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Smart spatial omics (S2-omics) optimizes region-of-interest selection to capture molecular heterogeneity in diverse tissues V.3

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S2-omics



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Manuscript citation:

This protocol is associated with our paper describing S2-omics. Paper link:
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ReadTheDocs page: <https://s2omics.readthedocs.io/en/latest/>

Github page: <https://github.com/ddb-qiwang/S2Omics>

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Protocol status: Working

We use this protocol and it's working

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Keywords: spatial molecular profiles across entire tissue section, resulting spatial omics data, using histology image, colorectal cancer tissue section, spatial omics data, smart spatial omic, spatial molecular profile, histology image, spatial omics experiment, making spatial omics study, spatial omics study, molecular heterogeneity in diverse tissue, preserving critical spatial molecular variation, diverse tissue, critical spatial molecular variation, entire tissue section, omic, selected rois

Abstract

A protocol describing the application of S2-omics on a colorectal cancer tissue section, designing 10x VisiumHD experiment.

S2-omics is an end-to-end workflow that automatically selects regions of interest for spatial omics experiments using histology images. Additionally, S2-omics utilizes the resulting spatial omics data to virtually reconstruct spatial molecular profiles across entire tissue sections, providing valuable insights to guide subsequent experimental steps. Our histology image-guided design significantly reduces experimental costs while preserving critical spatial molecular variations, thereby making spatial omics studies more accessible and cost-effective.

We demonstrate S2-omics's utility in selecting a 6.5mm × 6.5mm ROI and predicting the cell-type labels for the whole slide image after conducting VisiumHD experiment on the selected ROI.

Materials

he-raw.jpg: Raw histology image.

pixel-size-raw.txt: Side length (in micrometers) of pixels in he-raw.jpg. This value is usually between 0.1 and 1.0. For an instance, if the resolution of raw H6E image is 0.2 microns/pixel, you can just create a txt file and write down the value '0.2'.

annotation_file.csv(optional): The annotation and spatial location of superpixels, should at least contain three columns: 'super_pixel_x', 'super_pixel_y', 'annotation'. This file is not needed for ROI selection. For an instance, the first row of this table means the cell type of 267th row (top-down) 1254th column (left-right) superpixel is Myofibroblast.

Troubleshooting

Before start

Install Python. We recommend using conda, miniconda, or mamba for this.

To run the demo, first please download the demo data and pretrained model checkpoints file from:

google drive: https://drive.google.com/drive/folders/1z1nk0sF_e25LKMHyJxJVMtROFjuWet2G?usp=sharing

Please place both 'checkpoints' and 'demo' folder under the 'S2Omics' main folder.

In this demo, we mimic the situation that we need to select a 6.5 mm*6.5 mm ROI for Visium HD experiment from a colorectal cancer tissue section. To run the ROI selection (takes about 25 minutes with GPU),



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- 3 google drive:https://drive.google.com/drive/folders/1z1nk0sF_e25LKMHyJxJVMtROFjuWet2G?usp=sharing
- 4 Please place both 'checkpoints' and 'demo' folder under the 'S2Omics' main folder.
- 5 We provide both python files and notebooks for all examples. User can download them from S2-omics's github repository <https://github.com/ddb-qiwang/S2Omics>

Data format

- 6 Here are the files needed for running S2-omics. User can further refer to the demo data to check the detailed data formats.
- 7 **he-raw.jpg**: Raw histology image.
- 8 **pixel-size-raw.txt**: Side length (in micrometers) of pixels in he-raw.jpg. This value is usually between 0.1 and 1.0. For an instance, if the resolution of raw H&E image is 0.2 microns/pixel, you can just create a txt file and write down the value '0.2'.
- 9 **annotation_file.csv(optional)**: The annotation and spatial location of superpixels, should at least contain three columns: 'super_pixel_x', 'super_pixel_y', 'annotation'. This file is not needed for ROI selection. User can refer to the demo for more detailed input information.

ROI selection for single slide

- 10 For example, to select ROI on the demo colorectal cancer section:



```
11 python run_roi_selection_single.py --prefix
    './demo/Tutorial_1_VisiumHD_ROI_selection_colon/' --save_folder
    './demo/Tutorial_1_VisiumHD_ROI_selection_colon/S2Omics_output' --device 'cuda:0' --
    roi_size 6.5 6.5 --num_roi 1
```

ROI selection for multiple slides

12 To select ROI on the demo consecutive breast cancer sections:

```
13 python run_roi_selection_multiple.py --prefix_list
    './demo/Tutorial_3_Consecutive_ROI_selection_breast/breast_cancer_g1/'
    './demo/Tutorial_3_Consecutive_ROI_selection_breast/breast_cancer_g2/'
    './demo/Tutorial_3_Consecutive_ROI_selection_breast/breast_cancer_g3/' --
    save_folder_list
    './demo/Tutorial_3_Consecutive_ROI_selection_breast/breast_cancer_g1/S2Omics_output'
    './demo/Tutorial_3_Consecutive_ROI_selection_breast/breast_cancer_g2/S2Omics_output
    '
    './demo/Tutorial_3_Consecutive_ROI_selection_breast/breast_cancer_g3/S2Omics_output
    ' --device 'cuda:0' --roi_size 1.5 1.5 --num_roi 1
```

Cell type broadcasting

14 To broadcast the cell type label within the selected ROI to the entire slide on the demo colorectal cancer section:

```
15 python run_label_broadcasting.py --WSI_datapath
    './demo/Tutorial_1_VisiumHD_ROI_selection_colon/' --SO_datapath
    './demo/Tutorial_1_VisiumHD_ROI_selection_colon/' --WSI_save_folder
    './demo/Tutorial_1_VisiumHD_ROI_selection_colon/S2Omics_output' --SO_save_folder
    './demo/Tutorial_1_VisiumHD_ROI_selection_colon/S2Omics_output' --need_preprocess
    True --need_feature_extraction True
```

License

16 For commercial use of open source software S2-omics, please contact Musu Yuan (musu990519@gmail.com) and Mingyao Li (mingyao@penntestmed.upenn.edu).