to change the stack size. The red lines (stack limits) can only be changed symmetrically to the Current-Slice position within the HRZ 200 work range.

 Since the HRZ 200 moves from bottom to top during the creation of the Z Stack, top and bottom of the Axiovert 100 M have been switched round.

- A click on the **Start** button will start the recording of the Z Stack.
- When recording of the Z Stack is completed, click on **Next**.
 - The **Routine Mode - Image Save, Print & Export** window appears.

(2) Routine Mode - Image Save, Print & Export window

The set parameters are displayed in the **Information For Method: ...** panel.

The buttons **Save**, **Save As**, **Multi Print** and **Export** provided in the **Save** panel permit you to store, print or export scanned images. The procedure is identical to that in the Expert Mode.

- Click on **Save** or **Save As** to store the acquired stack image.
- Click on **Finish** to exit the Routine Mode and return to the LSM 510 Switchboard menu.

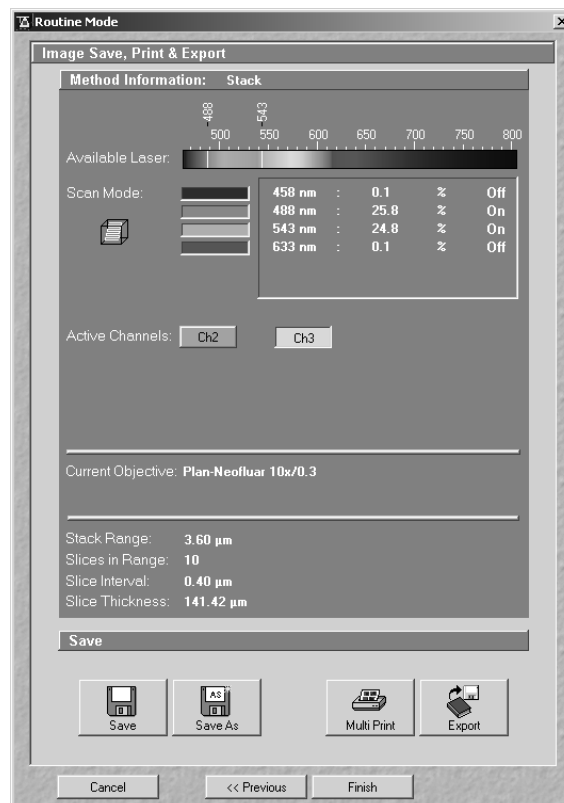


Fig. 7-19 Image Save, Print & Export window

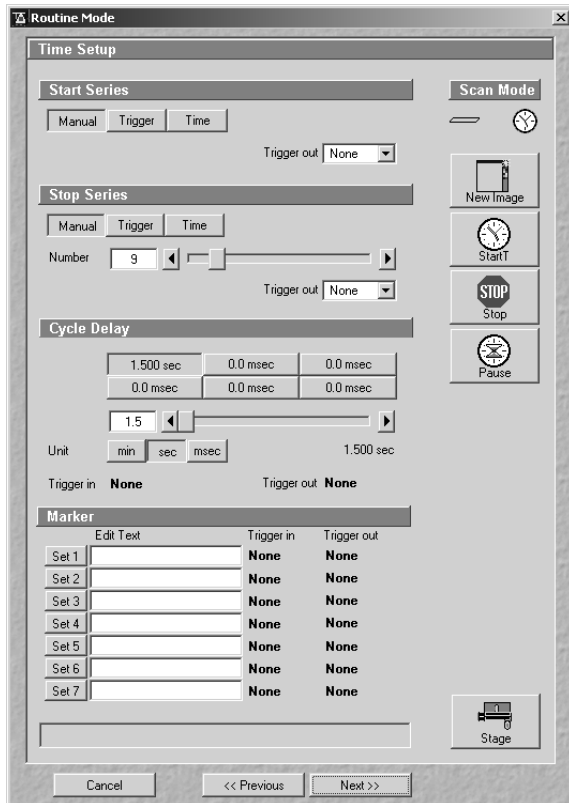


Fig. 7-20 Time Setup window

The following functions are available on the right side of the this window:

- | | |
|-------------------------|---|
| New Image button | Opens a new image window. |
| Start T button | Starts the time series. |
| Stop button | Stops the entire time series. A current scan is canceled. |
| Pause button | Interrupts the time series. The button labeling changes to Resume . A current scan is performed until its end. Pressing the Pause button again will resume the time series. |
| Stage button | Opens the Stage and Focus Control window. |

7.1.3.3 Acquisition of a Time Series

A time series is recorded in the Routine mode in the same way as a frame.

Furthermore, the Time settings are displayed in the **Routine Mode - Time Setup** window. The parameters set here correspond to those of the imported examination method and must be matched, if required.

The time series can be triggered / ended only in the way it was defined in the imported method.

- Load the required time series examination method from the **Routine Mode - Select Examination Method** window.
- Open the **Routine Mode - Microscope Setup** and **Routine Mode - Image Setup** windows one after the other by clicking on **Next**. Check the settings and change them, if required (see section 7.1.3.1, page 7-7).
- Click on the **Next** button.
 - The **Routine Mode - Time Setup** window becomes visible.

(1) Start Series panel

Depending on the imported examination method, the time series can be started and ended manually, via trigger or by entering a time (computer time). In the **Start Series & Stop Series** panel, the relevant mode is displayed. Switching between manual, trigger and time control is not possible in the Routine mode.

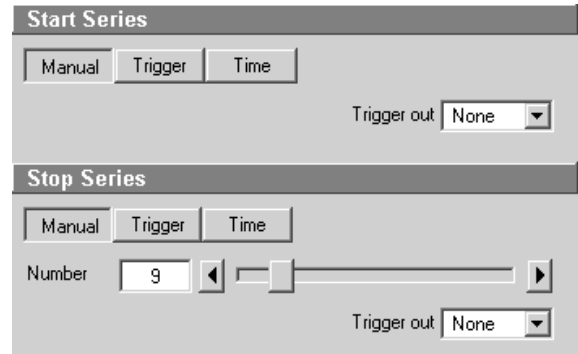


Fig. 7-21 Start & End panel

(a) Manual start of a time series

- You can take over the offered parameters by clicking on the **Start T** button.
 - The time series is recorded and displayed in the image window.
- Click on the **Stop** button to end the time series.

(b) Starting a time series via trigger

- The **Start T** button must be pressed first to start a time series via trigger.
 - **Waiting for Trigger** will be displayed in the status line.
- Then the relevant trigger key of the Trigger Control must be pressed to start the first scan procedure of the time series.
- The time series is ended by pressing the relevant trigger key.

If the number of cycles entered under **Number** has been processed without a trigger impulse having been given to end the procedure, the time series is finished.

If a trigger signal is given before the cycles have been processed, the time series will only be interrupted. **Waiting for Trigger** will be displayed in the status line. The time series can now be continued via a new trigger signal or ended via **Stop**.

(c) Starting a time series via the time

- To start a time series via the time, the **Start** button must be pressed first.
 - **Waiting for Start Time** will be displayed in the status line. The time series is started when the starting time has been reached.
 - The time series is interrupted when the end time has been reached.

If the entered **Number** of cycles has been processed, the time series is finished.

If the number of cycles has not yet been processed, the time series is only interrupted. **Waiting for Start Time** is displayed in the status line. The time series can now be continued by entering a new start time or finished via **Stop**.

The end time for the time series can be changed online.

(d) Input of the number of scan cycles

The currently set number of scan cycles is displayed in the **Number** input box of the **Start & End** panel. This number can be changed, if required.

- Set the required **Number** of scan cycles using the sliders or the arrow buttons.

(2) Cycle Delay / Time Interval panels

Depending on the Examination Method exported from the Expert Mode, the time series interval is defined either as a **Cycle Delay** or **Time Interval**. Accordingly, either the **Time Delay** panel or the **Time Interval** panel is displayed in the **Routine Mode - Time Settings** window.

Time Delay is the interval between the end of one scan process and the beginning of the next.

Time Interval is the interval between the beginning of one scan process and the beginning of the next.

The **Scan Interval** panel permits the intervals to be activated and changed.

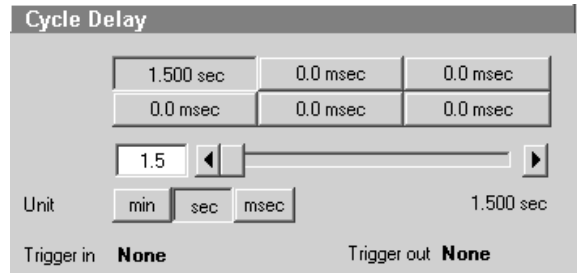


Fig. 7-22 Time Delay panel

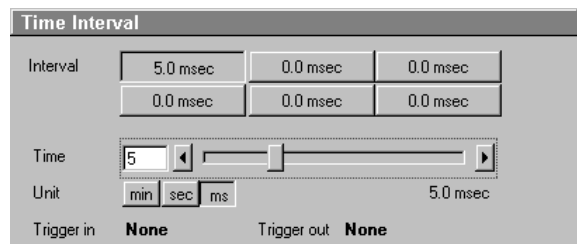


Fig. 7-23 Time Interval panel

The following functions are available:

Interval buttons	Activation of the interval set for the time series for the relevant button.
Time input box / arrow buttons / slider	Determination of the interval for the currently selected time button.
Unit buttons	Selection of the time unit: min , sec or ms .
Trigger in field	Display of the trigger keys (1-4) for the in signal.
Trigger out field	Display of the trigger keys (1-4) for the out signal.


- The interval to be used during the time series is set to a default value by activating a **Time** button.

The time displayed on the time buttons can be changed as follows, if required:

- Activate a **Time** button with a click of the mouse.
- Use the slider (arrow buttons or input box) near **Time** to set the required interval. The set time will be displayed online on the button. Select the required time unit by clicking on the relevant **Unit** button.

You can assign different times to all the six **Time** buttons.

- Changing to another interval is possible by activating another **Time** button during a current time series.

 If the interval change was made via trigger key in the imported examination method, the assigned **Time** button will be activated on pressing the displayed trigger key.

Marker			
	Edit Text	Trigger In	Trigger Out
Set 1	<input type="text"/>	None	None
Set 2	<input type="text"/>	None	None
Set 3	<input type="text"/>	None	None
Set 4	<input type="text"/>	None	None
Set 5	<input type="text"/>	None	None
Set 6	<input type="text"/>	None	None
Set 7	<input type="text"/>	None	None

Fig. 7-24 Marker panel

(3) Marker panel

The setting of a marker permits information about the moment in the current time series and any required comment to be assigned to the current scan. The time indication is set automatically, while comments must be defined before.

The markers (red squares) are visible in the Image window if the **Gallery** button (**Display** toolbar) is activated.

On storage of the image, all the markers, including the time indication and the comments, are stored along with the image contents.

The following functions are available:

- | | |
|----------------------------------|---|
| Set 1-7 button | Setting of a marker during the scan procedure. |
| Edit Text input box (1-7) | Entry of the comments for the marker. |
| Trigger in field (1-7) | Display of trigger keys (1-4) used to set the marker. |
| Trigger out field (1-7) | Display of trigger keys (1-4) for the out signal. |

- Click once in the edit text box of the required marker. Enter the comments via the keyboard. Then click outside the editing box to close this box.
- Clicking on one of the **Set 1** to **7** marker buttons will set a marker for the current scan.

(4) Recording a time series

- Set the relevant parameters for time control in the **Start Series**, **Stop Series**, **Time Delay** and **Marker** panels.
 - Start the time series by clicking on the **Start T** button.
 - If you use Trigger Control, press the relevant trigger key to start the time series with the first scan procedure.
 - Use the **Set 1** to **Set 7** buttons to set markers during the scanning procedure which will allow you to evaluate interesting scanning images later.
 - End the time series by clicking on the **Stop** button.
 - Click on **Next**.
- The **Routine Mode - Image Save, Print & Export** window appears.

 It is also possible to record a Z Stack in the form of a time series (4D Image).

(5) Routine Mode - Image Save, Print & Export window

The set parameters are displayed in the **Information For Method: ...** panel.

The buttons **Save**, **Save As**, **Print** and **Export** provided in the **Save** panel permit you to store, print or export scanned images. The procedure is identical to that in the Expert Mode.

The also available **Save** and **Save As** buttons permit scanned images to be stored in a database (see chapter 4).

- Click on **Finish** to exit the Routine Mode and return to the LSM 510 Switchboard menu.

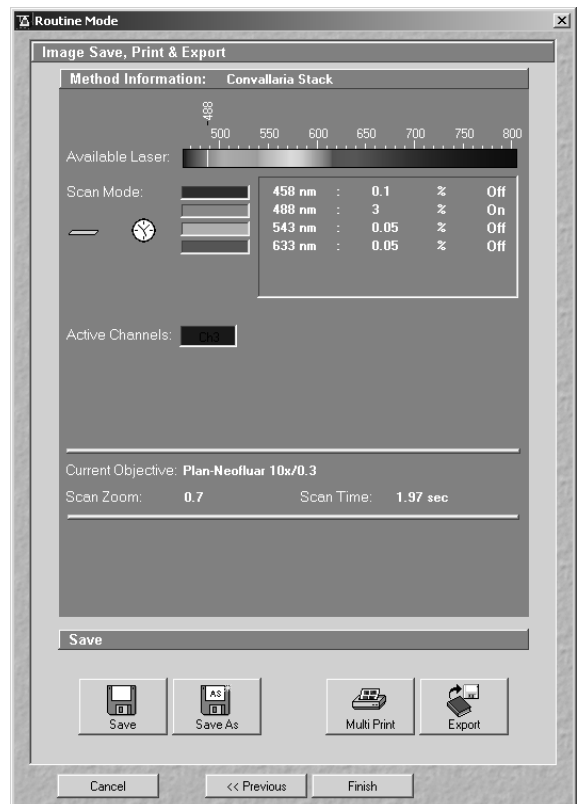


Fig. 7-25 Image Save, Print & Export window

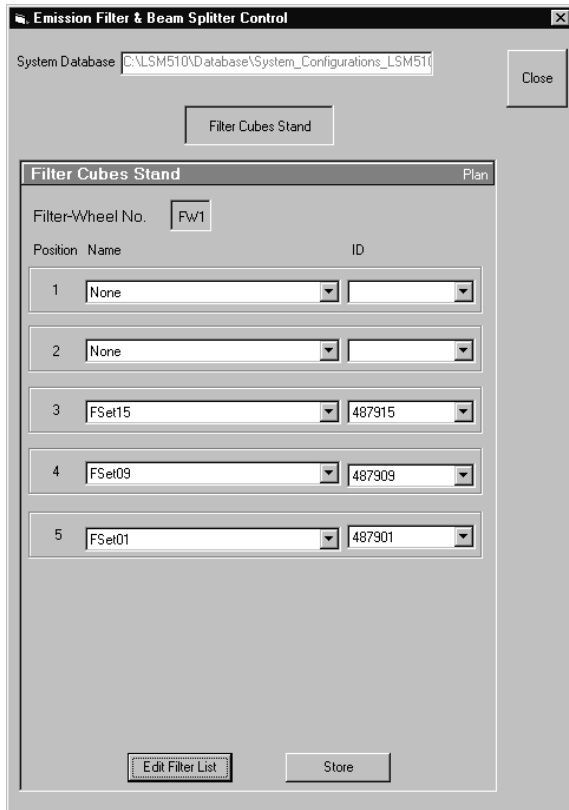


Fig. 7-26 Emission Filters & Beam Splitter Control window

7.2 Tools

7.2.1 Change Filters

The **Change Filters** tool is used to update the filter data in the software after a change of filters in the reflector turret.

- Close the LSM 510 software program.
- Insert the new filter module in the reflector turret.
- Double-click on the **Change Filters** icon on the desktop.
 - The **Emission Filter & Beam Splitter Control** window appears on the screen. The name of the currently used database is displayed in the **System Database** box, with the filter type being indicated below for checking purposes.
 - The **Filter Cubes Stand** panel shows the **Filter-Wheel No.** and the filter positions available.
 - Use the **Name** and **ID** selection boxes to enter the filters installed in the individual positions of the filter wheel.

- Open the **Name** (or **ID**) selection box of the relevant filter position and select the new filter set from the list.
- Click on the **Store** button to accept the new settings.
- Click on the **Close** button to close the **Emission Filter & Beam Splitter Control** window.



All available filter sets have to be registered in the filter list (see **Edit Filter List** function, next page).

Edit Filter List

The **Edit Filter List** function permits updating of the filter data in the software after a change of filters on the stand.

- Close the LSM 510 software program.
- Double-click on the **Change Filters** icon on the desktop.
- Click on the **Edit Filter List** button in the **Emission Filter & Beam Splitter Control** window.
 - The **Edit Filter/Beam Splitter List** window is opened.

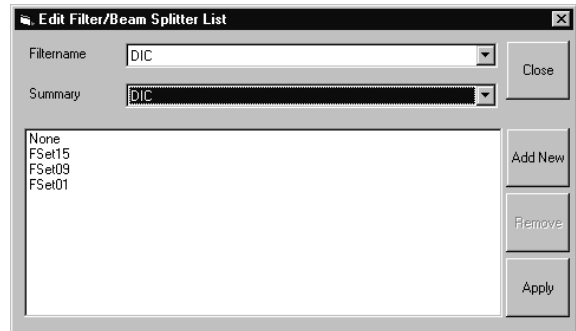


Fig. 7-27 Edit Filter/Beam Splitter List window

This window permits a list of the most frequently used filter sets to be compiled.

- Click on the arrow button in the **Filtername** list box to open it.
- Select the filter set which shall be included in the list.
- Click on the **Apply** button.

The selected filter set is included and displayed in the list (below the **Summary** list box).

This filter set is now also available in the **Name** selection boxes of the **Filter Cubes Stand** panel and can be assigned to a filter wheel position.

To remove a filter set which is no longer needed from the list, proceed as follows:

- Click on the name of the filter set concerned in the list box of the **Edit Filter/Beam Splitter List** window.
- Click on the **Remove** button. The filter set is deleted from the list and is then no longer available in the **Filter Cubes Stand** panel of the **Emission Filter & Beam Splitter Control** window.

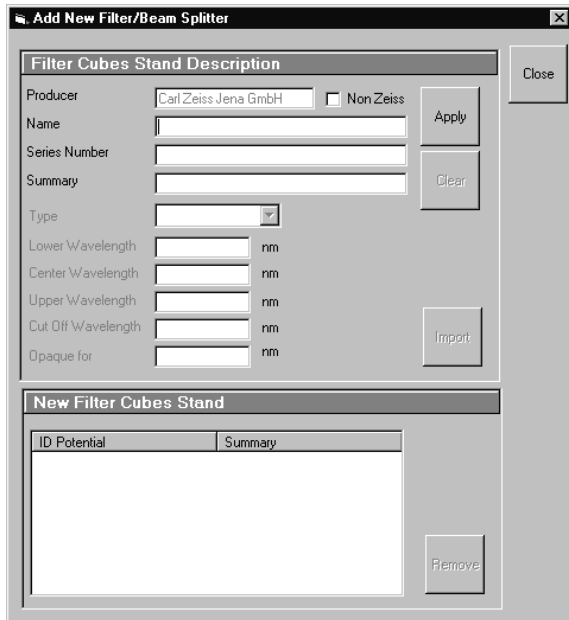


Fig. 7-28 Edit Filter/Beam Splitter List window

Add New

This function permits new filter sets to be added to the database.

For this, proceed as follows:

- Click on the **Add New** button on the **Edit Filter/Beam Splitter List** window.
 - The **Add New Filter/Beam Splitter** window is opened.
- Enter the data of the new filter set in the **Filter Cubes Stand Description** panel, then click on the **Apply** button.

The new filter set is stored in the database and included in the **New Filter Cubes Stand** panel. You can now activate the filter for a filter wheel position using the procedure described above.



If you have activated the **Non Zeiss** check box, filter sets from other manufacturers can also be included in the database.

- To remove an new filter set from the database, select it with a click of the mouse in the **New Filter Cubes Stand** panel and then click on **Remove**.
- Click on **Close** to close the **Add New Filter/Beam Splitter** window.
- Click on **Close** to close the **Edit Filter/Beam Splitter** List window.
- Click on the **Store** button to accept the new settings.
- Click on the **Close** button to close the **Emission Filter & Beam Splitter Control** window.

When you start the LSM 510 software, the filter data are updated.

7.2.2 Stand Select

The **Stand Select** tool permits a new or updated database to be assigned to the LSM 510 software program. This function should preferably be performed by authorized service personnel.

If this is not possible, proceed as follows:

- Close the LSM 510 software program and double-click on the **Stand Select** icon on the desktop.
 - The **Select Stand Database** window appears on the screen. The currently used database is displayed in the **Database** box.
- Click on the **Browse** button to activate the new database.
 - The **Open** window appears on the screen.
- Select the directory where the new database is stored.
- Click on the name of the database (file extension: ***.mdb**) and then on the **Open** button.
 - The **Open** window is closed and the name of the new database appears in the **Database** box.
- Click on the **Permanent** button. The **Select Name** window appears.
- Select the relevant stand icon from the **Icon** list box and click on **OK**. The **Select Name** window is closed and the desktop icon is updated.
- Then click on the **OK** button in the **Select Stand Database ...** window to accept the new settings and to close the window. (Clicking on **Cancel** will cancel the procedure.)
 - After the next restart of the LSM 510 software program, the new database will be automatically read in.

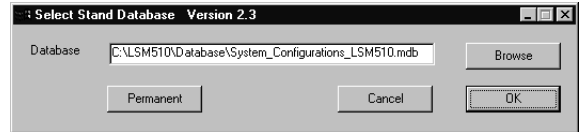


Fig. 7-29 Select Stand Database ... window

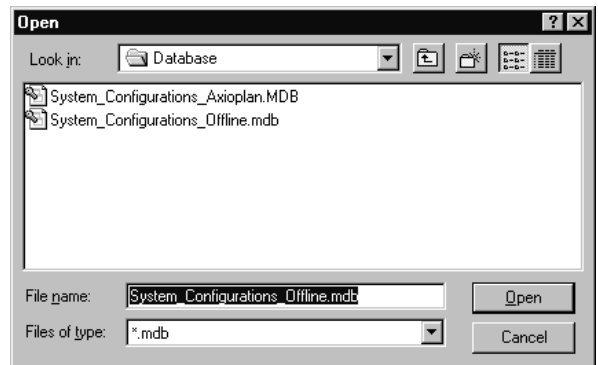


Fig. 7-30 Open window

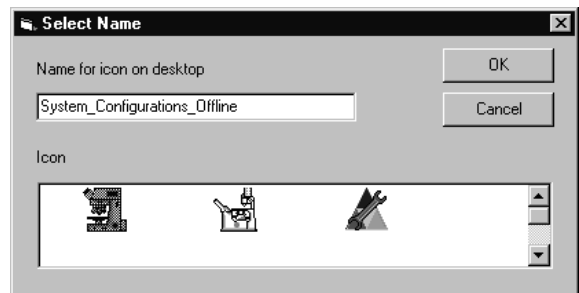


Fig. 7-31 Select Name window

7.2.3 LSM Image Browser

The **LSM Image Browser** permits images to be loaded, imported, exported and printed quickly without having to open the LSM 510 software. The **LSM Image Browser** can be used without dongle.

When images are opened, image processing functions of the LSM 510 software are available to a limited extent (**Chan, Zoom, Contr, Palette, Copy, Save, Save As, xy, Split xy, Prev, Info**).

- Click on the **LSM Image Browser** icon on the desktop of the PC. The **Zeiss LSM Image Browser** main menu is opened.



Fig. 7-32 Zeiss LSM Image Browser main menu

The following function buttons are available:

New button	Opens a new database.
Open button	Opens an existing database.
Save button	Saves the current image.
Save As button	Saves the current image under a new name.
Import button	Imports images.
Export button	Exports images.
Full Screen button	The current image is displayed on the full screen. Deactivation of the function with a click of the mouse.
Multi Print button	Several images are printed on one page.
RAM button	Use of the RAM memory for image display.
DISK button	Use of the hard disk as storage medium for image display.
Exit button	The Zeiss LSM Image Browser main menu is closed.

The functions **New, Open, Save, Save As, Import, Export** and **Multi Print** correspond to those of the Expert Mode of the LSM 510 software and have already been described in chapter 4.

7.2.4 LSM Image Examiner

The **LSM Image Examiner** can be used without having to open the LSM 510 software. However, this requires the installation of the relevant dongle. The **LSM Image Examiner** provides all the functions of the **LSM Image Browser**, plus the 3D functions and selected **Process** functions of the Expert Mode of the LSM 510.

When images are opened, a large scope of the image processing functions of the LSM 510 software is available (for further details see in chapter 4).

- Click on the **LSM Image Examiner** icon on the desktop of the PC. The **Zeiss LSM Image Examiner** main menu is opened.



Fig. 7-33 Zeiss LSM Image Examiner main menu

In addition to the buttons of the **LSM Image Browser** mentioned above, the following function buttons are available in the lower row of the **Zeiss LSM Image Examiner** main menu:

- | | |
|---------------------------|--|
| Projection button | One single projection or a series of projections can be calculated after rotation of the data package about the X, Y or Z axis. |
| DepthCod button | The depth information contained in a sequence can be colored with the colors of the rainbow. |
| Stereo button | Stereoscopic images can be generated. |
| Ratio button | Permits two channels to be linked into a new channel by the creation of a ratio. |
| Copy button | Permits one channel each of an existing image to be copied and stored as a new image. |
| Filter button | Permits the subsequent processing of scanned images via the integrated filters. |
| Interpolate button | Permits the continuous contrast and brightness change in a stack or time series through interpolation between the starting and end values. |
| Shift button | Produces a congruent image with relation to the pixels of the various channels. |

Duplicate button	Permits images (including Z Stacks and Time Series) to be duplicated completely.
Contrast button	Permits the subsequent modification of contrast and brightness of the stored image.

These functions correspond to those of the Expert Mode of the LSM 510 software and have already been described in chapter 5.