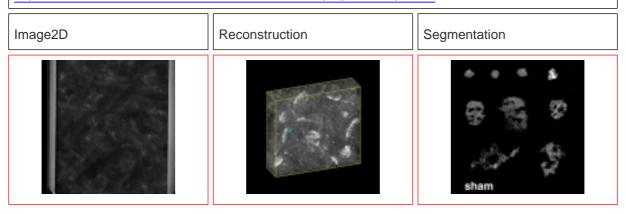
### **Cell Centered Database**

## University of California, San Diego Maryann Martone

### Microscopy Product #:21 shamca1

For the most updated information, please visit

http://ccdb.ucsd.edu/CCDBWebSite/main?event=displaySum&mpid=21



## **Project Information:**

PROJECT_ID	P1110
PROJECT_NAME	Synaptic alterations in transient ischemia
PROJECT_DESCRIPTION	Investigation of morphological alterations of post-synaptic densities following an episode of transient ischemia
LEADER	Bingren Hu
FUNDING_AGENCY	NIH
PROJECT_START_DATE	1995-01-01 00:00:00.0
PROJECT_END_DATE	
COLLABORATORS	Maryann Martone; Ying Jones
PUBLICATION1	Martone M. E.; Jones Y. Z.; Young S. J. Ellisman; M. H. Zivin; J. A. and Hu B. R. Modification of postsynaptic densities following transient cerebral ischemia:a quantitative and three dimensional ultrastructural study. J. Neurosci.19:1988-1997.1999
PUBLICATION2	
PUBLICATION3	

Experiment Information -	
PURPOSE	3D structure of post-synaptic densities selectively stained with EPTA
TITLE	Tomographic reconstruction of EPTA stained post-synaptic densities
EXPERIMENTER	Bingren Hu
EXPERIMENT_NAME	
EXPERIMENT_DATE	1997-05-01 00:00:00.0

Subject Information -	
GROUP_BY	time of reperfusion
SUBJECT_NAME	sham operated control
FIXATION_METHOD_ID	1
SCIENTIFIC_NAME	rattus norvegicus
SPECIES	rat
STRAIN	Wistar
AGE	
AGECLASS	adult
ANIMAL_NAME	
LITTER_ID	
SEX	male
VENDOR	
WEIGHT	275 grams

Tissue -	
ANATOMIC_LOCATION	hippocampus
MICROTOME	ultramicrotome
ORIENTATION	coronal
THICKNESS	1 um
TISSUE_PROD_STORAGE	
EXTERNAL_FILE_NAME	
TISSUE_GROUP_TYPE	

Microscopy Product Information -	
MICROSCOPY_PRODUCT_ID	21
IMAGE_BASENAME	shamca1
CREATE_DATE	1997-05-30 00:00:00.0
INSTRUMENT	JEOL4000 IVEM
MICROSCOPE_TYPE	IVEM
PLANE_COUNT	
PRODUCT_TYPE	single tilt
PURL	10066252
SESSION_NAME	shamca1/shamca1_seg.jpg
TELESCIENCE_SRB	P1110/Experiment_13/Subject_13/Tissue_16/Microscopy_21
X_RESOLUTION	.004 um
Y_RESOLUTION	.004 um
XSIZE	1024
YSIZE	1024

### **Protocol:**

Ischemia model: All experimental procedures were approved by the Subcommittee on Animal Studies of the Veterans Affairs Medical Center (San Diego, CA). Male Wistar rats (250-300 gm) were fasted overnight. Anesthesia was induced with 3% halothane followed by maintenance with

Catheters were inserted into the external jugular vein, tail artery, and tail vein to allow blood sampling, arterial blood pressure recording, and drug infusion. Both common carotid arteries were exposed and encircled by loose ligatures. Fifteen minutes before ischemia induction and 15 min after ischemia, blood gases were measured and adjusted to PaO2 >90 mmHg and PaCO2 35-45 mmHg, pH 7.35-7.45, by adjusting tidal volume of the respirator. Bipolar EEG was recorded every 5-10 min before ischemia, continuously during the ischemic insult, and every 5 min after ischemia until the rat recovered from the anesthesia. At the beginning of a 30 min steady-state period before induction of ischemia, the inspired halothane concentration was decreased to 0.5%, and 150 IU/kg heparin was administered intravenously. Blood was withdrawn via the jugular catheter to produce a mean arterial blood pressure of 50 mmHg, and ischemia was induced by clamping both carotid arteries. Blood pressure was maintained at 50 mmHg during the ischemic period by2 end of the ischemic period, the clamps were removed, and the blood was reinfused through the jugular catheter, followed by 0.5 ml of 0.6 M sodium bicarbonate. In all experiments, temperature was maintained at 37C before, during, and after ischemia (15 min of reperfusion). Halothane was discontinued at the end of ischemia, and all wounds were sutured. At 4 or 24 hr, 3 d, or 1 week after the ischemic episode, the animals were reanesthetized, tracheotomized, and artificially ventilated. For electron microscopic studies, the brains were perfused with ice-cold 2% paraformaldehyde and 2.5% glutaraldehyde in 0.1 M cacodylate buffer. Sham-operated control rats were subjected to the same surgical procedures but without induction of ischemia.

1-2% halothane in an oxygen/nitrous oxide (30/70%) gas mixture.

Electron microscopic studies: Tissue sections from experimental and control animals were stained either by 1% ethanolic phosphotungstic acid (E-PTA) (Bloom and Aghajanian, 1966, 1968) or by the conventional osmium-uranium-lead method. Briefly, coronal brain sections were cut at a thickness of 200 um with a Vibratome through the level of the dorsal hippocampus and post-fixed for 1 hr with 4% glutaraldehyde in 0.1 M cacodylate buffer, pH 7.4. For conventional osmium-uranium-lead staining, sections were post-fixed for 2 hr in 1% osmium tetroxide in 0.1 M cacodylate buffer, rinsed in distilled water, and stained with 1% aqueous uranyl acetate overnight. The tissue sections were then dehydrated in an ascending series of ethanol to 100%, followed by dry acetone, and embedded in Durupan ACM. Thin sections were counterstained with lead citrate before examination in the electron microscope. For E-PTA staining, sections were dehydrated in an ascending series of ethanol to 100% and stained for 1 hr with 1% PTA prepared by dissolving 0.1 gm of PTA in 10 ml of 100% ethanol and adding four drops of 95% ethanol. Sections were then embedded in Durcupan ACM.

Image Type -	
SINGLE_TILT_IMAGE_SEQ_ID	4
TILT_INCREMENT	2 degrees
SINGLETILTIMAGESEQ_ID	4

Image Type -	
TILT_INCREMENT	2 degrees
RANGE_MAX	60 degrees
RANGE_MIN	-60 degrees

Specimen Description -	
ANATOMICAL_DETAIL	21
ATLAS_COORD	, ,
CELL_TYPE	unspecified
ORGAN	brain
REGION	hippocampus
STRUCTURE	post synaptic density
SYSTEM	central nervous system

Electron Microscopy Product -	
EM_PRODUCT_ID	4
ACCELERATING_VOLTAGE	400 KeV
MAGNIFICATION	20000
RECORDING_MEDIUM	film

# Raw 2D Image

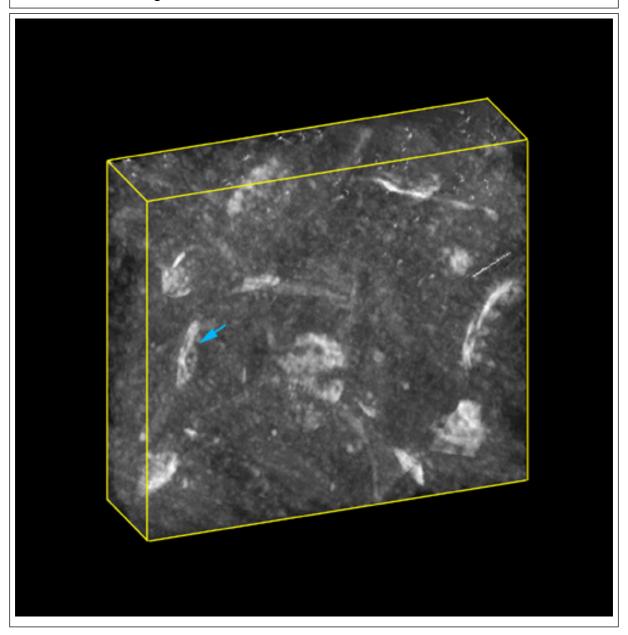
Raw Low Resolution 2D Image -



Raw 2D Image -	
IMAGE2D_ID	21
IMAGE_DESC	This tar file contains the original tile images; shamca1.???.f along with the fiducial mark file (shamca1.fido) used to align the data
IMAGE_FILE_FORMAT	suprim
IMAGE_FILE_NAME	shamca1/shamca1_img.jpg
MAGNIFICATION	20000
RAW_ANIMATION_FILE	shamca1/shamca1_img.qt
RAW_DATA_FILE	shamca1/shamca1_img.tar
THUMBNAIL_FILE	P1110/shamca1_rt.jpg

## Reconstruction

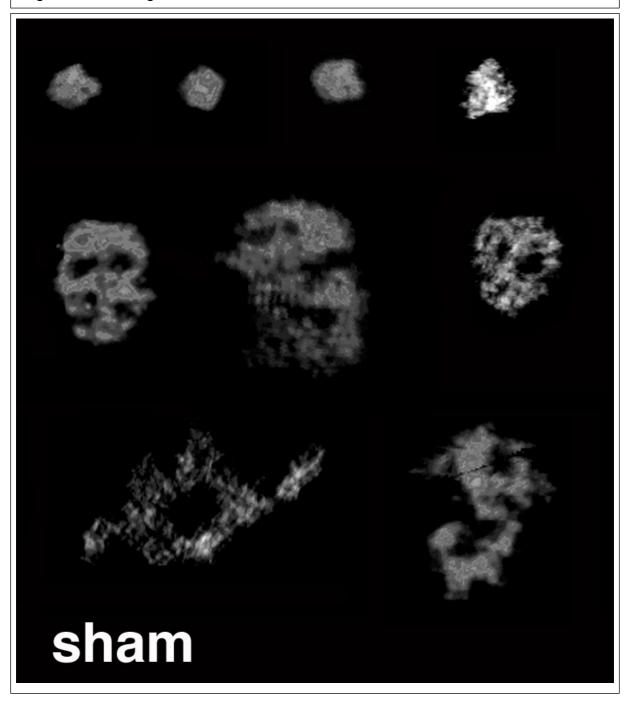
### Reconstruction Image -



·	
Reconstruction -	
RECONSTRUCTION3D_ID	21
ALIGNMENT_METHOD	manual
ALIGNMENT_PROGRAM	xfido
CROPPING_COORDINATE1	,
CROPPING_COORDINATE2	,
RECON_ALGORITHM	R-weighted back projection
RECON_DATE	1997-05-30 00:00:00.0
RECON_DESC	Volume reconstruction of selectively stained PSDs in Analyze format
RECON_PROGRAM	Suprim
RECON_TYPE	single tilt electron tomography
THUMBNAIL	P1110/shamca1_vt.jpg
VOLUME_DIMENSION	451, 411, 238
VOLUME_NAME	shamca1/shamca1_vol.zip
VOXEL_SCALE	.004, .004, .004
RECONSTRUCTION_IMAGES_I	21
RECON_IMAGE_DESC	maximum intensity projection of tomographic reconstruction of PSDs in area CA1 (blue arrow).Intensity is reversed so that PSDs appear bright against a dark background.
RECON_FILE_NAME	shamca1/shamca1.jpg
VOLUME_THUMBNAIL	P1110/shamca1_vt.jpg
ANIMATION_FILE	shamca1/rot_seq.qt
ANIMATION_DESC	rotation loop of maximum intensity projection of shamca1 in 5
	degrees. increments along the y axis. The contrast is reversed so
	that PSD appear bright against a dark background.

# Segmentation

Segmentation Image -



Segmentation -	
SEGMENTED_OBJECT_ID	83
NUMBER_OF_OBJECT	1
OBJECT_DESC	individual PSD
OBJECT_NAME	shamca1.obj
OBJECT_TYPE	volume
SEGMENTED_OBJ_2D_IMAGE	shamca1/a_shamca1.jpg
SEGMENTED_OBJECT_ID	83
SEGMENT_PERSON_NAME	Maryann Martone
SEG_ALGORITHM	simple threshold
SEG_DESC	Obj file contains individual PSDs segmented from shamca1.img using the "multiple objects" feature of Analyze.
SEG_FILE_NAME	shamca1/shamca1_seg.tar
THUMBNAIL	P1110/shamca1_st.jpg

#### **USER AGREEMENT**

Data Sharing and Citation Policy: The mission of the CCDB is to promote data sharing among scientists interested in cellular and subcellular anatomy and in developing computer algorithms for 3D reconstruction and modeling of such data. Data sets may be viewed or shared at the discretion of the author of the data. In some cases, the data may be freely viewed and downloaded without contacting the original author while in other cases, permission of the author may have to be obtained prior to downloading the data. In either case, failure to cite or give proper credit to the original authors who collected these data in subsequent published articles or presentations is a material breach of this User Agreement. CCDB requires all researchers re-analyzing these published data via the CCDB access to reference the original published article and the CCDB. An example of an appropriate acknowledgement is provided on the CCDB web site. CCDB is not in a position to police every intended use of these data. The scientific community will self-police the compliance of this contractual obligation.

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### **USER NOTIFICATION**

For large size image data, it will take several minutes to download, please be patient. Thanks!

### **ACKNOWLEDGEMENT**

Data used from the CCDB should be appropriately referenced, including both the author of the data and the CCDB. If the data were from a published study, the reference is included in the database record. The following reference should be cited for the CCDB:

Martone, M. E., Gupta, A., Wong, M., Qian, X., Sosinsky, G., Ludaescher, B., and Ellisman, M. H. A cell centered database for electron tomographic data. J. Struct. Biology 138: 145-155, 2002.

In addition, the support for the Cell Centered Database should be included in the acknolwedgement section of any publication: The Cell Centered Database is supported by NIH grants from NCRR RR04050, RR RR08605 and the Human Brain Project DA016602 from the National Institute on Drug Abuse, the National Institute of Biomedical Imaging and Bioengineering and the National Institute of Mental Health, and NSF grants supporting the National Partnership for Advanced Computational Infrastructure NSF-ASC 97-5249 and MCB-9728338.

Maryann Martone