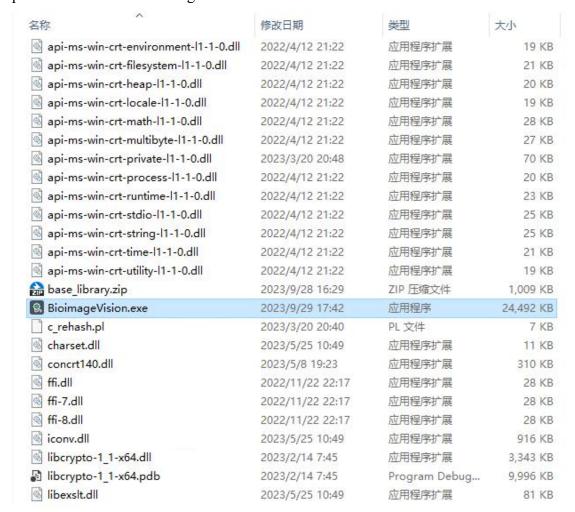
The user guider

1. Summary

BioimageVision software can generate the pyramid data from an image sequence and visualize the pyramid data. For any feedback on this software, please contact the authors: Tingwei Quan, quantingwei@mail.hust.edu.cn; Xuzhong Qu, quantingwei@mail.hust.edu.cn; Xuzhong Qu, quantingwei@mail.hust.edu.cn; Xuzhong Qu,

2. Install

BioimageVision Currently only supports Windows; after unpacking the software, please double-click BioimageVision.exe.

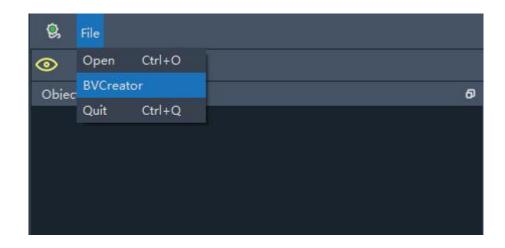


3. Quick Start

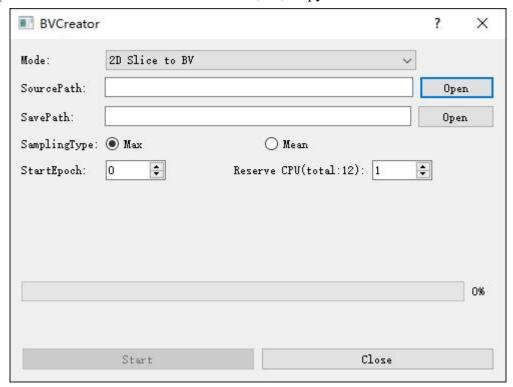
3.1 Pyramid data transformation

Step 1: Click in the toolbar for browsing mode, and then click BVCreator in

the menu bar.



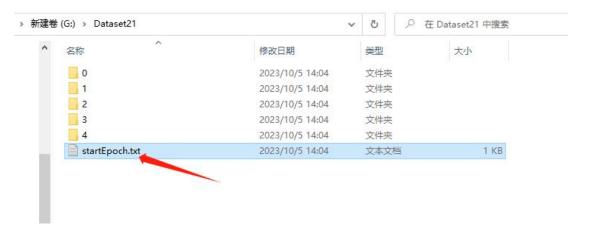
Step 2: Click open to select SourcePath where an image sequence is stored; Click 'Open' to select SavePath for save BV data,i.e., an pyramid data.



Note:

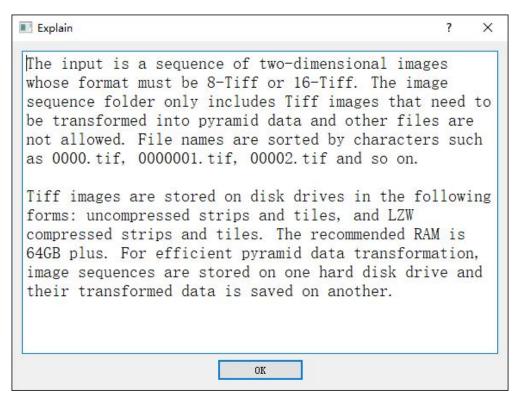
- 1. All paths cannot contain Chinese characters and Spaces.
- 2. Our software supports continue running where the program left off accidentally. In this case, the user should check the savepath and find a file named 'startEpoch.txt', then find number found in this file and fill in the number in

Normally, the program runs smoothly and number of 'StartEpoch' is a default value (0).

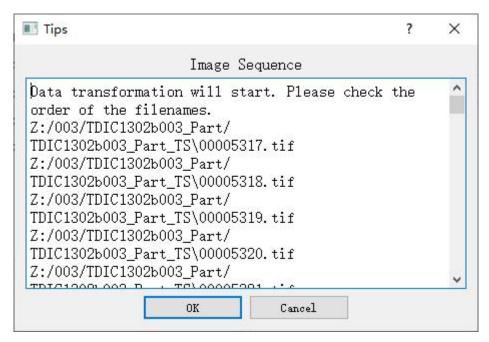


3. If a user wants to run the software with full power, he/she can fill in '0' in 'Reserve CPU'. However, we strongly suggest to fill in '1' instead for preventing system lag of the computer. Also, the software provides the total number of CPU cores in the running computer (i.e., there are 12 cores in the computer shown in the above figure).

Step 3. Select the SamplingType to generate multi-resolution chunks in the pyramid data, which includes max type and mean type. After clicking 'Start', the corresponding prompt box will appear.



Step 4: After clicking 'OK', the prompt below will appear.



Step 5 After clicking 'OK' in the figure above, the software begins to transform two-dimensional image sequence into the BV data set. The form of BV data storage is as following.

名称 个	修改日期	类型	大小
0	2023/5/4 20:14	文件夹	
1	2023/6/2 0:22	文件夹	
2	2023/6/2 2:10	文件夹	
3	2023/5/4 21:30	文件夹	
4	2023/5/4 21:30	文件夹	
5	2023/5/4 21:30	文件夹	
6	2023/5/4 21:30	文件夹	
7	2023/6/1 20:14	文件夹	
config.cfg	2023/6/6 15:43	CFG 文件	1 KB

3.2 Data visualization module

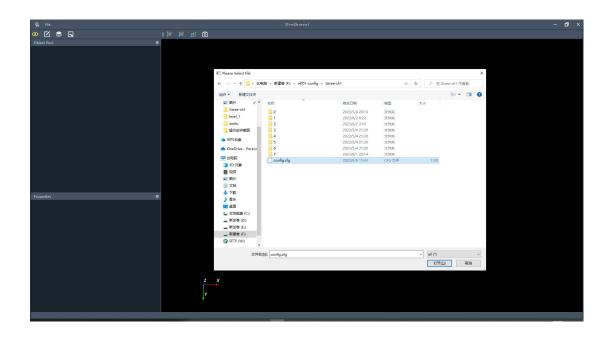
In the data visualization module, BioimageVision provides the following three display modes: the visualization of large-scale data (section 3.2.1), the visualization of local data (section 3.2.2) and the visualization of SWC data (section 3.2.3).

3.2.1 Visualization of BV data

Step 1 Click in the toolbar for browsing mode; Click 'File→Open' in the menu bar;



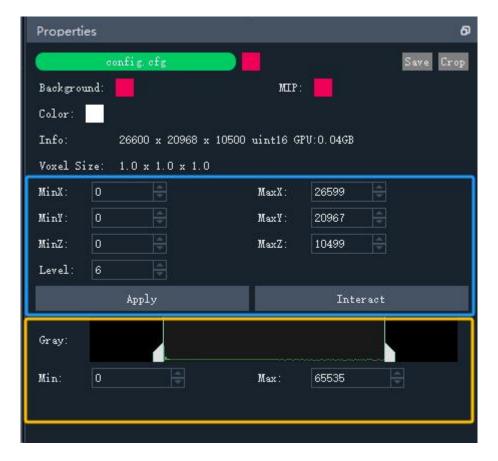
Step 2 Import data in BV data.



Step 3. The imported datasets are listed the menu bar: Object Pool; for visualizing the specified data, user should click refers to hiding the data, and refers to remove the data from Object Pool list.



Step 4. Set visualization properties of BV data.



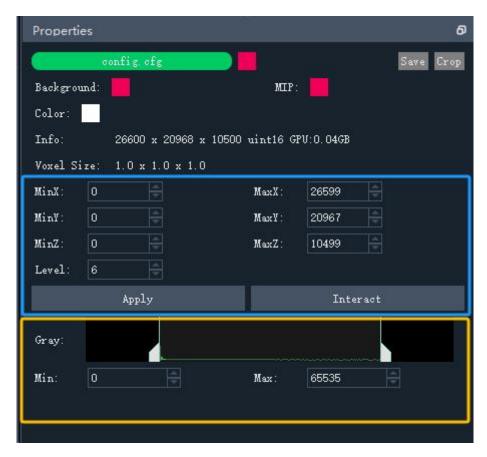
Note:

- 1. Click on the 'red box' of hidden.
- 2. Click on the 'red box' of Background: ____, only the region of interest is displayed;

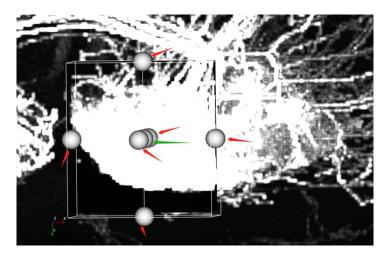
- 3. Click on the 'red box' of projection.
- 4. Click on the 'white box' of Color: , adjust the color of the displayed data.
- 5. Info shows the total size, format, and the size of the video memory occupied by the displayed data.
- 6. Voxel Size indicates the resolution of the image. Click 'Crop' in the upper right corner of the window. The voxel size can be adjusted by setting the suitable parameters as shown below.



Step 5. Select region of interest (ROI) in BV data. There are two ways for this purpose. One is to input the parameters highlighted with the blue box, and then click the button 'Apply'.



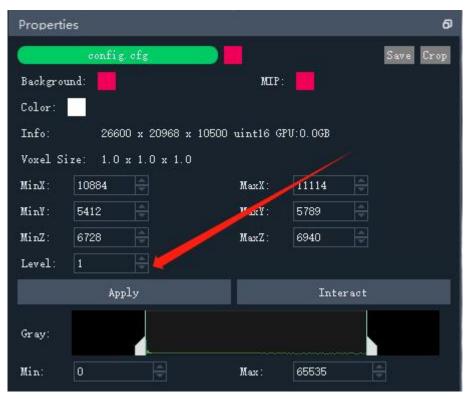
The other is directly click the button 'Interact'. After clicking this button, a selected ROI will shown with white balls. In addition, the user can adjust the gray value of displayed ROI freely by changing minimum/maximum gray value in the yellow box in the figure above.

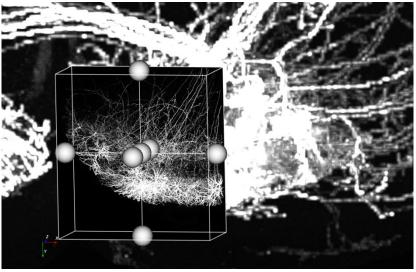


Note:

- 1. After selecting region of interest in BV data, it can be labeled with a rectangular box, shown in the above figure.
- 2. There are small balls located in the center of cuboid face as red arrows labeled.

- After single click the ball with left mouse, hold down the left mouse and drag the ball, the size of interesting region can be adjusted.
- 3. Single click the ball in the center of the rectangular box as green arrows labeled, hold down the left mouse, and drag the ball to a specified position. Correspondingly, the position of the region of interest is changed. Using the button 'Apply' to fix the region of interest.
- 4. Enter the Level value (default is 0-6) to adjust the display resolution of the image. Please see blow with red arrows.





5. When the size of ROI is large and its display resolution is high, the video memory will exceed limits.



Step 6. After adjusting the size and resolution of the region of interest, click the 'Save' button in the upper right corner to save the region of interest.



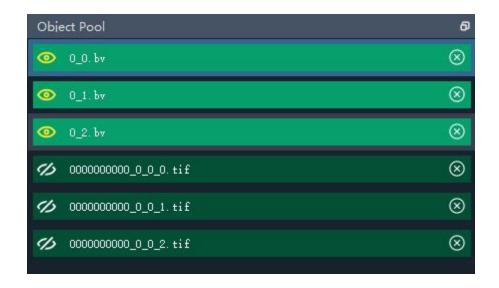
3.2.2 Visualization of volume images

The current version of BioimageVision is suitable for visualization of Tiff volume images and the chunks in the BV pyramid data. Here, we show an example of how to visualize the tiff image.

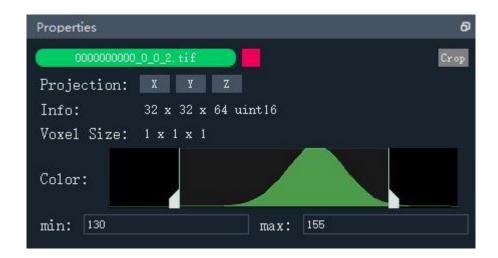
Step 1: Import the chunks in BV pyramid data: click in the toolbar to perform browsing mode and then click 'File→Open' in the menu bar.



Step 2. Select the images that needs to be displayed. After chunks are imported, the imported chunks (0_0.bv, 0_1.bv, etc.) are listed in Object Pool panel.



Step 3. Select the visualization properties of volume images. In the Object Pool panel, click the bar with left mouse button, the flowing window will appear. In this window, we can set the range of gray values for image visualization.



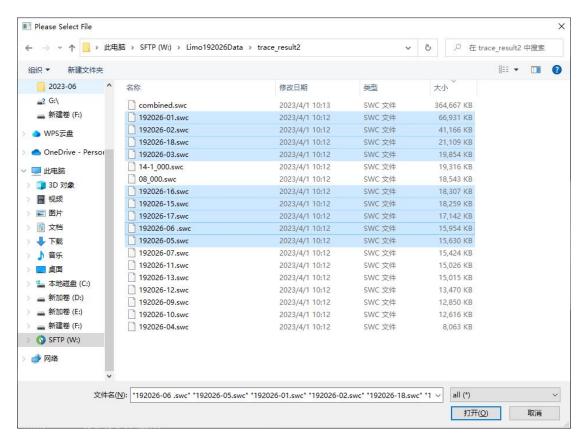
3.2.3 Visualization of SWC file

SWC file is widely used to store the reconstructed neurons which consists of a series of points. Each of points includes its position, its radius, the connection information, and its category. BioimageVision can visualize the SWC file, namely, the reconstructed neurons.

Step 1. Click in the toolbar to perform browsing mode; Click 'File→Open' in the menu bar.



Step 2 Import swc format data. BioimageVision supports importing multiple swc files at the same time.



Step 3. Select SWC files that need to be displayed. In the object pool panel, the imported SWC files are listed. The indicates whether the data is visualized or not. Click the square icon (shown below) for adjusting the display color of the data.

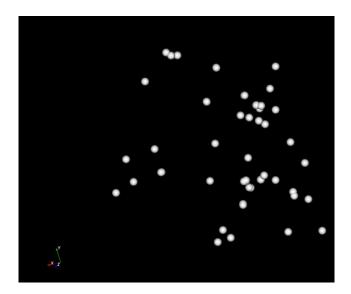
Click for removing the data from the panel.



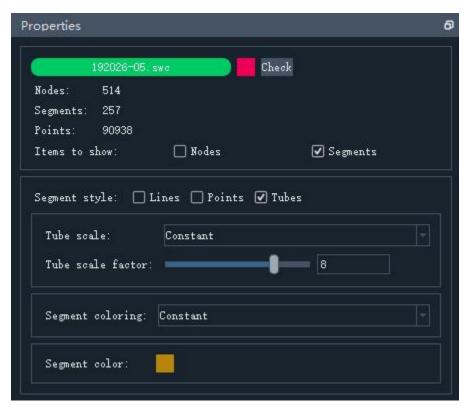
Step 4. Set the visualization parameters. Click the rectangular strip in the Object Pool panel, as shown in the above figure, and then the corresponding file information can be displayed below.

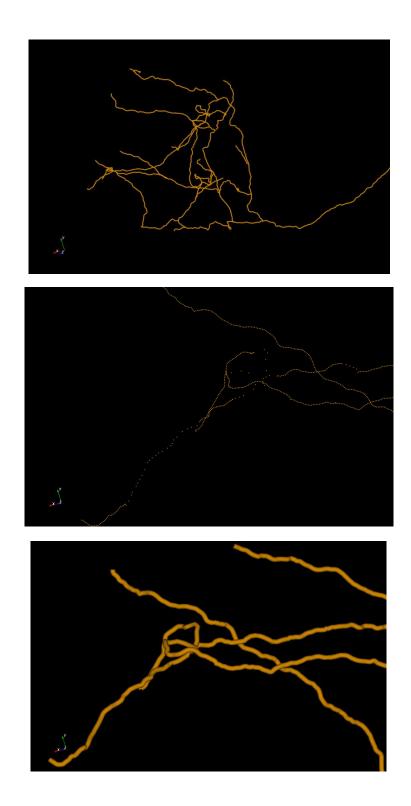


For example, when the SWC file (192026-05.swc) is selected, the number of nodes, segments and points are counted. The node size and color can be adjusted in the figure below.



In addition, the swc file can be displayed in segments mode. The segments of the swc file can be displayed in three ways: lines, points and tubes, shown in the figures below.





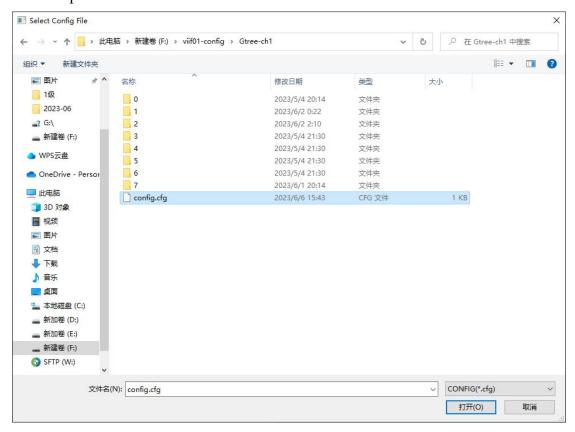
4. Application of BV data

Like other pyramid data structures, BV data format achieves multi-scale visualization of volume images. Moreover, BV can provide more visualization functions and data operations. From BV data structure, we can easily and quickly visualize specific slices

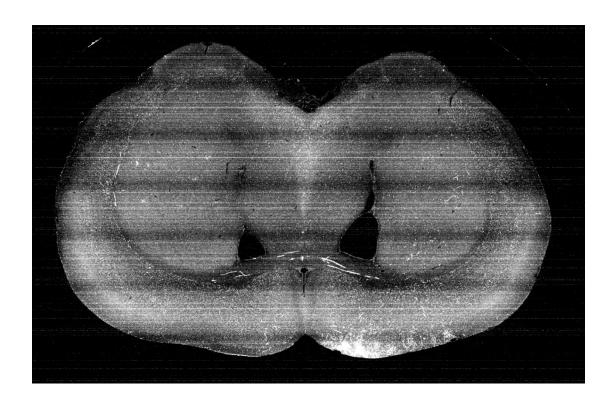
from the whole volume image, a series of small volume images around the skeletons. From BV data structure, it is convenient to generate the maximum projection of slices and visualize them.

4.1 Visualization of slices from the whole volume data

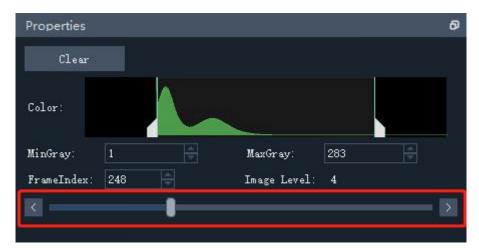
Step 1. Click ' in the toolbar to browse slices; Click 'File→Open' in the menu bar to import BV data format.



Step 2. Visualize the slices. After the BV data is successfully imported, its slice is automatically displayed, shown as below. The data slices can be visualized through sliding the mouse.



Step 3. Set data slice visualization parameters. The following panel controls the data slice visualization. The intensity distribution of data can be manually adjusted for displaying the weak signals or hiding the noise signal in the data visualization. Frame index refers to the identification number of the current displayed slice. Image Level is the tiers of pyramid data. As shown in the figure below, number 4 means that the displaying slice is generated by downsampling the data four times. The slices are also visualized automatically in the order of Frameindex, as red rectangular labeled.



4.2 Maximum projection of the volume image

For observing the overall information of large-scale volume data, maximum

projection operation is widely used. In general, the 3D volume image is divided into a series of image blocks along the z-axis. Each image block is mapped into a two-dimensional image by taking the maximum value along the z direction for the same x and y coordinates.

Step 1. Click on the toolbar to enter the maximum projection mode; Click the menu bar 'File—ProjectionCreator' for maximum projection.

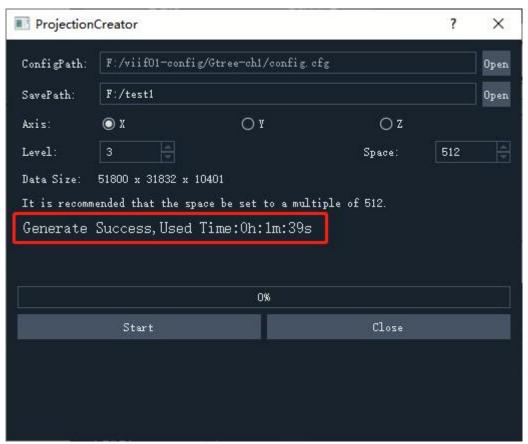


Step 2. Import BV data and set the save path for maximum projection of images. The following panel also provides the key maximum projection direction. Axis refers to the maximum projection direction. Level refers to the tier of BV pyramid data. A level value of 0 represents the original volume images. Space refers to the length of the projection interval.

Note: Level is generally set to a range of 0 to 6; it is recommended that the projection interval in X and Y direction be a multiple of 512, and the projection interval in Z direction be a multiple of 16;



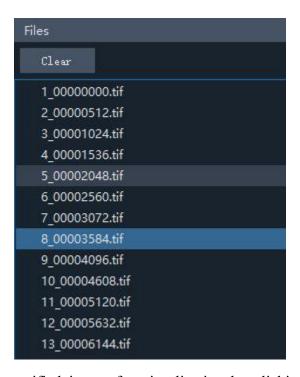
Step 3. Run the maximum projection operation by clicking start button. If successful, there will be a statement indicator as labeled by a red rectangular box. In this case, please click 'Close' button.



4.3 Visualization of maximum projection images

As described above, when the maximum projection operation is performed on a large-scale volume image, a series of maximum projection images with Tiff format are generated. The number of maximum projection images is determined by the length of projection interval. For example, for volume image with the z-dimension of 1000 and the set of projection interval length 100, the 10 maximum projection images will be generated.

Step 1. Import the maximum projection data by clicking 'File→Open' in the menu bar and selecting the file path. All images will be loaded in the BioimageVision.



Step 2. Select the specified image for visualization by clicking its file name. Its visualization setting is in the following window. The grayscale values of the image can be adjusted in the grayscale histogram. Pos is the coordinate where the current mouse is in the visualization area.



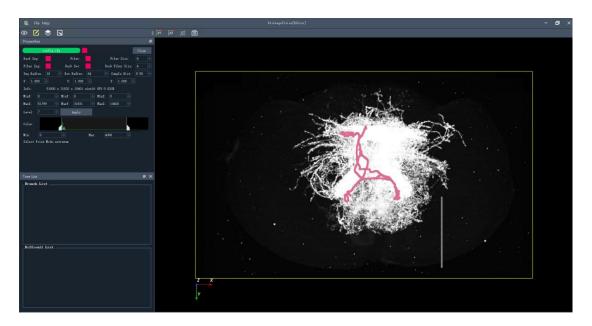
4.4 visualization of a series of small blocks surrounding the traced neurites

This function serves as an example of multiscale visualization of the quantified results and their corresponding images at the same time. To check the quantified results, it is helpful to display the image signal around them. This kind of visualization can significantly the mutual interference of signal and enhance the efficiency in the checking quantified results.

Step 1. Click ' in the toolbar to display the reconstructed neurites and their corresponding images. The following window contains three file paths. In Config Path, the BV images are stored. In SWC path, the reconstructed skeletons of neurons are stored as SWC file. SwcConfig refers to the pyramid SWC files, which can store multi-neuron reconstructed skeletons and can quickly retrieve the specified skeletons.



Step 2. Set the visualization parameters. If the data is successfully loaded, the corresponding information will appear in the panel labeled with a rectangular box.



Step 3. Select a neurite that is required to be visualized. Click red square of to hide images, and then click the specified neurite in the visualization panel. The clicked neurite is highlighted with red color.

Step 4. Visualize the reconstructed skeletons and its corresponding images, which is achieved by simultaneously pressing the Ctrl key and clicking the left mouse button. This kind of visualization will be presented continually along the reconstructed neurite by clicking null key.

