Supplementary Material

TDat: An efficient platform for processing petabyte-scale whole-brain volumetric images

Yuxin Li^{1,2}, Hui Gong^{1,2}, Xiaoquan Yang^{1,2}, Jing Yuan^{1,2}, Tao Jiang^{1,2}, Xiangning Li^{1,2}, Qingtao Sun^{1,2}, Dan Zhu^{1,2}, Zhenyu Wang^{1,2}, Qingming Luo^{1,2}, Anan Li^{1,2*}

¹Collaborative Innovation Center for Biomedical Engineering, Wuhan National Laboratory for Optoelectronics-Huazhong University of Science and Technology, Wuhan, Hubei 430074, China.

²Britton Chance Center and MOE Key Laboratory for Biomedical Photonics, School of Engineering Sciences, Huazhong University of Science and Technology, Wuhan, Hubei 430074, China.

*Correspondence: Anan Li (aali@mail.hust.edu.cn).

1 Supplementary Figures and Tables

1.1 Supplementary Figures



Supplementary Figure 1 | Comparison of the results between the TDat rotation and the rotation of the entire dataset.

An example of a 3DCM model data of size $512 \times 512 \times 512$ px was rotated 45 degrees around the zaxis using two methods. Two slices with the same position in the transformed data were shown for comparison. Four regions were enlarged to show the detailed pixel value. The pink dotted line represents the borders of different 3D blocks in the TDat file.



Supplementary Figure 2 | 3DCM model dataset.

(A) 3D visualization of a model dataset; the data size is $1,280 \times 1,280 \times 1,280$ px; the size of each slice is $1,280 \times 1,280$ px with an 8-bit TIFF. The size of each black or white block is $256 \times 256 \times 256$ px. (B) One slice in the 3DCM model dataset, and enlarged views of the red region. The Gaussian noise is shown. (C) The histogram of a slice in the model dataset, which reveals the 5 % Gaussian noise.



Supplementary Figure 3 | TDat-Fiji plugin for accessing TDat dataset in Fiji.

The TDat-Fiji plugin was used to read the ROI from the TDat dataset into the Fiji image stack, and Fiji was used to perform maximum intensity projections, image binary and cell counting. The ROI is a Thy1-EGFP F-line transgenic mouse hippocampus. The dataset was imaged using a light sheet microscope (UltraMicroscope II, Lavision Biotec GmbH) (see Dataset1 in **Table 2**).



Supplementary Figure 4 | TDat-Vaa3D plugin for accessing a TDat dataset in Vaa3D.

The TDat-Vaa3D plugin was used to read the ROI from the TDat dataset into Vaa3D and edit the color map for visualization; paint-and-trace was used to trace neurons in 3D space. The ROI is an SD rat cerebellum. The dataset was imaged using a MOST (see Dataset4 in **Table 2**).

1.2 Supplementary Tables

Image data	File size	Pixels	Bit depth	Noise	Reformatted size
100 GB	62.1 GB	4,753 × 4,753 × 4,753	8 bits	5 %	64.5 GB
200 GB	122 GB	5,989 × 5,989 × 5,989	8 bits	5 %	129 GB
400 GB	241 GB	$7{,}545 \times 7{,}545 \times 7{,}545$	8 bits	5 %	258 GB
800 GB	480 GB	$9{,}506 \times 9{,}506 \times 9{,}506$	8 bits	5 %	514 GB
1,600 GB	967 GB	11,977 × 11,977 × 11,977	8 bits	5 %	1 TB
3,200 GB	1.87 TB	15,090 × 15,090 × 15,090	8 bits	5 %	2.01 TB
6,400 GB	3.73 TB	19,012 × 19,012 × 19,012	8 bits	5 %	4.03 TB
12.5 TB	7.47 TB	23,954 × 23,954 × 23,954	8 bits	5 %	8.19 TB
25 TB	14.9 TB	30,180 × 30,180 × 30,180	8 bits	5 %	16.1 TB
50 TB	29.8 TB	38,024 × 38,024 × 38,024	8 bits	5 %	32.7 TB
100 TB	59.5 TB	47,908 × 47,908 × 47,908	8 bits	5 %	65.5 TB
1 PB	607 TB	104,032 ×104,032 ×104,032	8 bits	5 %	629 TB

Supplementary Table 1 Detail information of model dataset in this study reported here.

Image data	Amira-XLVolume		Fiji-bigdataviewer		TDat		Vaa3D-terafly	
	Run time	RAM usage	Run time	RAM usage	Run time	RAM usage	Run time	RAM usage
100 GB	6.6 h	0.8 GB	5 h	16 GB (a)	0.36 h	1.4 GB	1.3 h	1.4 GB
200 GB	25 h	0.8 GB	10.2 h	16 GB (a)	0.91 h	1.6 GB	2.5 h	2.2 GB
400 GB	N/A	N/A	21.3 h	16 GB ^(a)	1.96 h	2 GB	5 h	3.6 GB
800 GB	N/A	N/A	N/A	N/A	4.06 h	2.5 GB	10.5 h	11.5 GB
1,600 GB	N/A	N/A	N/A	N/A	7.7 h	3.2 GB	21.1 h	18.3 GB
3,200 GB	N/A	N/A	N/A	N/A	12.7 h	4 GB	N/A	29.1 GB ^(b)
6,400 GB	N/A	N/A	N/A	N/A	24.8 h	5 GB	N/A	92 GB ^(b)

Supplementary Table 2 Run time and memory usage for reformatting different size data between different methods.

All benchmarks were repeated three times for each experiment, and they were run on a graphic workstation with 3.1GHz Intel E5-2687w×2 (16 cores), 192 GB of RAM, a disk array.

^(a) The maximum heap size of Fiji was set to 16 GB.

^(b) the value was given by Estimated RAM usage in Vaa3D-terafly.

N/A: We did not test because the computation time was excessive.

	Workstation	HPC
100 GB	0.36 h	0.05 h
200 GB	0.91 h	0.07 h
400 GB	1.96 h	0.10 h
800 GB	4.06 h	0.17 h
1,600 GB	7.7 h	0.30 h
3,200 GB	12.7 h	0.55 h
6,400 GB	24.8 h	1.01 h
12.5 TB	N/A	1.97 h
25 TB	N/A	3.65 h
50 TB	N/A	7.15 h
100 TB	N/A	13.6 h
1 PB	N/A	144 h

Supplementary Table 3 Run time for reformatting different size data on workstation and HPC.

All benchmarks were repeated three times for each experiment, the computers configuration are provided in **Table 3**.

N/A: We did not test because the computation time was excessive or the capacity of storage was limited.