

User's Guide

# PMOD Image Segmentation Tool (PSEG)

Version 3.9



PMOD Technologies

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# PMOD Image Segmentation Tool Introduction (PSEG)

Preclinical imaging systems allow following the uptake and metabolism of labeled compounds in whole mice and major parts of rats. To exploit this wealth of information with regard to pharmacokinetics, it is necessary to localize all relevant tissue structures and assess their tracer concentration in time, which is then analyzed with kinetic models.

Manual segmentation is a tedious process and prone to considerable subjective variability. It is challenging for studies involving many animals, has a direct impact on the accuracy of the study endpoint and consequently on the required sample size.

The PSEG tool supports the semi-automatic segmentation of dynamic rodent PET studies within only a few minutes. It applies a patented clustering approach for grouping neighboring image pixels with similar kinetic behaviors. This process creates a hierarchy of compact tissue segments covering the whole body. Segments on low hierarchy levels are small and highly homogeneous regarding kinetics, whereas they grow towards higher levels and become kinetically more variable. The user can interactively browse through the hierarchy levels and assign an organ label when an appropriate representation has been found. If tissue structures cannot directly be represented by a segment or a union of segments, the user can interactively outline them in an anatomical reference image.

Once all relevant tissue structures have been segmented, PSEG readily calculates their average PET uptake curves (also time-activity curves, TAC) and optionally corrects them for the partial-volume effect.

## Methodology

### ORGAN SEPARATION by Local Means Analysis (LMA)

PSEG implements the local means analysis (LMA) method licensed from CEA, Orsay, France. This method aims at segmenting "functional organs" characterized by a particular pharmacokinetics. A functional organ only corresponds to an entire anatomical organ if the organ is functionally homogeneous. Otherwise, the functionality of different organ parts are separated and can be treated individually. The patented LMA method has been shown to be robust regarding the low signal-to-noise ratio, the limited spatial resolution and potential organ movement of dynamic rodent PET studies [1,2,3].

The LMA segmentation includes the following processing steps:

- ▶ Exclusion of the noisy background from segmentation.
- ▶ Automated localization of pixels in the organ centers.
- ▶ Computation of the local pharmacokinetics and global noise parameters.
- ▶ Parcellation of the body volume in the image into a predefined number of regions.
- ▶ Hierarchical fusion of the regions to form increasingly larger, connected regions. On each level of the hierarchy, the whole body is covered by disjoint regions.

Tissue structures which cannot be isolated by the LMA segmentation because they do not kinetically differ from the neighborhood need to be outlined using the general VOI features of PMOD.

### k-Means Clustering

In addition to the LMA method which incorporates prior knowledge, PSEG offers a k-means clustering as a general method for subdividing a volume into clusters of "similar" pixels [5]. The time-weighted Euclidean distance is used as the measure of dissimilarity (or distance) between TACs. In PSEG, the procedure performs the following steps for the pixels within a mask, given a prescribed number N of clusters

- 1) N non-background pixels serving as initial cluster centroids are randomly assigned.
- 2) Each pixel is assigned to that centroid with minimal distance between the TACs, thus forming N initial clusters.
- 3) For each cluster a new centroid TAC is calculated as the average TAC of all pixels in the cluster.
- 4) An iterative process with a maximal number of iterations is started which repeats the following two steps:
  - (1) Each pixel TAC is compared with all centroid TACs and assigned to the cluster with minimal distance.
  - (2) All centroid TACs are recalculated to reflect the updated cluster population.
 The iterations are repeated until no pixels are re-assigned to a different cluster, or the maximal number of iterations is exhausted.

Note that because no geometric information is used for the clustering, spatially disconnected pixels will most likely be included in the resulting clusters.

### Supervised Clustering by a Set of Kinetic Class TACs

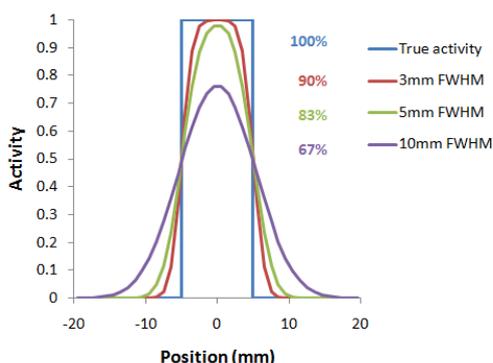
k-means clustering is working completely data-driven without any prior knowledge apart from the number of clusters. PSEG offers a complementary supervised clustering approach where the user specifies a set of TACs which are characteristic for the kinetics of different tissues. This method was introduced by Turkheimer et al. [6] for determination of gray matter reference tissue without specific binding in [11C]PK11195 brain studies. It requires preparation of time-activity curves which are representative for different tissues in the brain. They found that the method worked best after normalizing the data frame-wise by subtracting the frame average and dividing by the frame standard deviation.

The implementation in PSEG is close to the supervised clustering variant presented by Ikoma et al. [7] for use with [11C]PIB. Particularly, the weight ratio (eq. 3 in [7]) is used to assign each pixel to one of the classes, or to background.

Note that the data has to be normalized outside of PSEG using the z-score external tool.

### Partial-Volume Effect

PET images are inherently affected by the partial-volume effect. This means that the measured tracer activity concentration is not accurate due to the relatively low image resolution and the limited tissue sampling. The low spatial resolution of the PET system causes a blurring of the image, so that high activities (from a hot lesion) are spread to the surrounding as illustrated below. This effect is called spill-out. The same effect also causes a spill-in of background activity into the volume of interest.



As a consequence, hot lesions tend to appear less aggressive (reduced maximum) but bigger (spreading) than they are in reality.

Partial-volume effects are complex: Spill-in and spill-out depend on the geometry of the objects, the activity distribution of the tracer, and on the resolution of the scanner which may vary across the imaging field-of-view. Therefore, practical correction approaches have to assume certain conditions and can only be approximate.

### VOI Based Partial-Volume Correction (GTM Method)

The Geometric Transfer Matrix (GTM) method according to Rousset et al. [4] restricts partial volume correction to the signal of the true objects which are constituted by VOIs. The relation of measured PET values (affected by the partial-volume effect) to the true PET values is given by the matrix equation below

$$\vec{C}_{measured} = [GTM] \times \vec{C}_{true}$$

with the following notations:

- $C_{true}$  Vector of the true average activity concentration in the different VOIs of interest. The vector length  $n$  equals the number of object VOIs.
- $C_{measured}$  Actually measured average activity concentration in the different VOIs. Each VOI is assumed to have a homogeneous concentration.
- GTM Geometric Transfer Matrix which describes the spill-over among all the VOIs. The matrix is square with  $n \times n$  weighting elements  $w_{ij}$  which express the fraction of true activity spilled over from  $VOI_i$  into  $VOI_j$ .

In practice,  $w_{ij}$  is calculated as follows: A binary map is created with 1 in all pixels of  $VOI_i$  and 0 elsewhere. The map is convolved with the imaging Point-Spread Function (PSF), and in the resulting spillover map the weighted average of all  $VOI_j$  pixels calculated.

The GTM equation above represents a system of linear equations. Once the weights have been calculated, the system can be solved for the true values  $C_{true}$  by matrix inversion. It has been shown [4] that this algorithm is robust to noise propagation during the correction process.

### LMA Variant of GTM Method

The LMA (Local Means Analysis) GTM method [1] uses the homogeneous regions localized by the segmentation and calculates the average uptake in the inner of the structures. The percentage of pixels per segment considered for averaging is a parameter of the method. With 100% pixels included, the LMA GTM method equals the standard GTM method.

# User Interface

## Starting the PSEG Tool

The PSEG tool is started with the **Segment** button



from the PMOD ToolBox or by directly dragging image files onto the above button.

## Organization of the PSEG User Interface

The user interface of PSEG consists of three pages which can be selected by tabs:

- 1) **DB Load** page: Loading of the images from a PMOD database. The images are transferred to the processing page according to their modality. This page is not shown if the database functionality is disabled.
- 2) **Segment** page: Main segmentation page for creating VOIs by automatic and manual methods. They can be applied to the dynamic PET series for calculating the regional uptake, optionally with a partial volume correction.
- 3) **Statistics** page: Shows the resulting tissue time-activity curves. They can be saved or submitted to the kinetic modeling tool.

## Step-Wise Processing

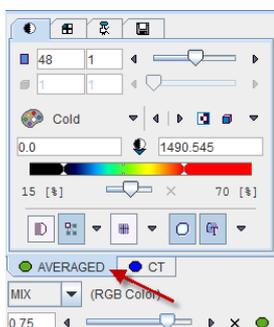
Data processing is consistently organized by a stepwise progression towards the end result. On each processing stage the user has to take some action such as data loading or parameter configuration, and then start the next processing step with the red action button located in the lower right. As soon as the result is calculated, it will be shown on a new page representing the new processing stage. The cascade of stages is available by the selection area in the upper right.



Selecting the processing pages in this way conveniently allows inspecting the results of prior stages without initiating any calculations. To repeat a calculation with modified parameters, please activate the action button in the lower right again.

## Fusion Image Display

The content of the image display area is controlled in the upper right. Often, more than one image contribute to the display. In these cases the tab corresponding to an image has to be first activated, before its color table or the color thresholds can be modified.



The fusion control section is located below the image control tabs. In the illustration above the colors of both images are mixed, whereby the weighting can be changed with the slider.

## Convenience Buttons

Next to the action button in the lower right is an area with three buttons

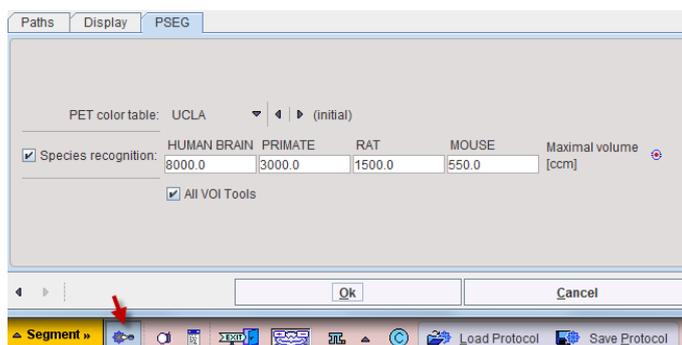


offering the following functions:

-  Hide the parameters panel to free some space in the user interface. With the panel hidden, the icon changes to . When this button is activated, the panel is shown again.
-  Reset the parameters on the page to their default values. If the button in the taskbar to the right is activated, the defaults are reset on all pages.
-  Display help information for the current page.

## Configurations

The PSEG tool can be configured according to user preferences in a dialog window as illustrated below.



The common configurations are available on the **Paths** and **Display** tabs, and in the upper part of the **PSEG** tab. The **All VOI Tools** flag allows defining the functionality level of the **VOIS** page. Default is a reduced set of VOI tools. To have all possible VOI tools available please enable the **All VOI Tools** box and restart.

### Further Information

This guide is focused on the dedicated rodent segmentation functionality. Please refer to the *PMOD Base Functionality Guide* for details about general functions such as data loading, image display, and VOI definition.

## Recommendations

### Motion Correction

Although the LMA segmentation algorithm is robust with respect to motion, movement during the dynamic acquisition should be avoided. While internal organ movement and bladder filling are difficult to handle, global relocation of the animal should be corrected before the segmentation process. PMOD's fusion tool includes a motion correction facility which has been successfully applied to preclinical data.

### Anatomical Reference Image

For the interpretation of the segments it is helpful to project them onto an anatomical image of the animal. Therefore, a PET scan is often complemented with a CT or MRI scan, preferably using the same support system and without moving the animal in between. If an offset of the reconstructed images occurs, it can easily be corrected by a rigid transformation.

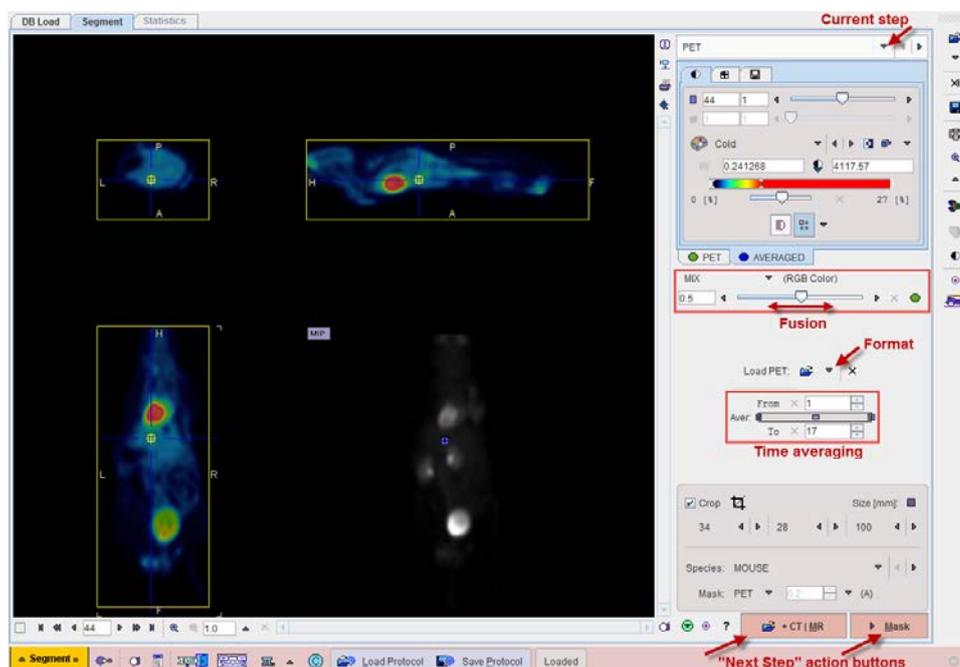
Multi-modal data needs to be matched before using them in PSEG. In order to conserve the better resolution of the anatomical modalities, it is recommended to match the PET series to the MR or CT series, for instance with PMOD's fusion tool.

## PSEG Workflow

PSEG guides the user step by step through the segmentation of a study, each step represented by a dedicated page.

## PET Image Loading and Time Averaging

Step-wise processing is started by selecting the **Segment** tab.



### PET Image Loading

The **Load PET** button for loading the PET images is located in the right control area. It behaves as an option button which needs to be set to the appropriate data format with the indicated arrow. For loading images which are not saved in a PMOD database it is recommended to apply the **Autodetect** format which will figure out the appropriate image loader.

PSEG assumes that the loaded PET series is dynamic. It generates a new series by averaging a range of frames and assigns it to the **AVGERAGED** tab. The averaging range can be defined by the **From** and **To** number fields, or dragging the range indicators in the **Aver** bar. After any modification of the range, the average is recalculated and the display updated. The aim of the averaging is to generate an image which is useful as an anatomical reference for the segment interpretation, as well as for creating the body mask. The original and the averaged images are shown in a fusion rendering which can be adjusted in the fusion control area below the tabs for selecting the individual images.

### PET Image Cropping

If the PET field-of-view is much larger than the body of the animal, the data set should be reduced in order to save RAM and optimize processing. To do so, please enable the **Crop** box and place the center of the yellow crop volume indication by clicking into the image. The edge size in [mm] of the box can be adjusted for each direction by selecting the size in the corresponding list, or by selecting the **Size** button and entering the three edge lengths. The  button initiates cropping, whereby the original data are replaced. If cropping is not activated manually, a request will be shown when proceeding to the next step. Note: The cropping operation is only allowed once.

## Body Mask Definition

The segmentation will operate on all pixels contained in a mask which is usually derived from the data. Please set the **Species** selection according to the studied subject. It establishes proper defaults for the morphological operations which are later applied to the data. The **Mask** selection allows switching between three methods of mask derivation: **PET**, **CT** and **MR**.

### PET-Based Body Mask

With **PET** masking the body mask is derived from the averaged PET series. There are five masking approaches available:

- ▶▶ **Otsu**: This method iteratively looks for the best threshold to separate an object from a background assuming two main gray-level regions.
- ▶▶ **Mean**: Includes all pixels above the image mean.
- ▶▶ **% max Value**: Includes all pixels above the specified percentage of the image maximum.
- ▶▶ **Threshold Value**: Includes all pixels above the specified absolute value.
- ▶▶ **Automatic**: This procedure calculates the image log-values and derives a threshold from the histogram.

In addition to the procedure selected above, morphological opening/closing operations will be applied to remove isolated pixels and fill holes.

Please select a definition appropriate for your PET series. If there are no matched MR or CT images, proceed with the **Mask** action button, otherwise proceed to the additional image loading with the **+CT | MR** button.

### MR- or CT-Based Body Mask

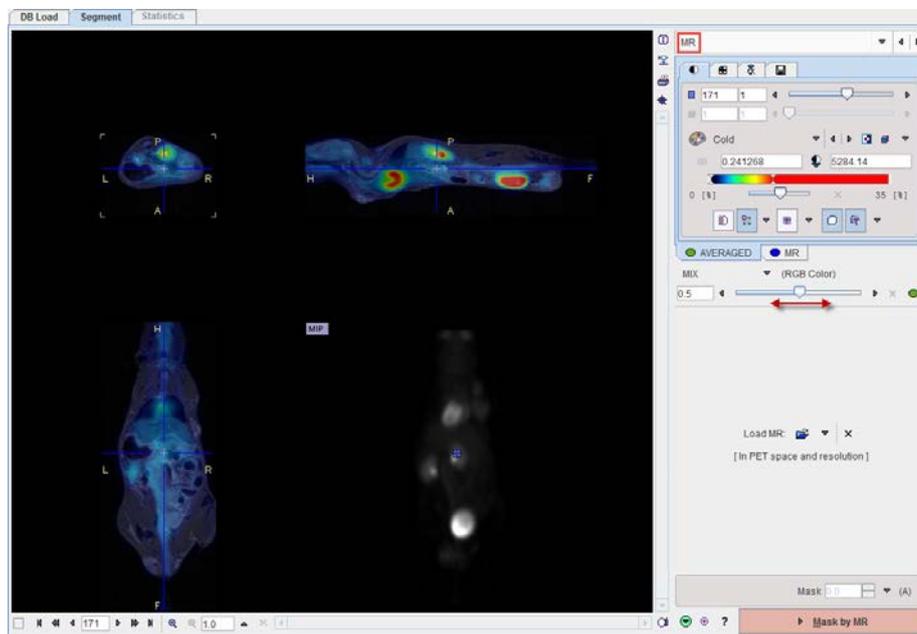
If matched CT or MR series of the same animal are available, they can not only be used as reference images for the segmentation assignment, but also for creation of the body mask. In order to apply MR- or CT-based masking please set the **Mask** choice accordingly. The change is reflected in the parameter area as illustrated below.



The PET masking options are no longer available, and the action buttons are replaced by a single button for progressing to the corresponding image loading page.

## MR or CT Reference Image Loading

The layout of the second page depends on the **Mask** setting on the **PET** page. The situation illustrated below represents the selection of **MR**-based masking. The page is titled **MR**, and in the lower right the **Mask** options appear with a corresponding action button **Mask by MR**. The situation with CT-based masking is analogous.



With PET-based masking the page is labeled **CT | MR**, the image loading button **Load Anatomical**, and the action button **Mask by PET**.

### Reference Image Loading

Please use the indicated button for loading an anatomical image series which is matched in space and resolution to the PET series. Appropriate images can easily be created using PMOD's image fusion tool. Note that the loaded images are automatically cropped by the same box as the PET. The correspondence can be verified with the fusion slider.

### MR-based Body Mask Definition

In the case of an MR-based mask selection the corresponding **Mask** options are available in the lower right:

- (O) - Otsu
- (M) - Mean
- (V) - % max Value
- (T) - threshold Value
- (A) - Automatic

- ▶▶ **Otsu:** This method iteratively looks for the best threshold to separate an object from a background assuming two main gray-level regions.
- ▶▶ **Mean:** Includes all pixels above the image mean.
- ▶▶ **% max Value:** Includes all pixels above the specified percentage of the image maximum.
- ▶▶ **Threshold Value:** Includes all pixels above the specified absolute value.
- ▶▶ **Automatic:** This procedure calculates the image log-values and derives a threshold from the histogram.

### CT-based Body Mask Definition

In the case of a CT-based mask selection the corresponding **Mask** options are available in the lower right:

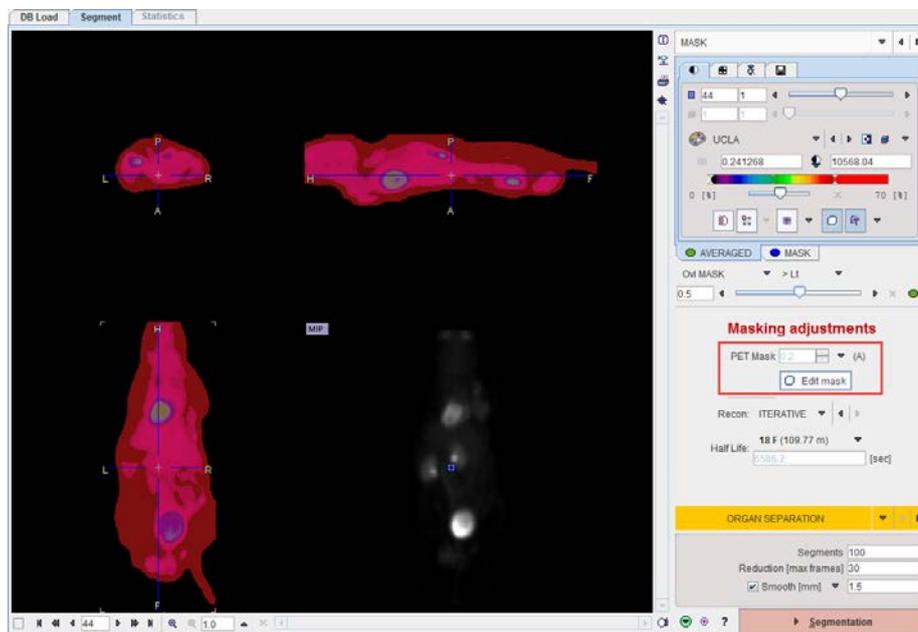
- ▶▶ **Otsu:** This method iteratively looks for the best threshold to separate an object from a background assuming two main gray-level regions.
- ▶▶ **Mean:** Includes all pixels above the image mean.
- ▶▶ **% max Value:** Includes all pixels above the specified percentage of the image maximum.
- ▶▶ **Threshold Value:** Includes all pixels above the specified absolute value.
- ▶▶ **Automatic:** This method applies a threshold, followed by morphological operations for removal of the bed, closing of the nostrils and filling of the lungs.

In addition to the procedure selected above, morphological opening/closing operations will be applied to remove isolated pixels and fill holes.

Please proceed with mask creation by activating the action button in the lower right.

## Body Mask Creation and Segmentation Configuration

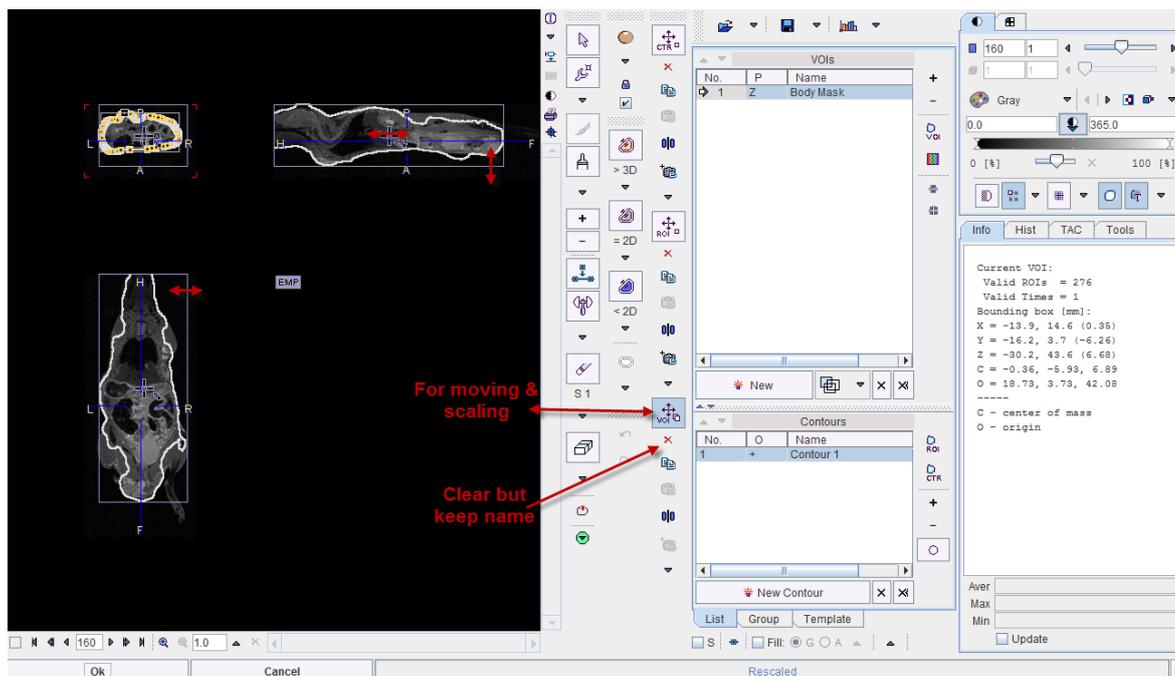
When arriving at the **MASK** page, the display shows a fusion of the **AVERAGED** PET with the generated **MASK**.



### Mask Adjustments

If needed the mask can be adjusted in the area indicated above. The **Mask** choices depend on the image which was defined as the masking base. As soon as the definition is changed, the mask is recalculated and updated in the fusion image.

Use the **Edit mask** button for fully interactive adjustments. The existing mask is contoured and opened in a dialog VOI window.

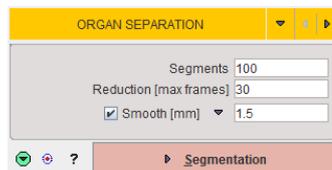


The contoured VOI can be adjusted using all VOI tools of PMOD, or cleared and redefined from scratch. Please refer to the *PMOD Base Functionality Guide* for details about VOI definition. When closing the window with the **Ok** button, the new mask is created by filling the VOI and shown on the **MASK** page.

There is a choice of two segmentation methods, the **ORGAN SEGMENTATION** method, as well as the **CLUSTER ANALYSIS**.

# ORGAN SEGMENTATION

## ORGAN SEGMENTATION Configuration

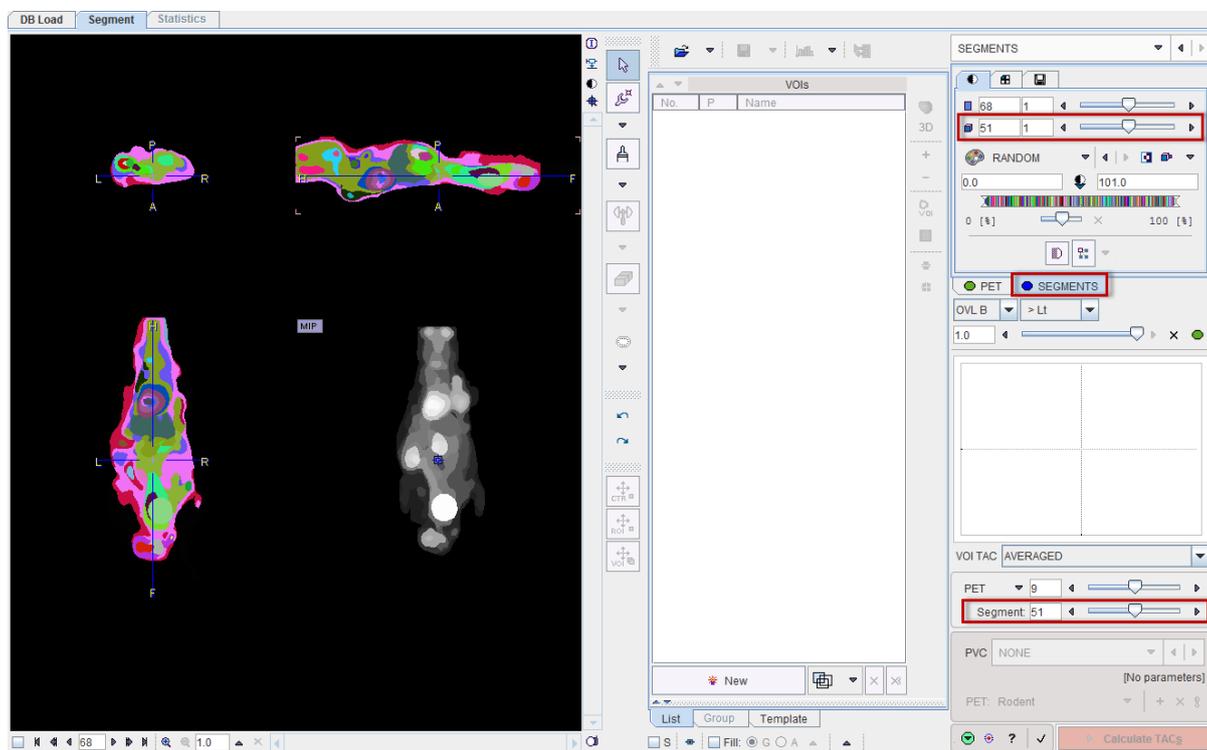


The **ORGAN SEGMENTATION** is the method developed for the *functional segmentation of dynamic rodent images* (on page 3), and incorporates related prior knowledge. The procedure also accounts for specific noise characteristics in the PET image. Therefore, the image **Reconstruction** method (**FBP** or **ITERATIVE**) and the isotope **Half Life** need to be specified properly. Additionally, three parameters need to be set for the segmentation:

- ▶▶ **Segments:** Specifies the maximal number of segments. The method applies a hierarchical approach, therefore segmentations with any smaller number of segments down to 1 will also be available.
- ▶▶ **Reduction:** If the specified number is smaller than the number of frames in the dynamic PET, they are reduced by averaging successive acquisitions.
- ▶▶ **Smooth:** Optional spatial smoothing of the data. The Gaussian kernel in mm can be entered numerically, or by selecting a specific PET system with the down-arrow. Note that smoothing reduces the level of detail which can be obtained by the segmentation.

## Segmentation

Once an appropriate body mask is ready and the segmentation parameters have been defined, please start calculations with the **Segmentation** button. The required processing time depends on several factors including the matrix size, and may take several minutes. The segmentation result is shown on the **SEGMENTS** page, initially in the layout shown below.

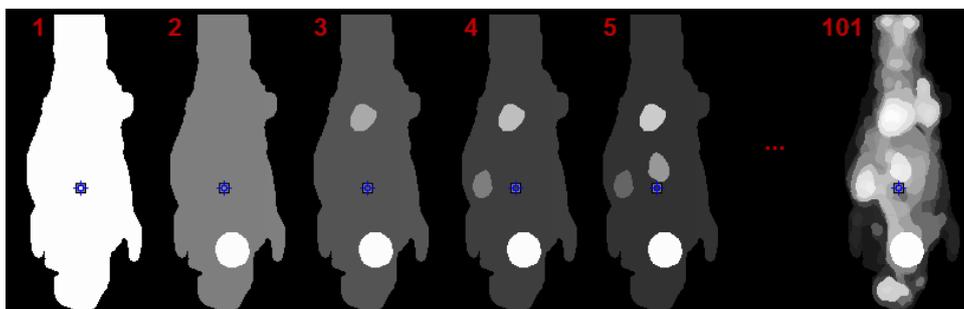


The display shows a fusion of the segmentation result (**SEGMENTS**) with the **PET** image as the reference.

## Image and Segment Inspection

### SEGMENTS Image

The ORGAN SEPARATION results in a hierarchy of segments which is organized as a "dynamic" series with increasing number of segments. The first "frame" has only a single segment which corresponds to the whole body mask. The second segment decomposes the area of the body mask into a background and a single segment, the third shows two segments, and so on, as illustrated below.



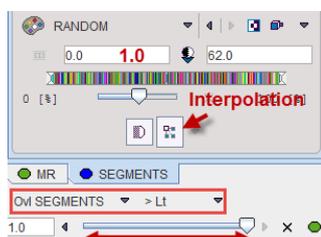
The number of segments can be adjusted by entering the number into indicated fields or dragging the slider. Note that the k-means CLUSTER ANALYSIS doesn't provide a hierarchical segmentation, just a single decomposition.

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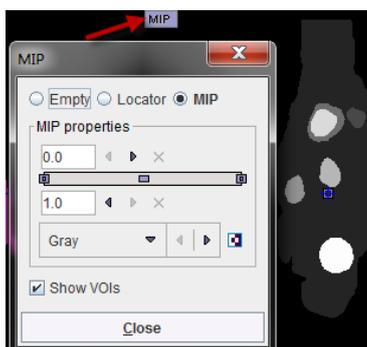
**Note:** On each hierarchical level, the segments decompose the body mask into disjoint, compact volumes.

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Per default the **RANDOM** color table is applied for showing the segments decomposition with the aim of separating neighboring segments. Interpolation is switched off to avoid distracting color effects at segment boundaries.

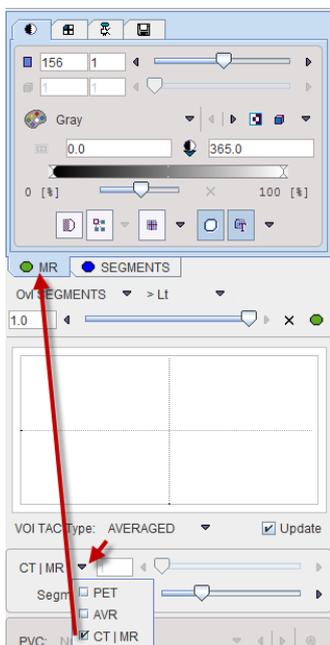


The **MIP** representation of the **SEGMENTS** image is a valuable tool for detecting segments of interest. To have it available please make sure the **SEGMENTS** tab is selected. Furthermore, the full range should be enabled for the MIP.



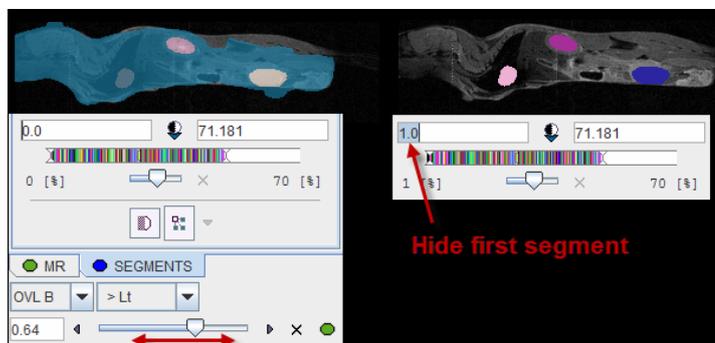
## Reference Image

The reference image which is used as a backdrop for the segments can be selected as illustrated below. The dynamic **PET** as well as the averaged PET (**AVR**), are always available, and the anatomical image optionally.



## Image Fusion

Per default the fusion mode is set to **OVL B >Lt**, meaning that only the segments above the lower threshold are shown in the image. It may be helpful to set the threshold to 1.0 in order to hide the first "background segment". Please use the blending slider to inspect the reference image below the segments.

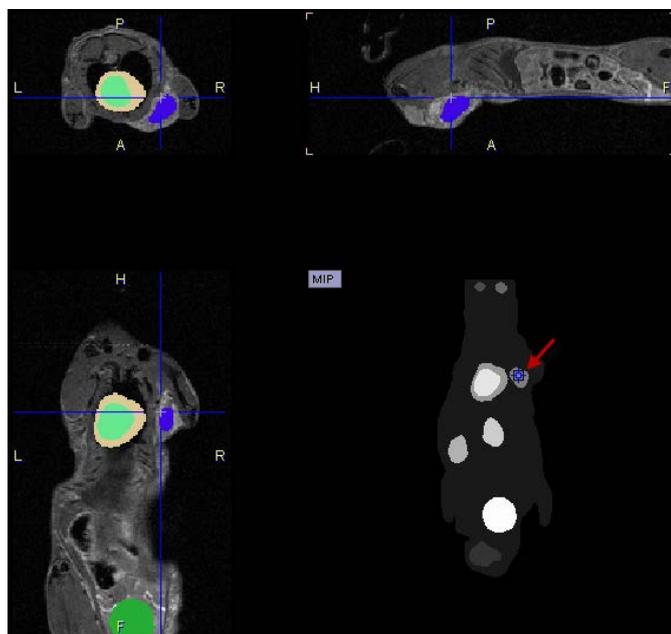


## Navigational Elements

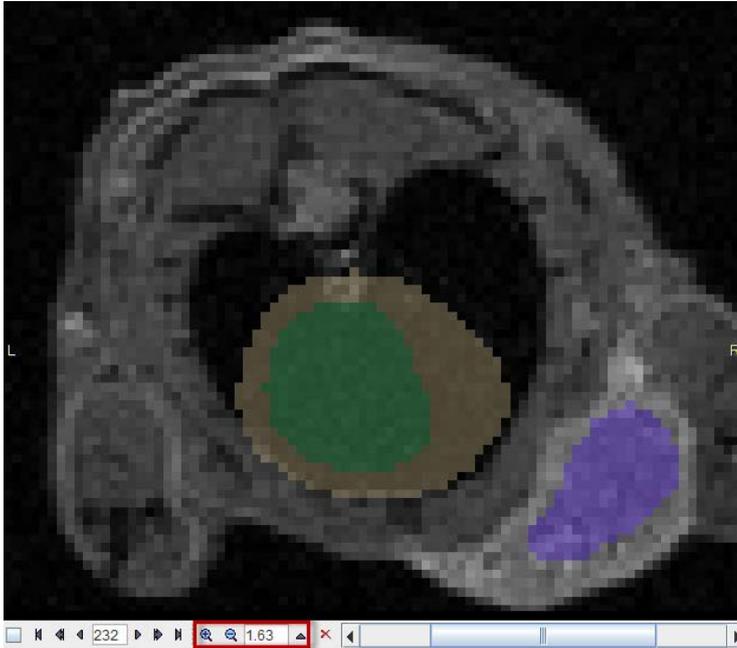
The number of segments is best changed in the lower right slider area. The recommended procedure is to set the **Segment** number to 1, and then gradually increase it by the increment arrow.



The easiest way to localize segments in the image is via the MIP image: Clicking at a segment will update the orthogonal image planes with intersection at the segment. To center the intersection point on the segment please click on the slice images, or use the mouse wheel for scrolling slices.



Close examination might require switching the image layout from the orthogonal layout to a single plane layout and some zooming, and then adjusting the blending. There are zooming elements available below the image, but the image can also be zoomed by the mouse wheel when holding down the `Ctrl` key.

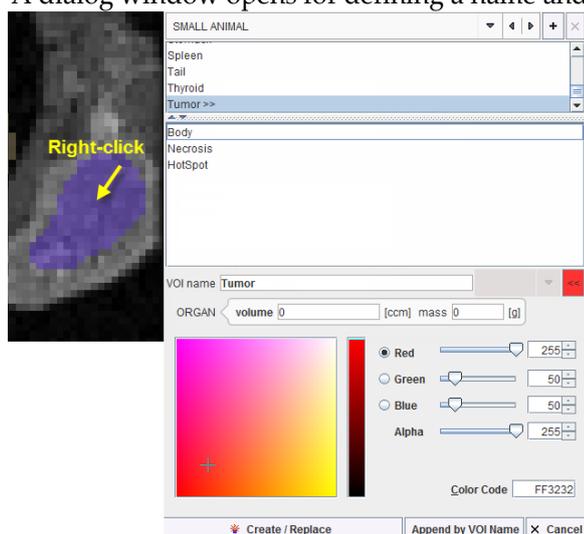


## Segment to VOI Assignment

The basic idea of the whole processing is, that the segmentation produces on some decomposition level segment definitions which represent meaningful tissue structures in the body. Please use the following procedure for creating a named VOI corresponding to a tissue structure of interest (called "organ" or "lesion" in the following):

- 1) Select an appropriate reference image wherein the organs can be seen.
- 2) Adjust the transparency with the fusion slider so that the organ is visible below the segment image.
- 3) Operate the **Segment** slider until one of the segments in the image corresponds to the organ.
- 4) Make sure the **SEGMENTS** tab is selected. Otherwise the assignment will not work as expected.
- 5) Right click into at the segment into any of the slice images shown (NOT the MIP image).

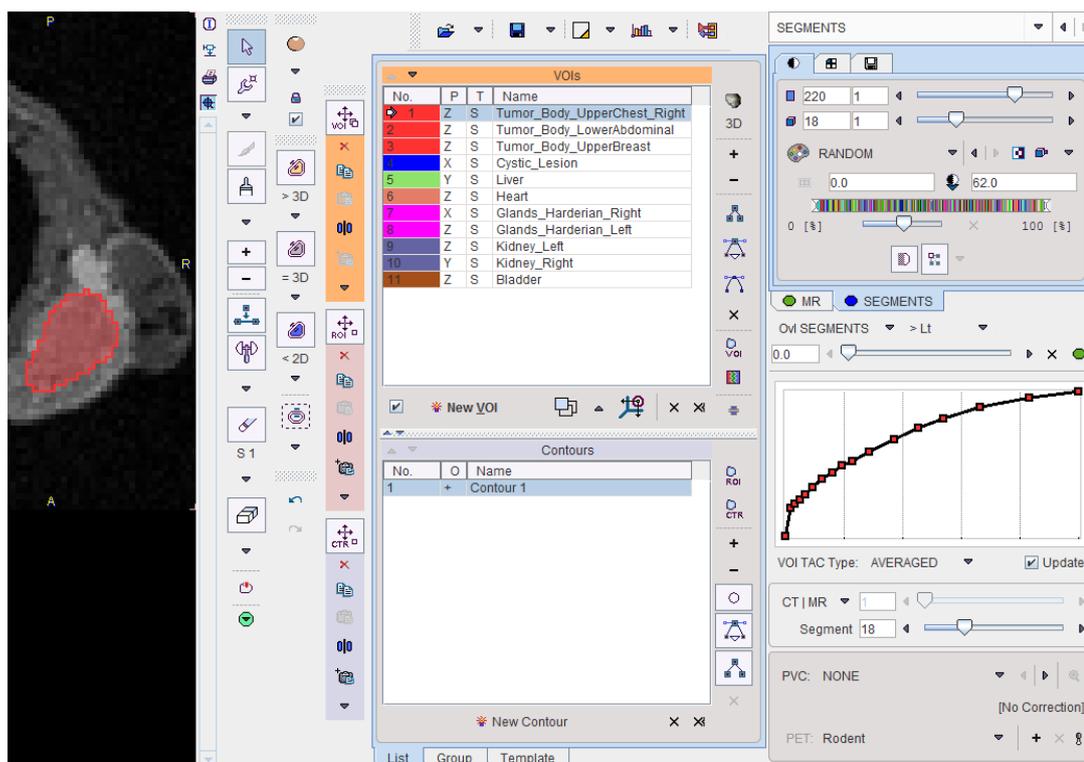
- 6) A dialog window opens for defining a name and a color.



Four lists with pre-defined organ names are available: **DICOM** and **SMALL ANIMAL**. Select either in the list selection in the upper left. Custom name lists can also be prepared and will be available in the selection.

- 7) Select an organ name from the selected list. Organs with trailing >> such as **Tumor>>** offer a substructure list. Other organs like the **Kidneys** offer a **Laterality** choice. Select as appropriate.
- 8) The **VOI name** shown in the text entry field is composed from the selections, but can be edited or completely redefined.  
**CAUTION:** An existing VOI with exactly the same name will be overwritten. In this case please add a suffix to differentiate.
- 9) Change the VOI color.
- 10) Confirm the dialog window with one of the buttons. **Create/Replace** adds the VOI to the list, and overwrites any VOI with the same name. **Append by VOI name** causes the VOI to be merged with an existing VOI of the same name. **Cancel** discards the definition.

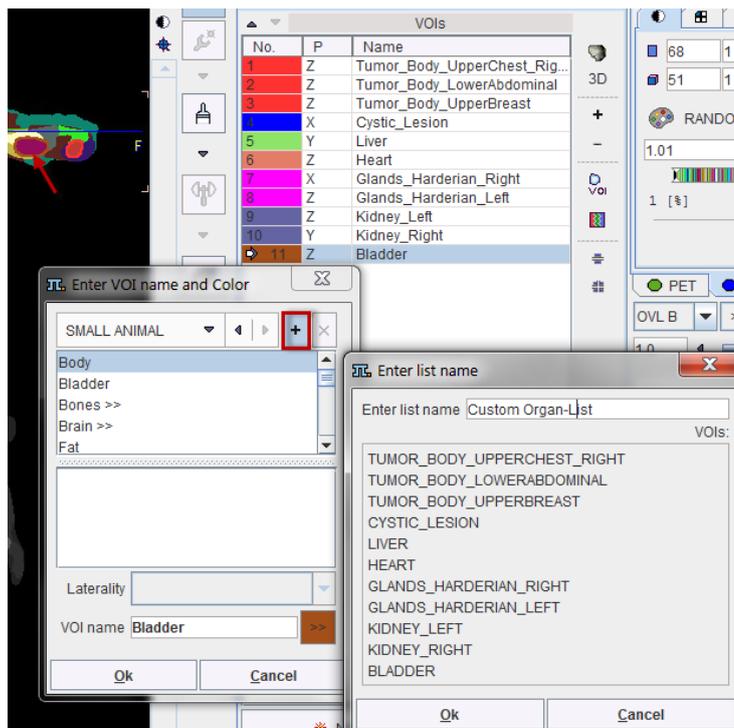
The VOI name appears in the VOIs list, the contours are shown in the image, and the time-activity curve is calculated.



Please repeat the sequence above for all organs of interest.

### Custom Organ List

In the end the **VOIs** list contains all structures of interest. It can easily be converted into a custom list of names for the assignment as illustrated below. Right click into any segment to open the assignment window. Activate the + button, specify a **list name** in the appearing dialog window and confirm the dialog window with ok.

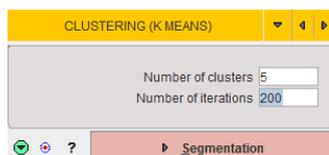


From now on, this list can be used alternatively to the pre-defined lists.

## CLUSTER ANALYSIS (K MEANS)

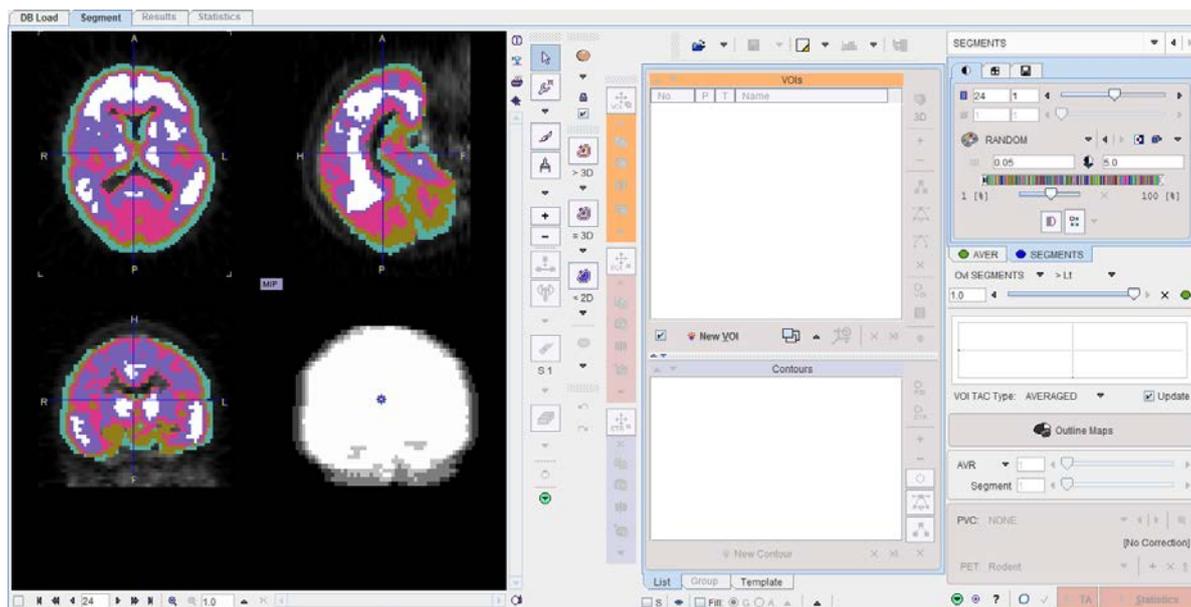
### K-Means Cluster Analysis

The **CLUSTER ANALYSIS** method implements a simple *k-means clustering procedure* (on page 3) [1]. The **Number of clusters** (N) has to be defined, as well as the **Number of iterations** during which the clusters are adjusted. Note that all pixels within the body mask are assigned to one of the clusters, and the process doesn't include any kind of neighborhood relation. The classification is solely based on the signal time course in each pixel.



### Segmentation

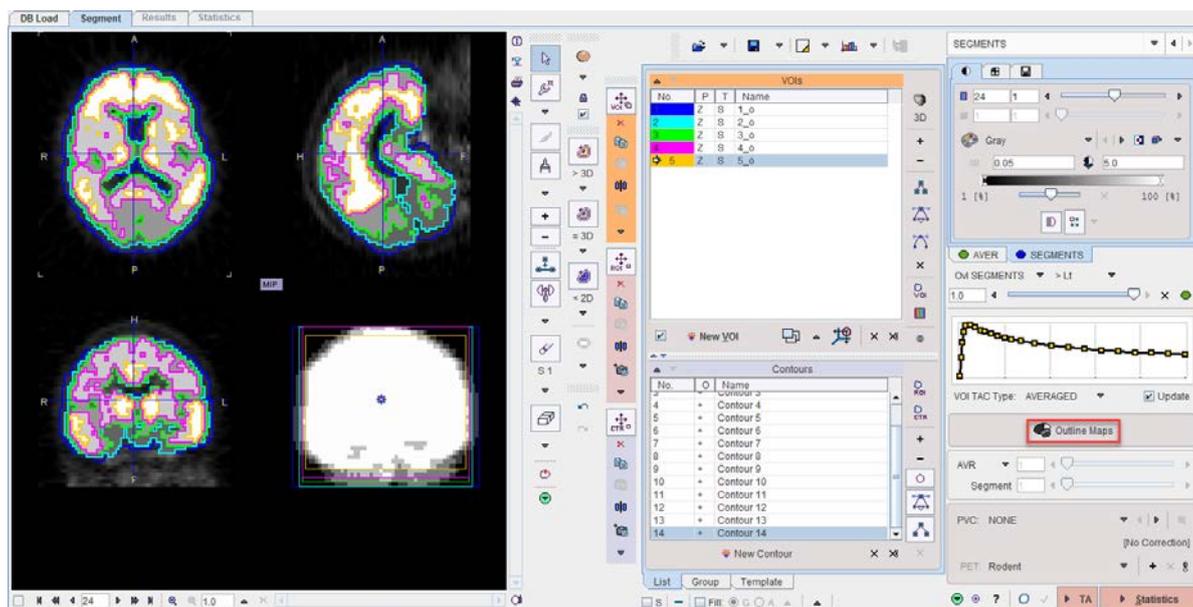
Once an appropriate body mask is ready and the segmentation parameters have been defined, please start calculations with the **Segmentation** button. The required processing time depends on several factors, particularly the number of iterations, and may take several minutes. The segmentation result is shown on the **SEGMENTS** page, initially in the layout shown below.



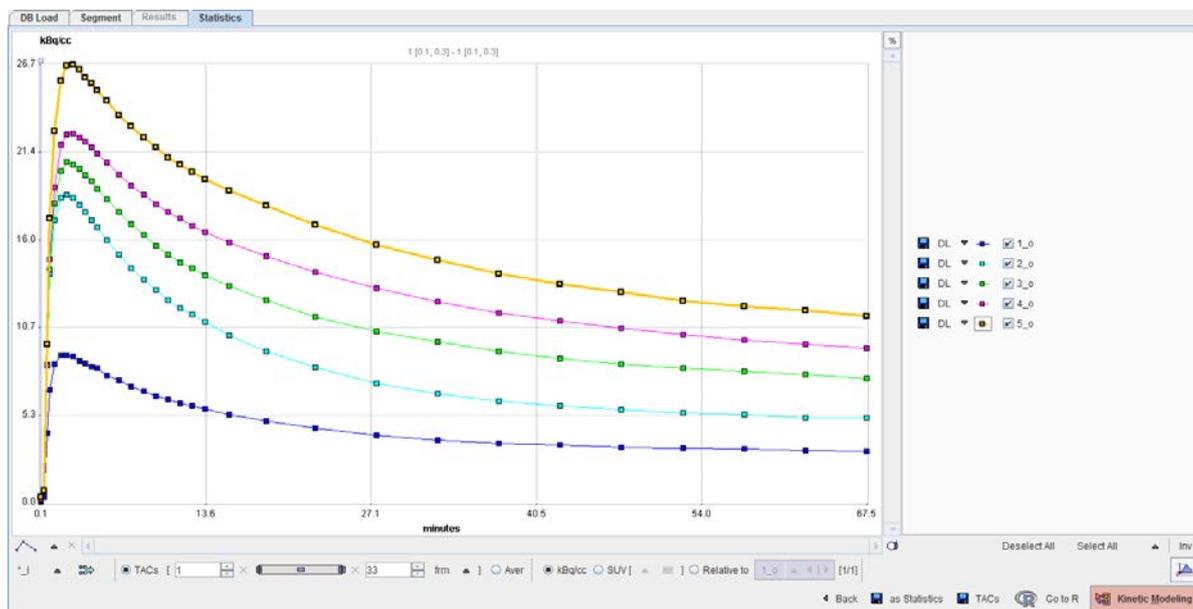
The display shows a fusion of the segmentation result (**SEGMENTS**) with the **PET** image as the reference.

The procedures for *converting clusters into VOIs* (on page 21) are the same as for the **ORGAN SEGMENTATION**, except that the clustering only provides a single decomposition of the volume. Correspondingly, there is a static label images in the **SEGMENTS** tab rather than a dynamic series showing the volume in a hierarchical structure of decompositions, and the **Segment** slider is inactive.

All clusters can be outlined at once using the **Outline Maps** button, as illustrated below.



Finally, **Statistics** will calculate the average TACs of the clusters and show them on the **Statistics** page.



## CLUSTER ANALYSIS (SUPERVISED)

*Supervised clustering* (on page 3) is based on the specification of a set of TACs which represent the average time course in specific tissues. Typically, these kinetic class TACs are generated using a representative set of PET series which are segmented by a different means, e.g. by using the anatomical information in matched MR series [6,7]. For use in PSEG it is assumed that the kinetic class TACs are already available as one file per TAC.

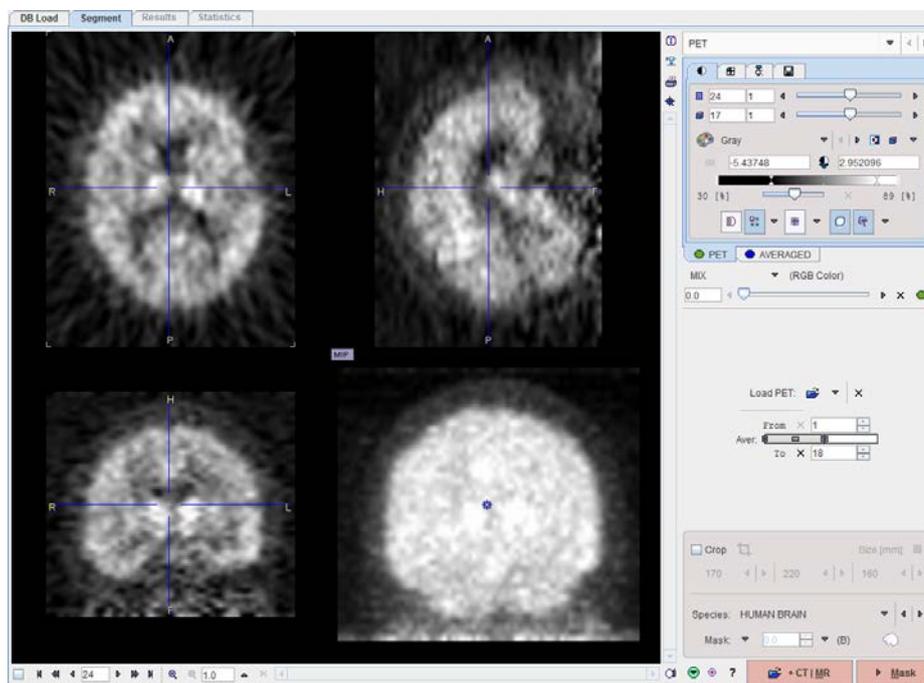
---

**Note:** If the class TACs have been created using z-score normalized data, the image series to be segmented also has to be normalized before loading into PSEG.

---

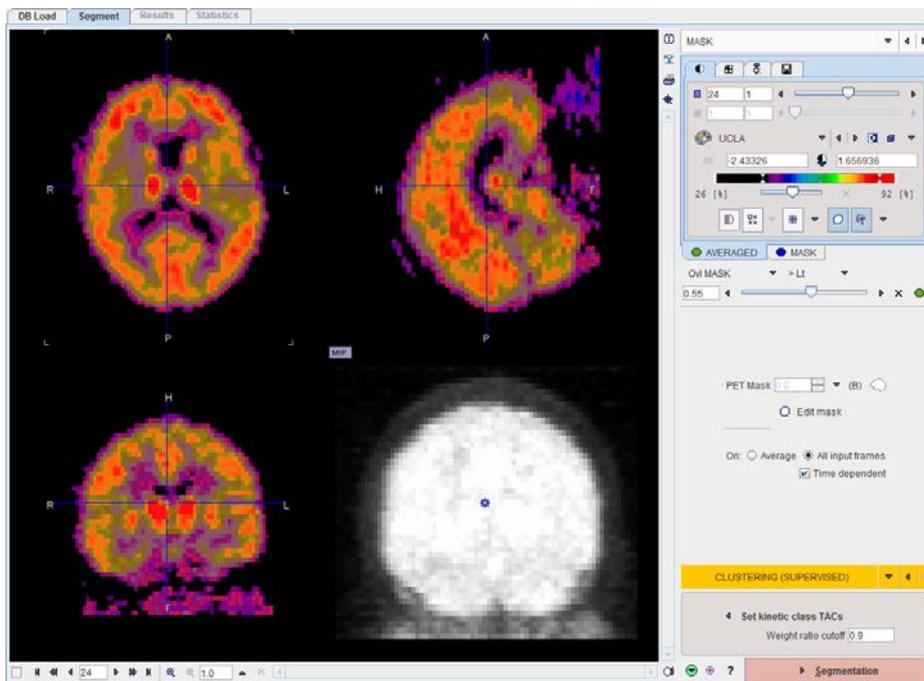
### Image Loading

Use **Load PET** for loading the image series to be segmented. The example below illustrates the situation after loading a [11C]PIB scan which has been z-score normalized. This process results in negative numbers, which are evident in the lower colorbar threshold.



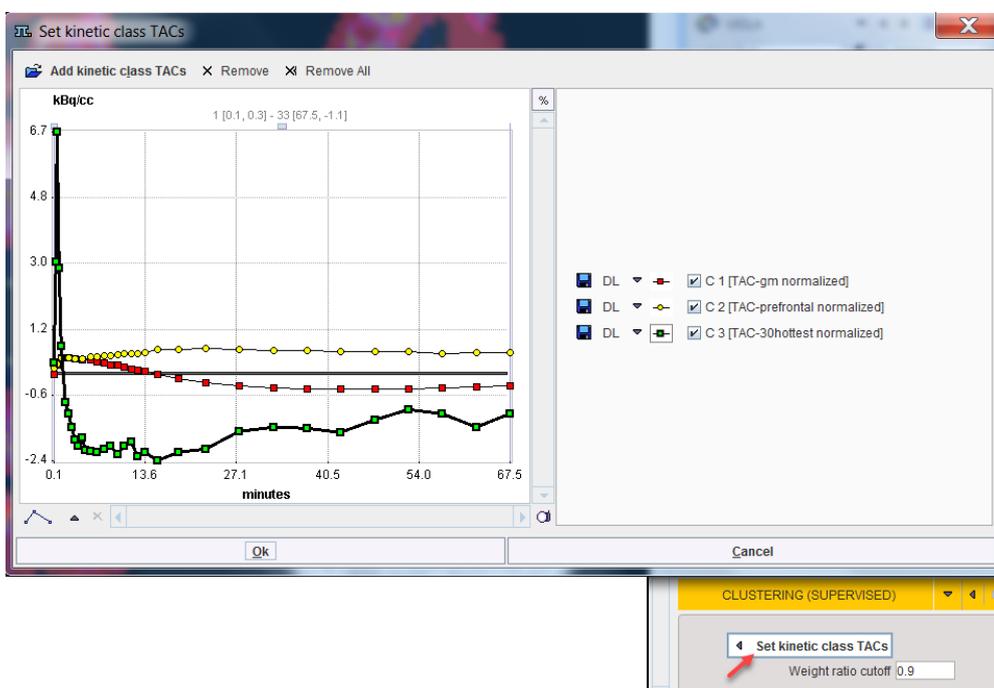
## Masking

After configuration of the mask and proceeding with **Mask** to the MASK page, the segmentation method can be switched to **CLUSTERING (SUPERVISED)**.

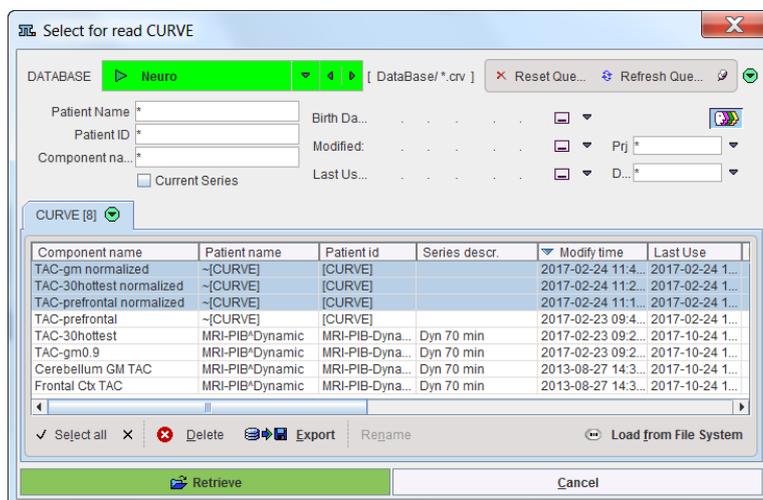


## Kinetic Class TACs Definition

**Set kinetic class TACs** opens a dialog window for specifying the kinetic class TACs. In the illustration below C1 corresponds to normal grey matter, C2 to specific binding, and C3 to the blood pool similar to Fig. 1 in [7]. Note the normalized curve shapes which are very different from normal uptake TACs.



**Add kinetic class TACs** opens a dialog window for selecting the curves, either from a database as below, or via **Load from File System** from the disk. Retrieve adds the selected curves to the panel. Curves can be discarded by selecting them to the right and use **Remove**, or **Remove all**.

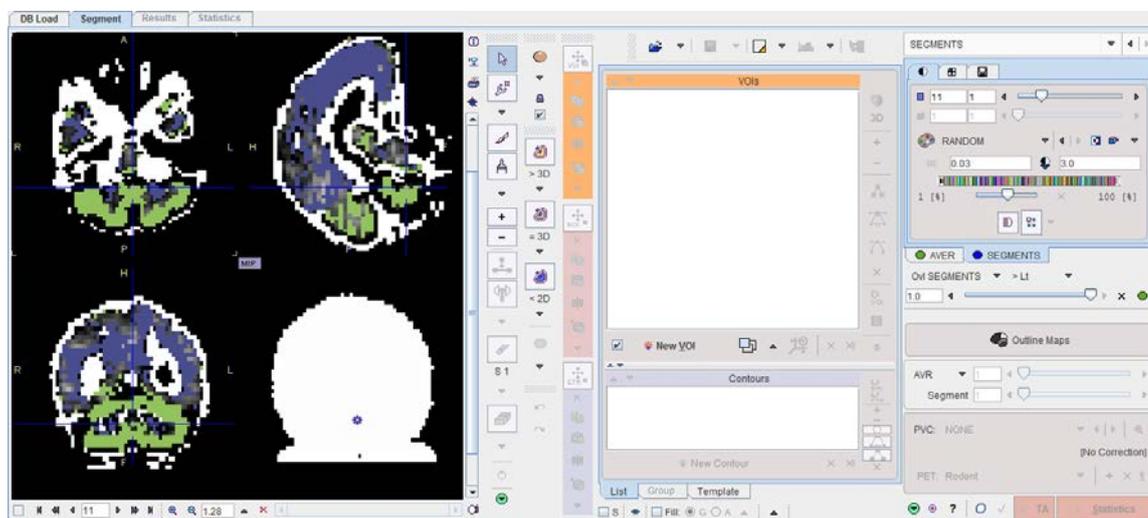


Note that the selection is serialized during subsequent uses of **CLUSTER ANALYSIS (SUPERVISED)**.

## Segmentation

**Segmentation** starts the the classification procedure. For each pixel in the mask it finds the optimal linear combination of all kinetic class curves so that all coefficients are non-negative (NNLS, non-negative least squares). It then assigns the pixel to the class with the highest coefficient (or weight), if its ratio to the sum of all coefficient is greater than **Weight ratio cutoff**.

The resulting segment image is shown on the **SEGMENTS** page, and the segments can be converted into VOIs using the **Outline Maps** button.

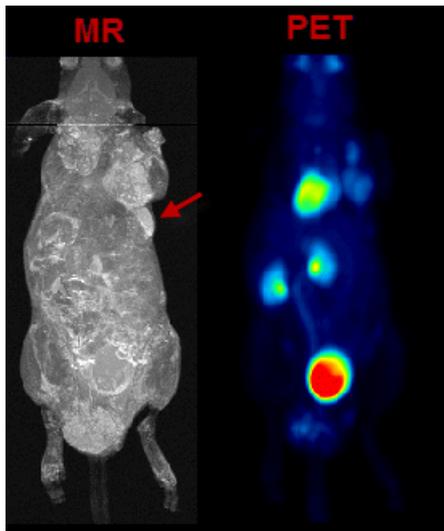


## VOI Editing and Manual VOI Addition

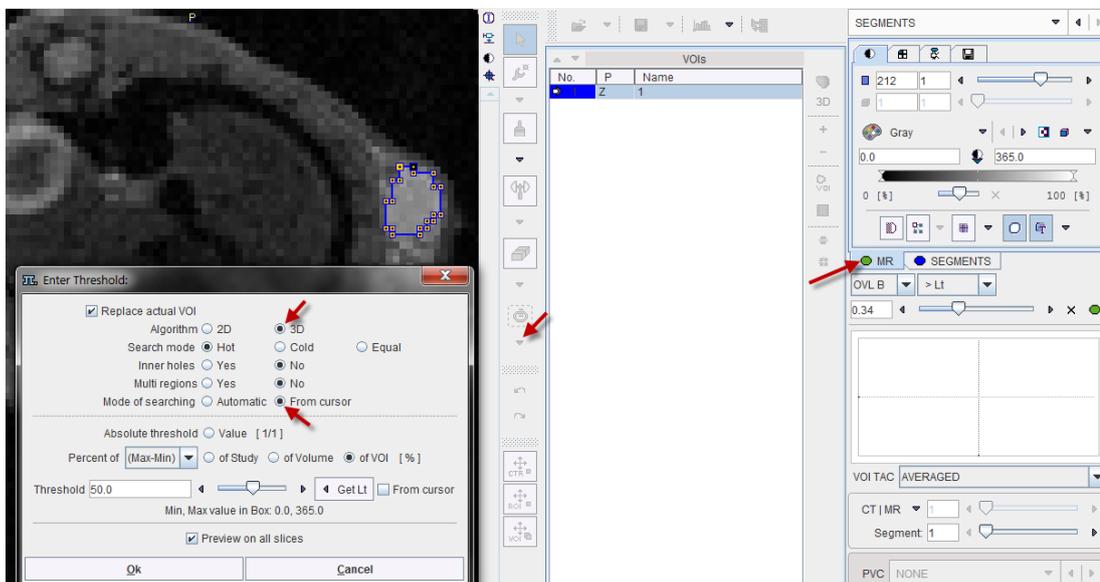
The VOIs created by the segment assignment process are regular PMOD contour VOIs which can be adjusted in many ways. For instance, if the segmentation produced two complementary parts of a structure, their VOIs can easily be merged into a single VOI.

### Manual VOI Addition

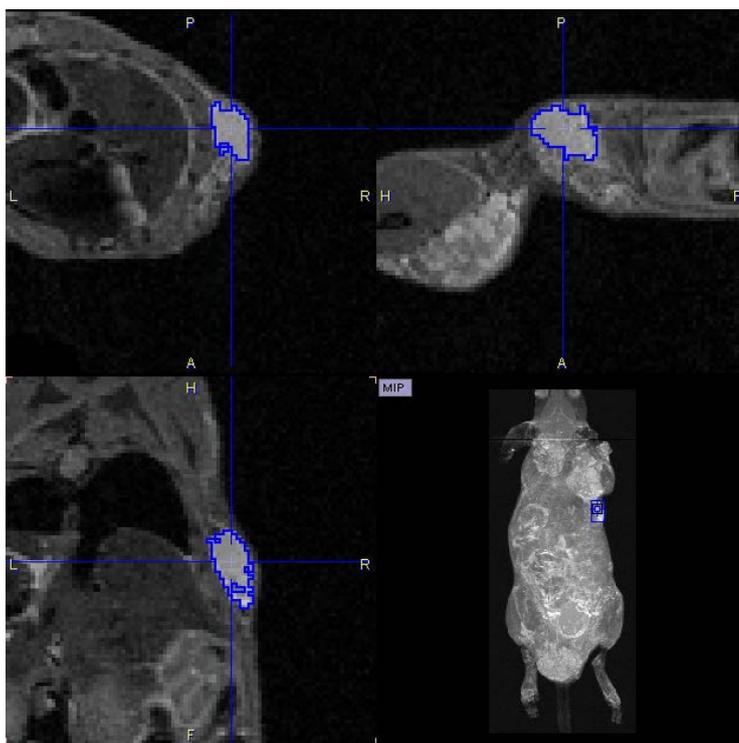
VOIs can also be added in a completely manual manner. In the example data set, for example, there is a bright part in the MR image which does not take up FDG.



In the present situation, a VOI can be obtained from the MRI image using the iso-contouring tool as illustrated below. First, the MR image tab is selected to direct the iso-contouring operation to the MR image content, not the SEGMENTS. Then, the iso-contouring is started. The parameters are set to 3D with Multi regions off, to create a single compact VOI. Setting the Mode of searching to From cursor will ensure that VOI creation starts at the location where the user clicks. The iso-contour is shown in the image, and the contouring level may be adjusted by the Threshold setting.

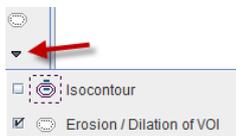


After closing the dialog window with **Ok**, the contouring result is shown as illustrated below, and the VOI can be labeled.

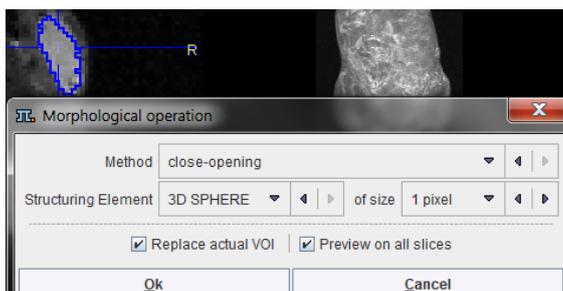


## Morphological Operations

Morphological operations support convenient functions for modifying the created VOIs, such as shrinking, growing, removing holes and single pixels. Please select the VOI to be modified in the list, then activate the **Erosion/Dilation** function from the VOI tools area.

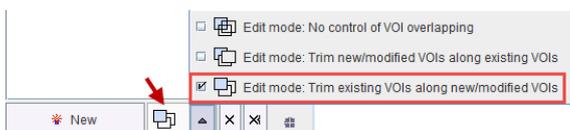


A dialog window appears which offers a list of **Methods** with corresponding **Structuring Elements** and their **size**. The changes to the VOI are immediately reflection when configuring the operation. Note that the result may replace the existing VOI, or added as a new VOI.



## Non-overlapping of VOIs

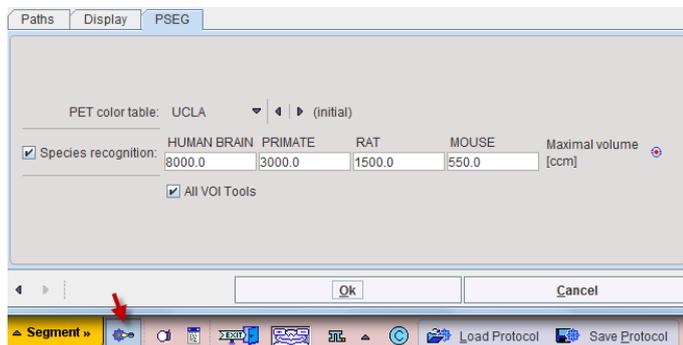
In order to avoid the overlapping of VOIs, which causes problems with the partial-volume correction, the VOI **Edit mode** is pre-configured accordingly.



If VOIs are created by the merging of existing VOIs, please make sure that the original VOIs are removed.

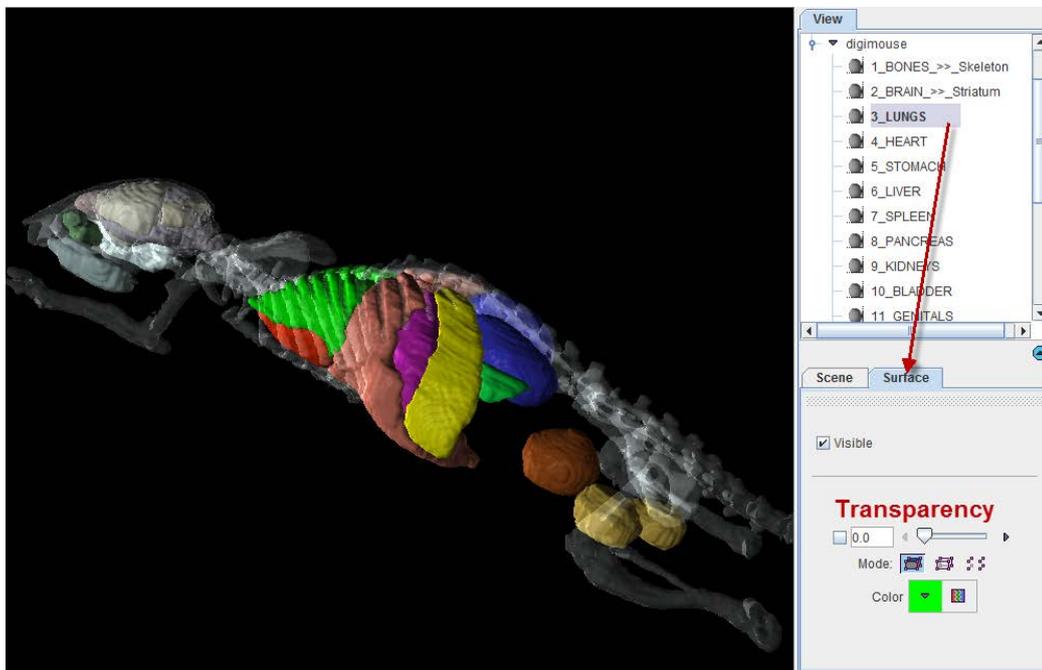
## Palette of VOI Tools

Per default only a subset of the PMOD VOI tools is available in PSEG. To be able to use the full VOI functionality please enable **All VOI Tools** in the configuration.



## Digimouse Visualization (Option)

During the assignment of segments to organs it may be helpful to relate the information in the image display with an anatomical whole-body atlas. The PSEG tool includes a 3D rendering of the Digimouse Atlas which is only available if the PMOD license contains the P3D rendering option. In this case the corresponding  button in the lateral taskbar is active and launches the tool as illustrated below.



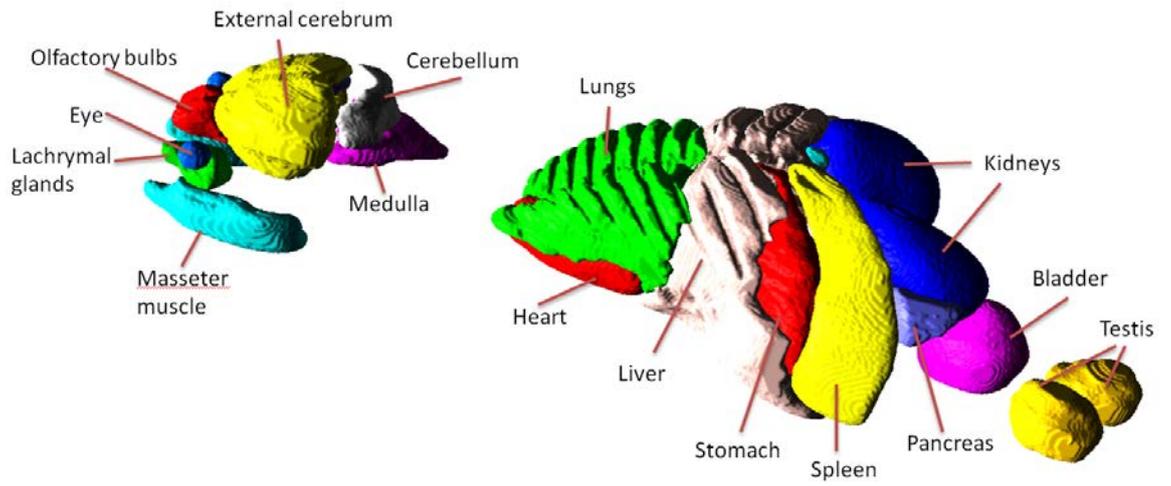
Note the organ list in the **View** panel to the right which allows selecting individual objects and changing their representation. In the example above, the **3\_LUNGS** object is selected, with the corresponding **Surface** panel giving access to its properties. For instance, by removing the **Visible** check the lungs can be hidden from the rendering. Note that multiple objects can be selected and a property such as the transparency changed for all of them at once. Please refer to the *PMOD 3D Rendering Tool Guide* for more information about the renderings options.

---

**Note:** The 3D Digimouse window can remain open while assigning the segments. After hiding irrelevant structures it is recommended to reduce the window size and hide the control area with the  button in the lateral taskbar.

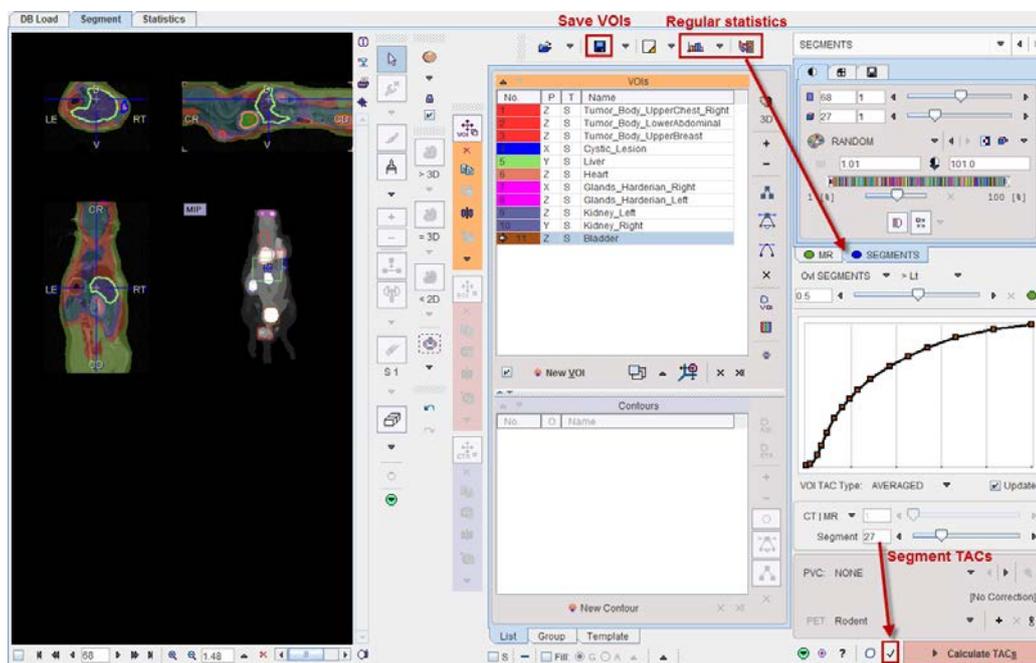
---

The structures available in the Digimouse phantom are illustrated below.



## Time-Activity Curve Calculation and Statistics

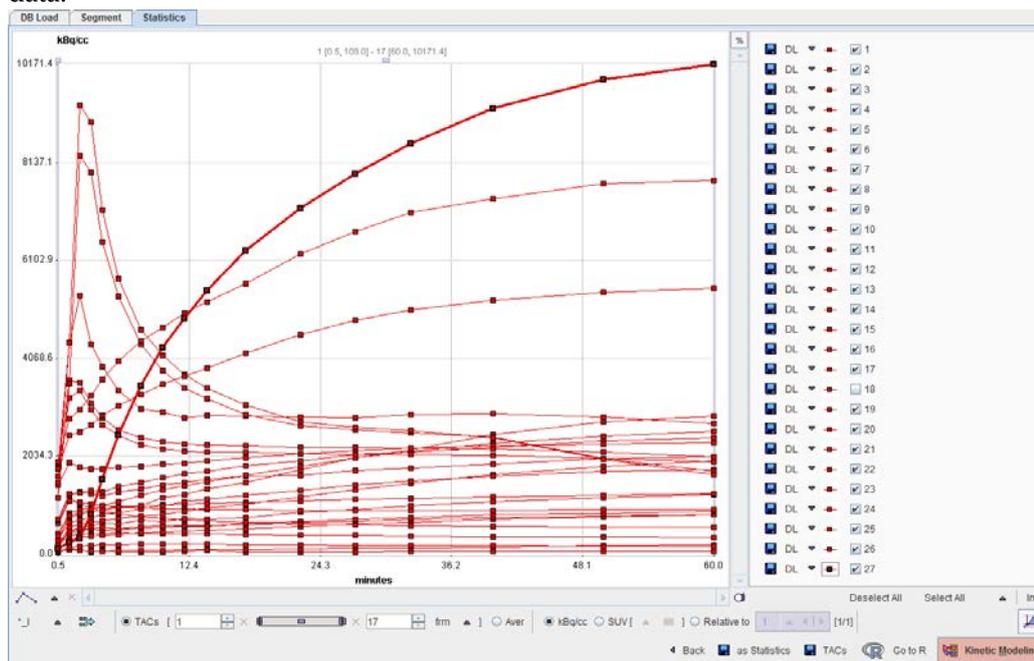
At the end of segment assignment and VOI editing the VOIs list contains all defined structures of interest. It is recommended saving them separately using the button illustrated below.



There are different variants for calculating statistics.

- 1) The regular statistics buttons above the VOIs list will operate on the selected image. In the example above, statistics would be calculated from the SEGMENTS image and be useless. However, the user might select the first image tab and the averaged PET as the reference image. In this case, statistics will result in the average PET uptake.
- 2) The  button in the lower right calculates the TACs of all segments decomposing the body mask at the level defined by the **Segment** number. The example result with 27 **Segments** is illustrated below. It provides a convenient overview of the kinetics in the

data.



- 3) Normally the **Calculate TACs** action button will be used for producing the end result: The average PET uptake curves in all defined VOIs are calculated and presented on the **Statistics** page.

### Partial-Volume Correction (PVC)

Optionally, the **PVC** can be enabled.

PVC LMA GTM

Voxels to be kept 15 [%]

PET: Rodent

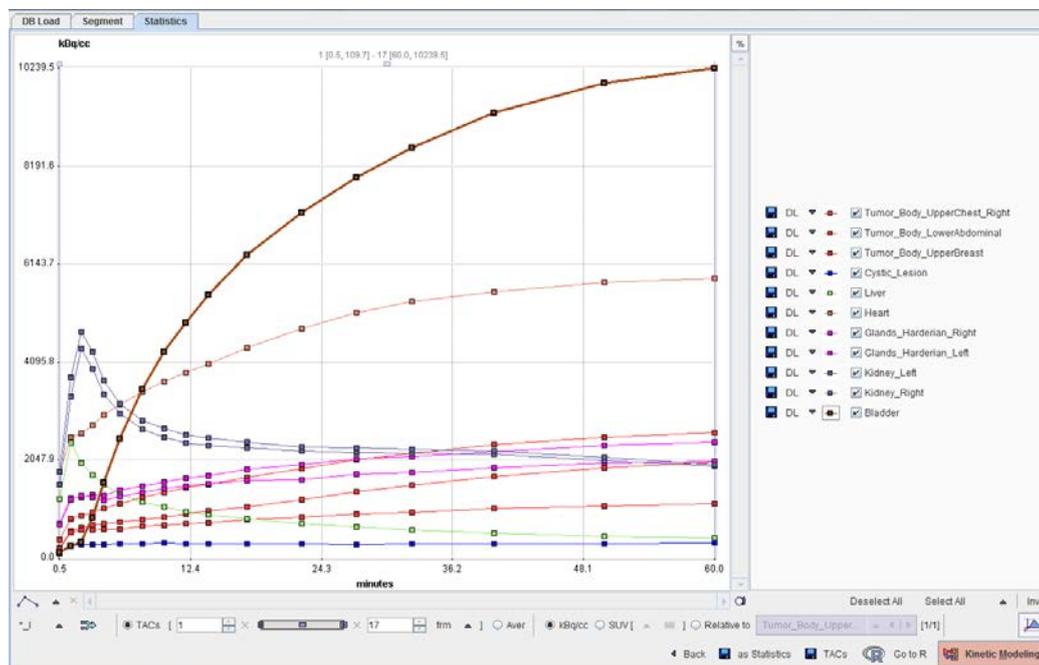
The **LMA GTM** method uses information from the segmentation process. Assuming that the inner-most part of the segments is least affected by the partial-volume effect, it uses only a fraction of the segment data for the segment average calculation. The percentage can be set by the **Voxels to be kept** parameter. The algorithm uses the Geometric Transfer Matrix (GTM) approach for spillover compensation at a fine segmentation level which is not shown to the user. The corrected signal for all user-assigned segments is then constructed from the corrected signals contained in the segments. The PVC procedure requires that the defined VOIs are non-overlapping. If there is overlap, the results will not be correct.

The resolution of the imaging system is crucial for the GTM method. There is a list-selection for pre-defined scanners in the lower part of the PVC area, and new scanners can easily be added. Default is the generic **Rodent** scanner with isotropic resolution of 1.5mm FWHM.

With PVC enabled, PSEG will provide both the corrected and uncorrected uptake curves on the **Statistics** page.

## Statistics Results

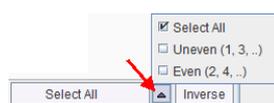
The **Statistics** page is dedicated to the visualization and further use of the time-activity curves. The example below illustrates the result of calculating TACs with PVC enabled.



The **Back** arrow in the lower right is a convenience button for switching back to the **Segment** page of PSEG which generated the statistics.

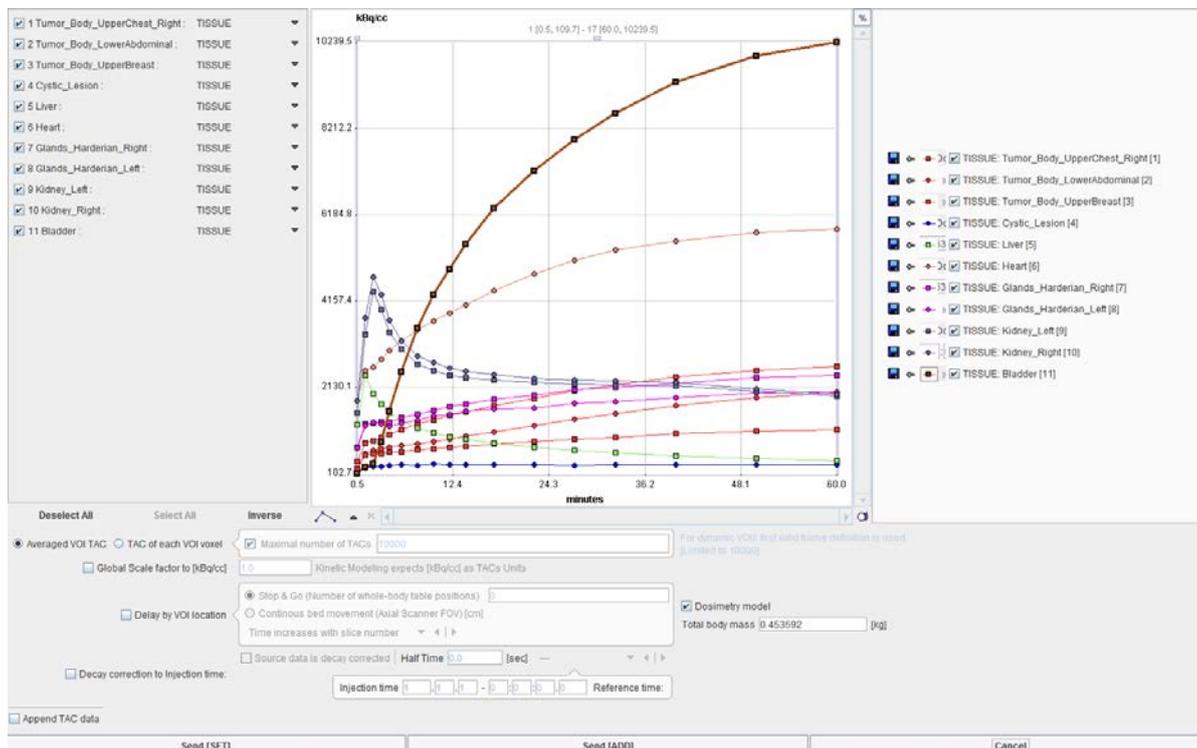
### Selection of Curve Subsets

Sometimes, only a subset of the TACs may be relevant for further processing and should be selected in the list to the right. For instance, if the optional partial-volume correction is enabled for the statistics calculation there will be pairs of curves for all VOIs: one curve representing the original data, and one curve ending with **\_C** representing the partial volume corrected data. There are some convenience buttons supporting this selection: **Deselect All** switches all curves off. **Select All** switches all curves on. **Inverse** inverts the current selection. Note the arrow button which allows to quickly select dedicated subsets.



## Transfer of TACs to the PKIN Tool

The **Kinetic Modeling** button allows directly transferring the TACs to the PKIN tool for modeling. It opens the following dialog window.



The TACs to be transferred can be selected by checking the box in the left column. The right side lists the currently selected TACs. **Send [SET]** transfers the selection to the active workspace in PKIN. If the **Append TAC data** is enabled, the TACs will be added as additional tissue regions, otherwise the existing tissue data is overwritten. **Send [ADD]** first creates a new workspace in PKIN before actually transferring the selection. If **Dosimetry model** is enabled, the **OLINDA Residence times** model is selected in the PKIN tool.

## Saving the Statistics

The **Save TACs** button allows saving all curves in a simple tab-delimited text file as illustrated below. Such a file can easily be imported into other programs for processing and visualization.

start[seconds]	end[kBq/cc]	Tumor_Boi	Tumor_Boi	Tumor_Boi	Cystic_Les	Liver	Heart	Glands_Ha	Glands_Ha	Kidney_Lel	Kidney_Rig	Bladder	Tumor_Boi	Tumor_Boi
0	60	380.114	221.5528	207.4992	102.6775	1238.673	1803.045	725.8459	707.2146	1535.606	1806.721	109.6588	387.7057	267.4343
60	120	820.8457	576.1053	539.8545	243.1883	2402.688	2528.803	1260.343	1227.332	3365.928	3770.818	249.8382	854.3848	692.7699
120	180	880.5907	640.0921	579.2888	279.2121	1988.089	2597.902	1276.144	1295.933	4378.474	4721.757	330.0487	950.0287	774.1116
180	240	944.6486	682.1565	580.5773	278.2239	1731.391	2780.089	1321.961	1267.1	3953.037	4303.564	838.6104	1043.522	796.8466
240	300	1031.131	713.262	602.1958	289.8592	1532.681	2987.612	1296.596	1209.838	3421.908	3700.216	1570.082	1123.159	846.2061
300	420	1134.957	758.1786	605.9648	301.8691	1341.723	3215.362	1418.684	1293.919	3030.759	3218.521	2486.536	1287.885	889.2018
420	540	1261.409	803.9566	661.589	292.8661	1172.9	3480.49	1495.719	1368.695	2683.507	2866.252	3519.747	1424.484	921.5853
540	660	1365.036	858.1505	682.6136	321.1853	1061.131	3695.217	1592.272	1445.226	2518.211	2702.75	4307.815	1527.267	1041.758
660	780	1470.753	917.2937	720.9745	308.083	972.9532	3876.665	1678.036	1495.743	2408.823	2578.388	4908.41	1681.522	1080.932
780	900	1542.412	979.6508	741.7846	306.457	906.2761	4061.367	1726.597	1559.568	2363.485	2506.825	5493.842	1749.019	1196.057
900	1200	1691.433	1075.638	801.5291	303.2694	813.1308	4392.316	1845.886	1620.312	2297.27	2422.741	6327.608	1944.444	1319.51
1200	1500	1874.588	1224.403	858.6415	303.1281	719.8015	4799.821	1946.554	1641.805	2235.158	2329.366	7218.813	2136.715	1523.124
1500	1800	2060.648	1379.77	919.2335	290.6183	650.2844	5121.743	2070.251	1750.531	2209.377	2302.028	7945.154	2323.963	1746.191
1800	2100	2192.781	1519.666	956.6143	301.2032	592.5841	5361.922	2122.306	1792.865	2193.197	2279.914	8575.592	2464.544	1919.583
2100	2700	2368.967	1704.15	1032.841	301.993	523.9605	5567.85	2216.431	1879.409	2174.376	2241.131	9307.16	2657.097	2160.183
2700	3300	2526.163	1880.994	1091.014	307.5017	456.5826	5764.392	2355.619	1990.5	2038.278	2100.789	9921.196	2838.526	2383.876
3300	3900	2618.536	2001.92	1132.796	310.3426	410.5404	5839.346	2416.858	2026.613	1923.791	1955.769	10239.54	2953.921	2509.716

Save as Statistics allows storing the information in a format suitable for statistical analysis.

### Average in Frame Range

For dynamic data there is an easy way to calculate the average regional uptake in the regions in a certain frame range: with the **TACs** radio button selected define the frame range to be averaged. As soon as the **Aver** radio button is switched on, the averaged uptake statistics is calculated and listed, replacing the curves display.

Name	AVERAGED	PVC	Volume [ccm]	Unit
Tumor_Body_UpperChest_Right	2092.8842063789966	2360.4969738375758	0.04325359518170275	kBq/cc
Tumor_Body_LowerAbdominal	1482.2243946852848	1854.5470206968246	0.09898793919852261	kBq/cc
Tumor_Body_UpperBreast	939.600301747989	1147.8828518775201	0.03863181787869844	kBq/cc
Cystic_Lesion	303.5237516878906	253.78431037164503	0.027613656797696617	kBq/cc
Liver	676.8367141227446	661.3894357988911	0.6509100540914673	kBq/cc
Heart	5040.657957176254	5574.471329227571	0.12730363811819456	kBq/cc
Glands_Hardierian_Right	2068.632903829329	3079.1018637380294	0.010569634169739807	kBq/cc
Glands_Hardierian_Left	1770.9036320198343	2579.7764951644403	0.00795647738238716	kBq/cc
Kidney_Left	2248.7737113828143	3045.541566910282	0.05840600431433712	kBq/cc
Kidney_Right	2345.1029622395445	3136.008497668851	0.06521191266348693	kBq/cc
Bladder	7884.777375630602	12759.211473034275	0.1985804146687462	kBq/cc

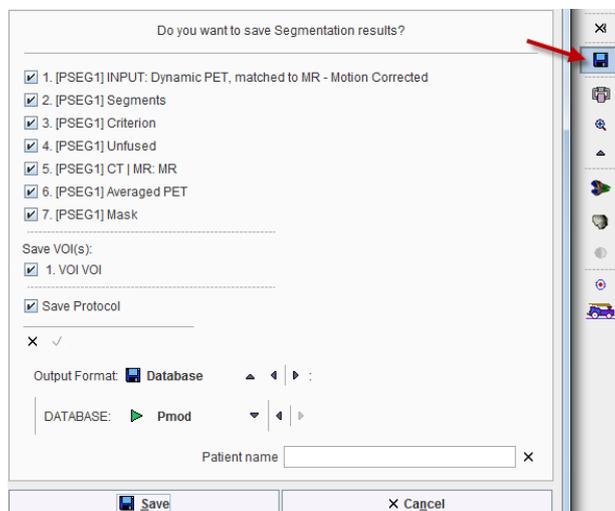
\*\_C ▲ TACs 17 frm Aver %

as Statistics Back Copy to Clipboard

## Results Saving

### Saving of Data

The easiest way to save information is via the *Save all* button in the lateral taskbar as illustrated below.



All images arising during processing are available for saving, as well as the final VOIs and a protocol. Please select the information of interest and saved them to a database or a directory.

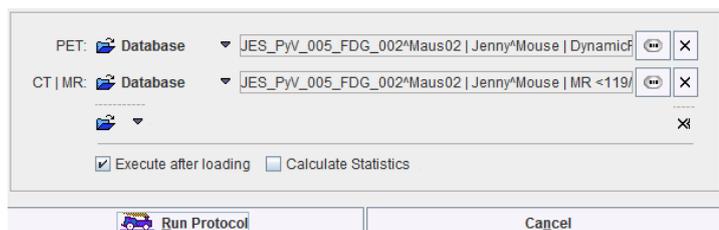
### Protocol Saving

The final processing configuration can separately be saved using the **Save Protocol** button.



Such a protocol includes definition of the input data, the parameters of the different processing stages, VOIs generated by assigning the segments to organs as well as manually edited VOIs. It is recommended saving a protocol after every completed data processing, so that at any later time the configuration can be retrieved, verified and modified to try variations of the processing parameters.

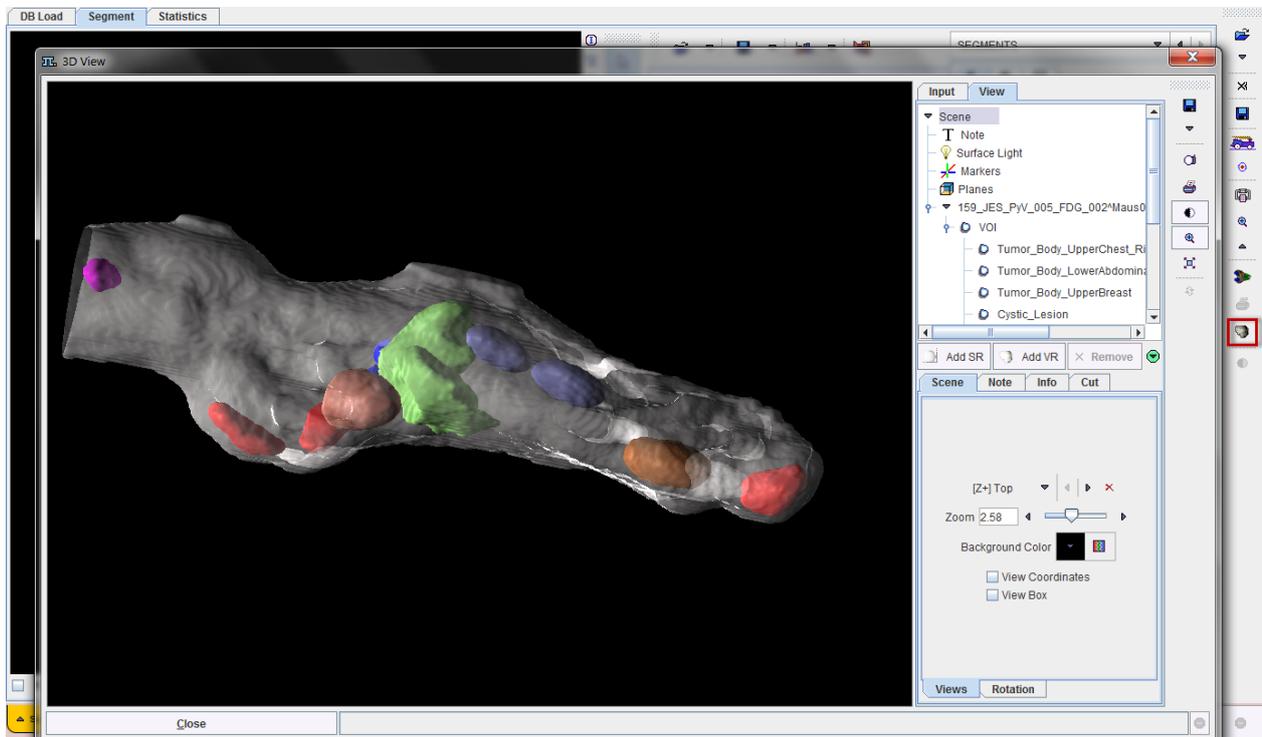
**Load Protocol** allows retrieving a protocol. It shows the following dialog window.



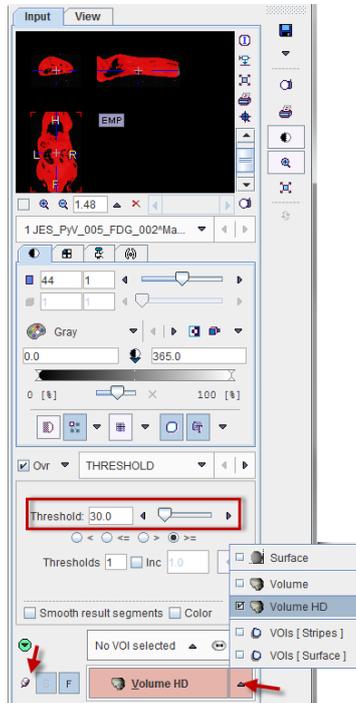
The data sources may be redefined. If **Execute after loading** is disabled, only the data will be loaded and the configurations recovered. Otherwise, the full segmentation will be performed and the result shown together with all VOIs of the saved session.

## 3D Visualization (Option)

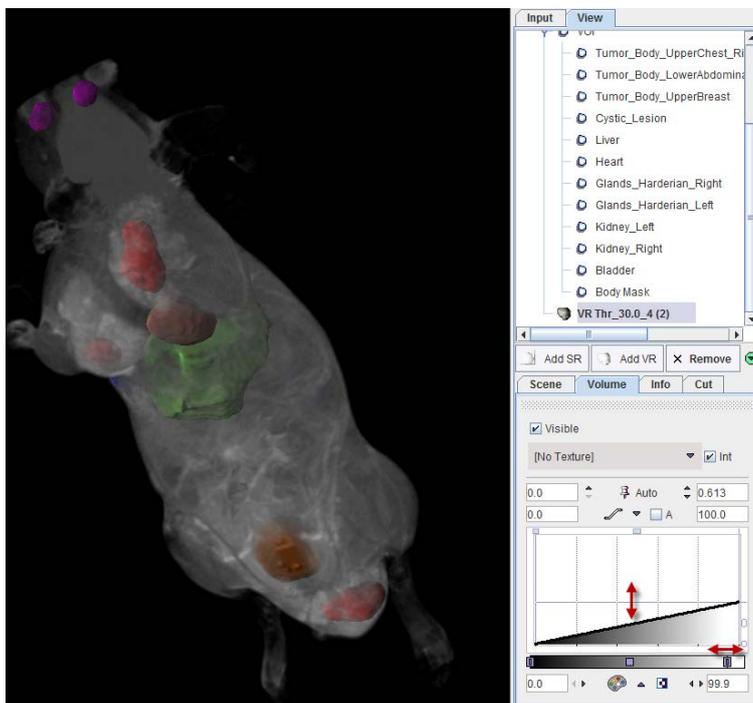
If the 3D option is part of the PMOD license, the VOIs created in the PSEG tool can be conveniently visualized as a 3D scene. When activating the  button in the lateral taskbar, a **3D View** window is opened, the data loaded, and the VOIs rendered as surfaces together with the body mask.



The image data is also available for the scene. Illustrated below is adding a volume-rendering (VR) of the MRI. The **Input** tab is selected, a **Threshold** level set such that all body pixels are included (**Ovr** check), the "Append" pushpin set for adding to the existing VOI renderings, and **Volume HD** activated.



The result after switching off the body mask and increasing the transparency of the VR object is illustrated below. Interactive exploration of such a scene will greatly add to the understanding of the lesion localization.



Please refer to the *PMOD 3D Rendering Tool User's Guide* for more information about how to

## References

explore and enrich the 3D scene.

- 1) Maroy R, Viel T, Boisgard R, Comtat C, Trebossen R, Tavitian B. Fast and Accurate PET Preclinical Data Analysis: Segmentation and Partial Volume Effect Correction with no Anatomical priors. *IEEE Nuclear Science Symposium*; 2008:5498-5001.
- 2) Maroy R, Boisgard R, Comtat C, Frouin V, Cathier P, Duchesnay E, Dolle F, Nielsen PE, Trebossen R, Tavitian B. Segmentation of rodent whole-body dynamic PET images: an unsupervised method based on voxel dynamics. *IEEE Trans Med Imaging*. 2008;27(3):342-54.
- 3) Maroy R, Boisgard R, Comtat C, Jegou B, Fontyn Y, Jan S, Dubois A, Trebossen R, Tavitian B. Quantitative organ time activity curve extraction from rodent PET images without anatomical prior. *Med Phys*. 2010;37(4):1507-17.
- 4) Rousset OG, Ma Y, Evans AC. Correction for partial volume effects in PET: principle and validation. *J Nucl Med*. 1998;39(5):904-11.
- 5) Velamuru PK, Renaut RA, Guo H, Chen K. Robust Clustering of Positron Emission Tomography Data. Paper presented at: Joint Conference of the Classification Society of North America and Interface Foundation of North America, 2005; St. Louis.
- 6) Turkheimer FE, Edison P, Pavese N, Roncaroli F, Anderson AN, Hammers A, Gerhard A, Hinz R, Tai YF, Brooks DJ: Reference and target region modeling of [11C]-(R)-PK11195 brain studies. *J Nucl Med* 2007, 48(1):158-167.
- 7) Ikoma Y, Edison P, Ramlackhansingh A, Brooks DJ, Turkheimer FE: Reference region automatic extraction in dynamic [(11)C]PIB. *J Cereb Blood Flow Metab* 2013, 33(11):1725-1731. DOI <http://dx.doi.org/10.1038/jcbfm.2013.133>

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