

Standard Operating Procedure for Manual Segmentation of the Spinal Cord and Gray Matter within the Lumbosacral Cord

Abbreviations

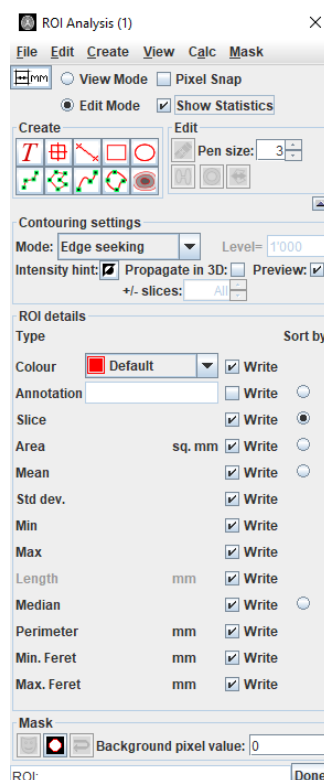
GM, gray matter; SC, spinal cord; WM, white matter

Purpose

The standard operating procedure was established for manually segmenting the SC and GM in multi-echo gradient echo (ME-GRE) images of the lumbosacral cord acquired on a 3T Siemens Prisma scanner.

Before segmentation

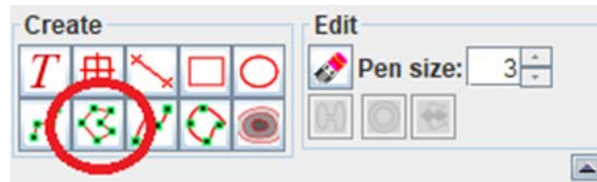
- Open the image using JIM 7 (Xinapse Systems, <https://www.xinapse.com>)
- Apply interpolation by ticking View → Interpolated
- Launch the ROI Analysis tool: Go to Toolkits → ROI Analysis
- Within the ROI Analysis window, make sure to:
 - enable extended statistics by ticking View → Show Extended Stats
 - click to set the measurement units to mm (upper left corner)
 - tick "Show Statistics"
 - select the options displayed below:



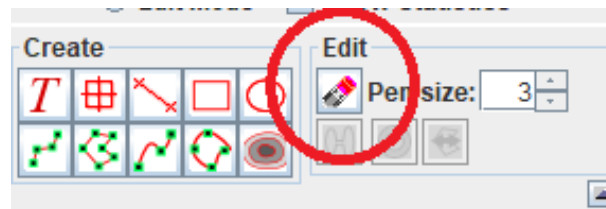
- Display all slices to assess image quality for potential motion or other artifacts. Suggested zoom factor: 700 - 800%.
- Select the initial slice for segmentation. Choose a high-quality slice that can be segmented reliably. Begin with segmenting the SC followed by the GM.

Segmentation

- Create a new Irregular ROI within the ROI Analysis window:



- Use the Erase points tool to make edits to segmentations or erase segmentations.



- Segment slices consecutively. Recommended zoom factor: 1200 - 1300%. Adjust the intensity window for each slice using the sliders on the right, following these guidelines:
 - *for SC segmentation:*
lower threshold: increase to the point where WM begins to saturate (turning black)
upper threshold: decrease to the point where CSF begins to saturate (turning white)
 - *for GM segmentation:*
lower threshold: same as for SC segmentation
upper threshold: decrease to the point where GM begins to saturate (turning white)
- **Follow the segmentation guidelines provided in the following chapters.**
- Remember to save your progress regularly during segmentation in the ROI Analysis window by selecting File -> Save ROIs As
- At the end, confirm the segmentation by displaying three adjacent slices simultaneously (Layout: 3) and scrolling through them. Additionally, plot the curves of the slice-wise SC and GM cross-sectional areas. Pay close attention to smooth transitions from one slice to the next. Adjust segmentation in case of abrupt changes between adjacent slices.

Segmentation guidelines: spinal cord

Maintaining a consistent segmentation along the spinal cord boundaries

Issue:

It is important to draw the segmentation line along voxels with the same intensity, ensuring that the segmentation follows an isointense contour. However, this needs to be balanced with maintaining the anatomical smoothness of the SC.

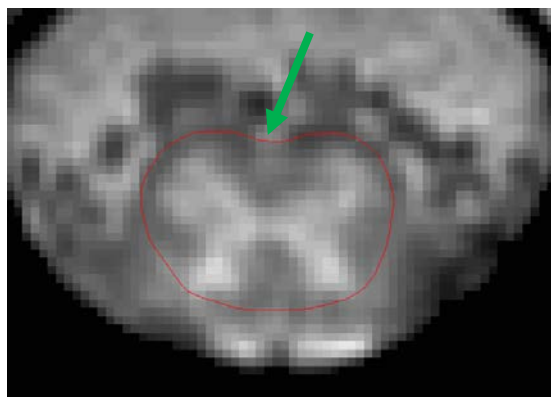
Guidelines:

Avoid creating a zigzag segmentation line by following the isointense contour too closely. Instead, aim for a smooth segmentation line, aligning with the anatomically smooth boundary of the SC.

Segmenting the ventral boundary of the spinal cord

Issue:

Anatomically, there exists a fissure on the medio-ventral part of the SC known as the anterior medial fissure. While the fissure itself may not be directly visible, the entry point of the fissure forms an anatomical indentation and is discernible on the MRI images (indicated by the green arrow in the figure below). Additionally, a major blood vessel is situated slightly ventral to the fissure.



Guidelines:

- Ensure that the major blood vessel situated ventral to the SC is not included in the segmentation.
- Follow any visible anatomical indentations, but if none are present, do not force them into the segmentation.

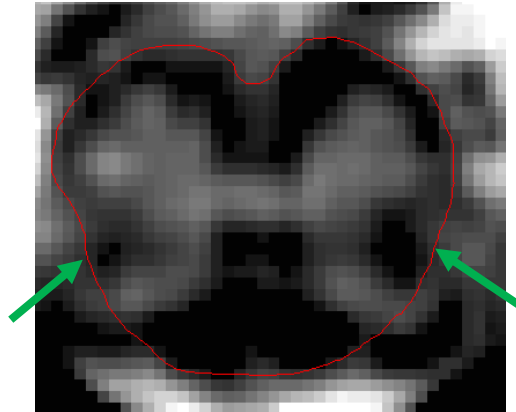
Segmenting the lateral boundary of the spinal cord

Issue:

The lateral boundaries of the SC are sometimes not clearly visible, as the region just outside the SC has similar intensities (as indicated by the green arrows below).

Guidelines:

Draw a relatively straight line extending towards the tip of the dorsal GM horn as illustrated below. If necessary, small "dents" are acceptable, but refrain from creating large dents in the segmentation.



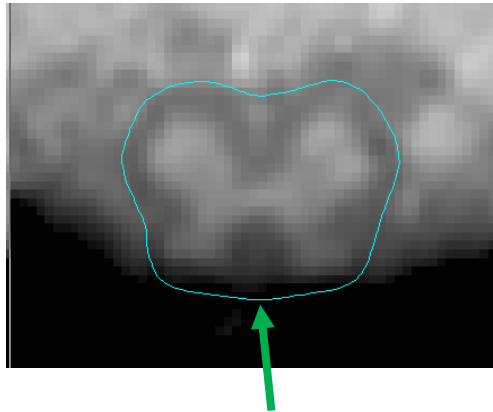
Segmenting the dorsal boundary of the spinal cord

Issue:

Hypointense regions caused for example by susceptibility artifacts can compromise SC segmentation along the dorsal boundaries of the SC.

Guidelines:

Maintain a roundish shape of the SC in the hypointense region. Looking at the neighboring slices can be helpful, as often they are not affected by these artifacts. Note down the affected slices.



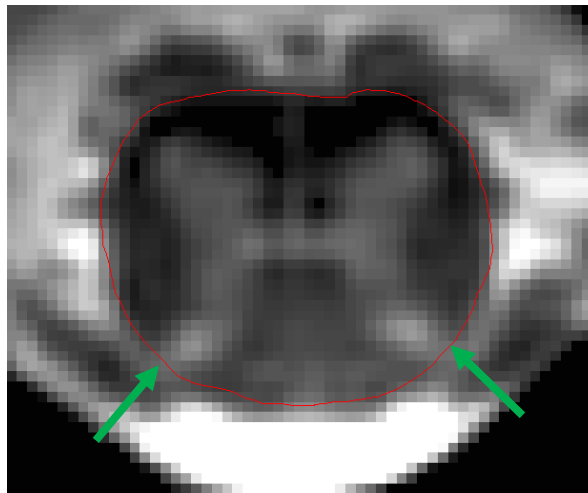
Segmenting at the tip of the dorsal gray matter horn

Issue:

Segmenting the SC can be less straightforward at the tip of the dorsal GM horns, as they may appear to extend beyond the boundaries of the SC (as indicated by the green arrows below).

Guidelines:

Maintain the roundish shape of the SC and draw the segmentation line as illustrated below.



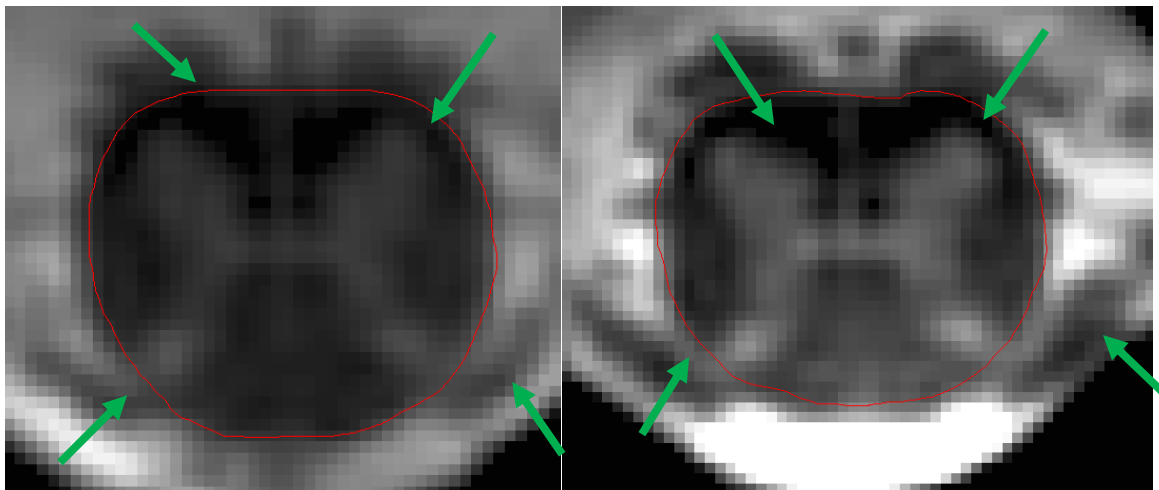
Separating nerve rootlets from the spinal cord

Issue:

Nerve rootlets, roots, and nerves exhibit a hypointense signal similar to that of the SC, which makes it difficult to distinguish them from the SC itself (indicated by the green arrows in the figures below). This issue is particularly notable in the lumbosacral cord, where spinal nerves are located in close proximity to the SC. The rootlets enter/exit both the dorsal and ventral SC, but in our experience, the issue is more pronounced in the ventral boundaries of the SC.

Guidelines:

Exclude rootlets from the segmentation. Identify the anatomical boundaries between the rootlets and the SC. If these boundaries aren't immediately visible, aim to preserve the roundish shape of the SC, as illustrated in the figures below. When in doubt, looking at neighboring slices can be helpful; often, a boundary becomes evident in the slice above or below, as illustrated in the example below (the boundary is not visible in the left slice, but is more discernible in the right slice).



Segmentation guidelines: gray matter

Maintaining a consistent segmentation along the gray matter boundaries

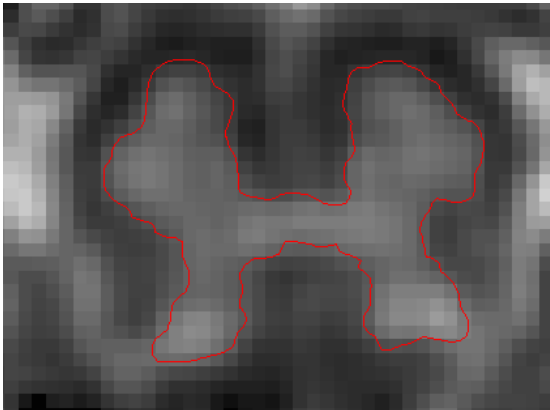
Issue:

It is important to draw the segmentation line along voxels with the same intensity, ensuring that the segmentation follows an isointense contour. However, this needs to be balanced with maintaining the anatomical smoothness of the GM.

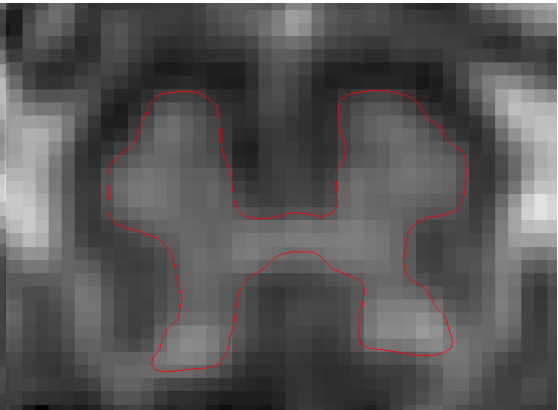
Guidelines:

Avoid creating a zigzag segmentation line by following the isointense contour too closely. Instead, aim for a smooth segmentation line, aligning with the anatomically smooth boundary of the GM (as shown in the right figure below).

Zigzag pattern (not recommended)



Smooth pattern (recommended)



Conservative vs. liberal segmentation

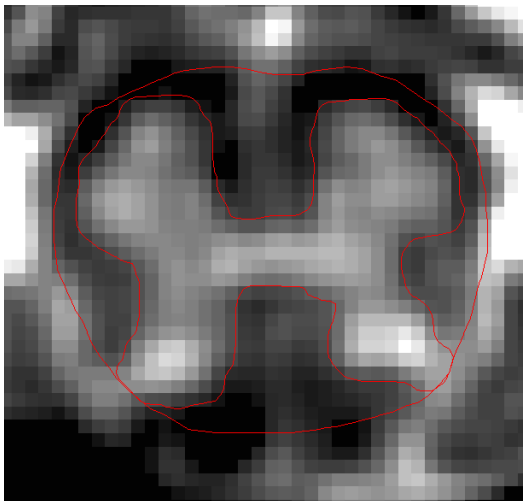
Issue:

As previously mentioned, it is important for the segmentation line to follow an isointense contour. The next consideration is: which intensity level to select? In these images, GM exhibits high intensity (I_{GM}) and WM shows low intensity (I_{WM}). Therefore, the segmentation line should fall within the intensity range between I_{GM} and I_{WM} . This intermediate intensity region is called the partial volumes. It's worth noting that the width of the partial volumes increases with larger voxel sizes. If the segmentation line follows an intensity which is closer to I_{GM} , it is referred to as conservative segmentation, as it excludes much of the partial volumes and encompasses mostly voxels located entirely within the GM. Conversely, if the segmentation line follows an intensity which is closer to I_{WM} , it is referred to as liberal segmentation, as it encompasses a significant portion of the partial volumes.

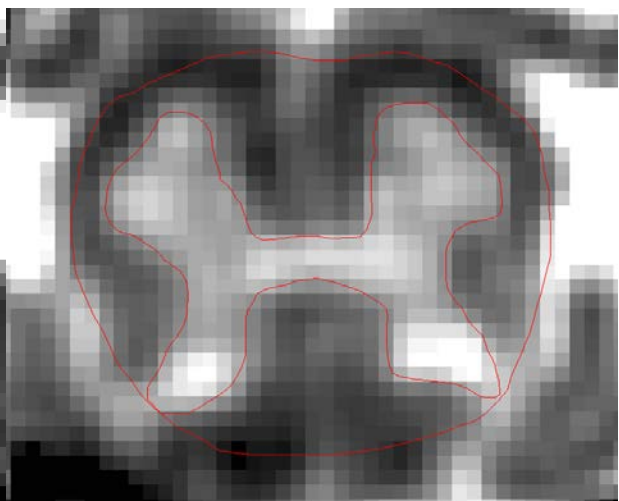
Guidelines:

The segmentation line should be in the middle of the partial volumes; therefore, it should follow the isointense contour $(I_{GM} + I_{WM})/2$. See images below for an example.

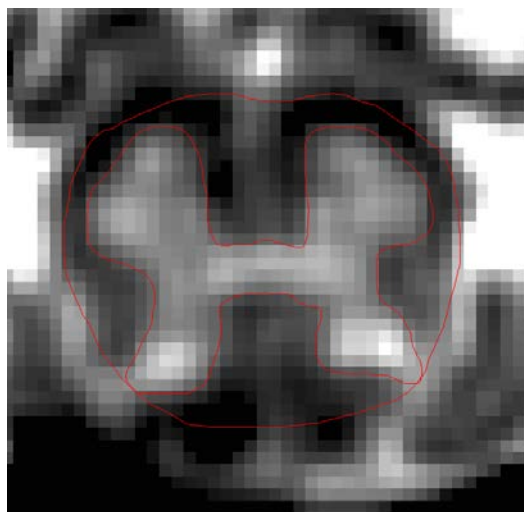
Liberal segmentation



Conservative segmentation



Recommended segmentation



Segmenting the tip of the dorsal gray matter horn

Issue:

Establishing a boundary between the dorsal GM horn and the dorsal rootlet is not straightforward.

Guidelines:

In slices where the characteristic butterfly-shaped GM is evident, ensure that the GM segmentation line touches the SC segmentation line, without extending beyond it (as indicated by the green arrows in the figure below). In caudal slices where the characteristic butterfly-shaped GM is not evident any more, the GM segmentation is not required to touch the SC segmentation line.

