

# Australasian Neuroscience Society 2022 Image Competition Submissions

### 1. Connectome deep brain stimulation

# Nicola Acevedo (Swinburne University, St Vincent's Hospital)

The image shows a reconstruction of deep brain stimulation (DBS) electrodes implanted in a patient with refractory obsessive-compulsive disorder (OCD), that deliver electrical impulses leading to modulation of local pathological neural circuits and global diffuse pathways. The pathways in red and blue represent white matter fibers respectively associated with good and poor clinical response, the white dots model the estimated stimulation artefacts. The image exemplifies recent advances in neurotechnology, biomedical engineering and computational neuroscience, that are influencing mechanistic understanding of refractory psychopathology, and targeting methods of neurostimulation therapy.

# 2. Deep brain stimulation electrode reconstructions

#### Nicola Acevedo (Swinburne University, St Vincent's Hospital)

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# 3. Deep brain stimulation electrode reconstructions 2

#### Nicola Acevedo (Swinburne University, St Vincent's Hospital)

The image shows a reconstruction of deep brain stimulation (DBS) electrodes implanted in patients with refractory obsessive-compulsive disorder (OCD); the field has recently shifted to evaluating whole brain circuitry (connectomes) in the identification of optimal stimulation targets. Our analysis supports this approach, responders (blue electrodes) received stimulation near fibers predictive of good response (red fibers), and non-responders (purple electrodes) received stimulation near fibers predictive of poor response (blue fibers). Therefore, representing advances in computational neuroscience, that are influencing mechanistic understanding of refractory psychopathology, and targeting methods of neurostimulation therapy.

# 4. The calcium signalling in dendrites

#### Ana Batallas Borja (Queensland Brain Institute)

This video shows the dynamics of calcium signalling in dendritic compartments of a principal hippocampal neuron following synaptic potentiation, a process that is essential for synaptic plasticity, learning and memory. A neuron-specific calcium-binding protein responded throughout the dendrites and spines in fluorescent cyan, while the cell filler Td-Tomato is in yellow.

# 5. Human induced Pluripotent Spiderweb

# Juan Botto (Queensland Brain Institute)

Human induced pluripotent cells-derived neurons expressing GFAP in cellular bodies and axonal projections

#### 6. Neuronal Chromesthesia

#### Juan Botto (Queensland Brain Institute)

Lentivirus-infected neurons stained for NeuN, Calcium-dependent activator protein for secretion 2 (CAPS-2) and vesicular glutamate transporter 2 (Vglut2).





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# 7. Neurovascular interactions

#### Jake Cashion (University of Tasmania)

Immunofluorescent labelling of pericytes (magenta), blood vessels (green) and microglia (cyan) in the mouse brain.

# 8. Human iPSC-derived POMC neurons

# Cortina Chen (University of Cambridge)

Hypothalamic POMC neurons differentiated from human iPSC. Pan neuronal MAP2 in red and POMC in yellow.

# 9. Neurons and Astrocytes

# Cortina Chen (University of Cambridge)

Human iPSC-derived neurons in red (MAP2) and mouse primary astrocytes (GFAP) in green. Image taken with Operaphenix high-content screening system.

# 10. Mouse Brain 3 Days Post-Stroke

# Hannah Coombe (University of Tasmania)

This image is of a mouse brain 3 days after a Rose Bengal photothrombotic stroke. The image was taken with a laser speckle to highlight the cerebral blood flow at this time point. The stroke was induced 1.5mm right of the midline at bregma where there is a clear blood flow deficit shown by the circular blue area amongst the red and yellow.

# 11. BV4-eyes

# Laura Carr (University of Adelaide)

BV2 cells treated with the proteasome inhibitor MG132 were stained with phalloidin, DAPI and anti-K48-linked ubiquitin antibody. Upon imaging using confocal microscopy BV2 microglia cells were apparently wearing glasses. Perhaps they were observing me.

# 12. Culture of Connection

# Elysa Carr (Florey Institute of Neuroscience and Mental Health)

As they grow, cultured hippocampal neurons (red – neurofilament M) reach for one another to establish connections in a sprawling network. Nuclei are also visible (blue – DNA). We grow these neurons to understand how they communicate at the molecular level. Growing in community is vital to the survival of these cells.

# 13. Amygdala: A "Switch" in the Brain

# Izel Eraslan (Deakin University)

A confocal microscope image (x20) of the central amygdala mirrored horizontally to present as a bow. Image is from a salt consuming male mice - neurons contain Fos (red), Enkephalin (green), and PKC-delta (blue).

# 14. Dendrite spine density of a parvalbumin interneuron Nicholas Crosbie (Monash University)

Created using immunohistochemistry, confocal microscopy and IMARIS neuron package. Blue: NeuN, Green: Parvalbumin, White: Dendrites, Pink: Dendrite Spines. Bregma 1.70mm. Scale bar is 10µm.

# 15. Dendrite spine density of a parvalbumin interneuron Nicholas Crosbie (Monash University)

Created using immunohistochemistry, confocal microscopy and IMARIS. Blue: NeuN, Green: Parvalbumin, White: Dendrites, Pink: Dendrite Spines. Bregma 1.98mm. Scale bar is 10µm.

# 16. Dendrite structure of parvalbumin interneurons

Nicholas Crosbie (Monash University)





Created using immunohistochemistry, confocal microscopy and IMARIS. Blue: NeuN, Green: Parvalbumin, White: Dendrites, Pink: Dendrite Spines. Bregma 1.98mm. Scale bar is 70µm.

# 17. A neuron in a hurry

# Eugenia Ferreiro (Queensland Brain Institute)

Immunostaining on mouse midbrain primary cultures. TH positive neuron (green) - Lamin nuclear envelope (magenta) - ORF1p (red) – DAPI

# 18. Networking

# Eugenia Ferreiro (Queensland Brain Institute)

Immunostaining on murine midbrain primary cultures. TH+ neuron (green) surrounded by other neurons in the culture (NeuN, magenta). Red dots indicate the expression of ORF1 protein

# 19. Nuclei full of stars

#### **Eugenia Ferreiro (Queensland Brain Institute)**

RNA in situ hybridization in murine primary culture

# 20. F-Actin Staining of iPSC-derived Pericytes

#### Alastair Fortune (Menzies Institute for Medical Research)

iPSCs from people with MS were differentiated into pericytes and stained with Phalloidin to visualise the F-actin cytoskeleton.

# 21. A Colony of Induced Pluripotent Stem Cells

# Alastair Fortune (Menzies Institute for Medical Research)

Blood from people with MS was collected and reprogrammed into induced pluripotent stem cells. To ensure these cells are pluripotent the cells were stained for key pluripotency markers Oct4 and Tra-1-60

### 22. A Sun of Stem Cells

# Alastair Fortune (Menzies Institute for Medical Research)

Blood was collected from people with Multiple Sclerosis and was reprogrammed into induced pluripotent stem cells (iPSCs). The iPSCs were then allowed to form embryoid bodies which were dissociated and stained for the ectoderm marker Nestin.

# 23. Cholinergic Microglia Symbiosis

#### Rashmi Gamage (Western Sydney University)

The image represents a cholinergic neuron (green channel) in the medial septal nuclei of ChAT(BAC)-eGFP (GFP tagged cholinergic neurons) mouse basal forebrain, surrounded by lb1+ microglial cells (red channel), while all nuclei stained with DAPI (blue channel).

# 24. Ankyrin down cell potential

# Sian Genoud (Macquarie University)

Action potentials are produced at the axon initial segment. Here I immunobael 5DIV primary hippocampal neurons with cell bodies in blue, dendrites in red and a marker called AnkyrinG to identify early axonal development in green.

# 25. Microglia in mouse model of epilepsy

#### Tabitha Green (University of Colorado)

This is a 3D rendering of microglia in a 40 µm mouse brain slice that was stained with lonized calcium binding adaptor molecule 1 (Iba1). This image was taken on a Zeiss ApoTome.2 (3D renderings and pseudocoloring were done in Zen software, and no other manipulations were made to the image), as part of a study that examined the morphology of dividing microglia after cortical kainic acid injection.





# 26. Defasciculated associations

#### **Elizabeth Haines (Queensland Brain Institute)**

In the dunnart forebrain axons initially project laterally, fasciculated in a white matter tract. Later, medial association projections appear, coursing through the ipsilateral cortical grey matter.

# 27. The Joker

### Olivia Haller, Ines Semendric (University of Adelaide)

Western blot analysing rat hippocampal tissue for GFAP. Upon visualisation, western blot appeared to be taunting us. Contender for the western blot hall of shame.

#### 28. Developing Human Dorsal Root Ganglion Kateleen Hedley (University of Newcastle)

Immunofluorescence was used to label DRG at 10 WG for parvalbumin (green) to show proprioceptive neurons, as well as NF200 (blue) a mechanoreceptive marker and SubP (red) a nociceptive marker, which display extensive cross over at this age (purple).

# 29. Glial Environment in the Dorsal Vagal Nucleus Kateleen Hedley (University of Newcastle)

At 12 weeks after a neonatal respiratory infection, immunofluorescence was used to label the brainstem region of the dorsal motor nucleus of the vagus. This showed neurons (blue), microglia (red) and astrocytes (green), with all three cells types clearly independently labelled.

# 30. (Not) Connected

# Patrick Heisterkamp (Ludwig Maximilian University of Munich)

Cerebral organoids grown in a microdevice connect to each other through axonal fibre tracts. The four organoids on the bottom right failed to develop properly and thus did not become part of the network. The image shows a staining for TUJ1, a neuron-specific tubulin found in axons and neuronal cell bodies.

# 31. Gang meeting

# Simon Brookes, Nan Chen, Tim Hibberd (Flinders University)

Human colonic myenteric ganglion. All morphological classes of human enteric neurons shown in a single ganglion. Nerve cell bodies labelled by neuronal tracer biotinamide, and antisera to the neurofilament proteins, peripherin and NF200.

# 32. Branching out our life

# Chia-Wei Huang (National Yang Ming Chiao Tung University)

Cortical neurons establish countless, dynamic, and crucial connections with other cells through synapses located in the branching dendrites. Let's embrace the wonderful world and branch out our colorful life as a great neuron!

# 33. Disco DRG

# Jacqueline Iredale (University of Newcastle)

Immunofluorescent staining of a mouse dorsal root ganglion (DRG) to confirm localisation of ChR2 to TRPV1+ neurons. Blue=NF200 labelling small and large myelinated A-fibre neurons, Green=GFP labelling TRPV1::ChR2 positive neurons, Red=CGRP labelling the peptidergic subpopulation of primary afferent neurons.

# 34. Bouquet of Neural Rosettes

Ayda Issa (University of Tasmania)





Immunofluorescent image of embryonic stem cell-derived pericytes exposed to 24h hypoxia stained with fluorescently labelled Phalloidin (red), with hypoxia revealed in green (detected using Hydroxyprobe Kit), co-labelled with DAPI (blue). Magnification 20x.

# 35. Hypoxic Pericytes

# Ayda Issa (University of Tasmania)

Immunofluorescent image of embryonic stem cell-derived pericytes exposed to 24h hypoxia stained with fluorescently labelled Phalloidin (red), with hypoxia revealed in green (detected using Hydroxyprobe Kit), co-labelled with DAPI (blue). Magnification 20x.

#### 36. Movin' Mito

#### Leanne Jiang (University of Queensland, University of Western Australia)

Live cell imaging of mitochondria (red). Cells are iPSC-derived lower motor neurons from a healthy patient. Mitochondrial dynamics clearly show mitochondria are motile and are constantly undergoing fusion and fission.

#### 37. How memories are made?

#### Merja Joensuu (Australian Institute for Bioengineering and Nanotechnology)

Hippocampal neurons (magenta) are responsible for learning and formation of precious memories. Lipid modifying enzymes (cyan) that are involved in the memory acquisition control membrane trafficking in the nerve endings and in the early secretory pathway (yellow).

#### 38. Cortical Constellations

### Dominic Kaul (University of Wollongong)

Astrocytes (star-shaped cells) form unique constellations in the human brain. These constellations help to control many functions, such as behaviour. Image consists of fluorescent tilescans of 16 human cortex sections, showcasing the diversity of individual astrocyte constellations.

# 39. That's my gut?

# Michaela E Johnson, Adam Humenick, Melinda Kyloh, Simon Brookes (Flinders University)

An "ascending nerve" running through the wall of the human colon. This image shows the nerve fibres of the ascending nerve (cyan) surrounded by a protective sheath (magenta) and a few myelinated nerve fibres (green), which was stained using immunofluorescence and imaged with a confocal microscope. Research into these nerves could lead to a better understanding of their important role in coordinating colonic function and how disruption of them may contribute to several common gut disorders.

# 40. Networking is Everything

# Narges Mahdavian (Monash Institute of Pharmaceutical Sciences)

Cells in the gut rely on forming networks with each other to control essential gut functions. Interstitial cells of Cajal (c-kit, pink) communicate with nerves (substance P, cyan) to mediate gut contractions. This image is of a myenteric wholemount preparation from donated paediatric colon, labelled by indirect immunofluorescence and acquired on a Leica SP8 laser scanning confocal microscope (40x objective, NA 1.3, 1024x1024 pixel resolution).

# 41. Connections in a dish

#### Simon Maksour (University of Wollongong)

Microglia are the brain's resident immune cell and play a role in preserving and protecting normal brain function. In neurodegenerative disease, microglia conversely play roles in the progression and degeneration of neutrons. The use of induced pluripotent stem cells allows for the generation of microglia in a dish from patients to explore what causes these in functions and homeostasis.





# 42. Microglia: friend or foe

# Simon Maksour (University of Wollongong)

The use of induced pluripotent stem cells (iPSCs) have allowed for the generation of cell types in a dish that are limited by access. These include neurons that can be differentiated in a dish from patients for disease modelling. This image shows neurons differentiated to explore differences at a molecular and functional level (green is the neuronal marker B-III-tubulin).

#### 43. Axonal-Glial Crosstalk - 'Talk to me like lovers do!' Gila Moalem-Taylor (University of New South Wales)

A co-culture of dorsal root ganglion sensory neurons and microglia in a microfluidic device, showing axonal outreach from the neuronal chamber (top; green) to the microglial chamber (bottom; orange). This setup allows the investigation of axonal-microglial crosstalk in vitro. (Image was taken by Chiettha Prajnadewie from the Neuropathic Pain Research Group at UNSW).

# 44. Microglia and pericytes interacting in the adult brain

# Gary Morris (University of Tasmania)

Microglia in the brain (green) and pericytes (magenta) on brain capillaries interacting in the adult mouse somatosensory cortex, without the interference of astrocyte endfeet (white, DAPI labelled nuclei are in blue). These interactions may influence brain blood flow, the blood brain barrier and new blood vessel formation. This image was obtained with an inverted Ti Eclipse microscope (Nikon, Japan) equipped with a CSU-X1 spinning disk scanner (Yokogawa Electric Corporation, Japan), using a 100x oil objective (for more information refer to our pre-print: <a href="https://www.biorxiv.org/content/10.1101/2022.08.08.503250v1.full">https://www.biorxiv.org/content/10.1101/2022.08.08.503250v1.full</a>).

# 45. Blue fireworks

# Jenny Ngo (Florey Institute of Neuroscience and Mental Health)

Motor neurons in middle aged mouse displaying autophagy flux, the degradative pathway responsible for the removal of protein aggregates.

# 46. Shedding a light on developing brains

# Ishara Paranawithana (Monash University/Bionics Institute)

The aim of this study is to investigate how brain functional networks of hearing-impaired infants change during infancy compared to normal-hearing peers and its effects on language development using non-invasive optical brain imaging technique; functional near-infrared spectroscopy (fNIRS). The figure shows the sensitivity profile of the probes used to record brain activity from auditory and language areas (blue and red colors in the heatmap represent low and high sensitivity, respectively). Eight sources and eight detectors placed over bilateral temporal and prefrontal regions are marked by red and blue filled circles, respectively and fNIRS measurement channels are shown in yellow lines.

# 47. Pathfinder

# Igor Bonacossa Pereira (University of Queensland)

This is a cut touch-sensing neuron on the right in C. elegans trying its best to reach its disconnected part on the left. Neuron is shaded in light blue and the epidermal membrane cytoskeleton is shaded in purple.

# 48. Structures of Memory

# **Emmanuel Prikas (Flinders University)**

Depiction of complexity of neuronal network arrangements underlying learning & memory. The segregated localisation of Map2 (red) & Vamp2 (yellow) give clear non-overlapping regions of red and yellow, which look like red tree branches sprouting golden yellow leaves.

# 49. A good bowl of noodle





# Hazel Quek (QIMR Berghofer Medical Research Institute)

3D culture of neurons (nestin-red) and (double cortin-green)

#### 50. Yummy

#### Hazel Quek (QIMR Berghofer Medical Research Institute)

3D culture of monocyte-derived microglia (green) clustering around an amyloid plaque (blue)

### 51. Dance of Oligodendrocytes around Myelin

#### Shwathy Ramesan (Florey Institute of Neuroscience and Mental Health)

The image shows a whole organoid consisting of Oligodendrocytes ensheathing axons with mature myelin (in green). The organoids also have active microglial cells (in red) surveying the surroundings constantly for any signs of inflammation. This is a working model to study demyelination and demyelination in Multiple Sclerosis.

#### 52. Golden hippocampal neurons

### Belal Shohayeb (Queensland Brain institute)

Hippocampal neuron neurons beautifully overlapping, expressing green fluorescent protein (gold), and stained with filament polymerised actin marker (cyan).

#### 53. Neuronal galaxy

#### Belal Shohayeb (Queensland Brain Institute)

Hippocampal neurons expressing green fluorescence protein and stained with gobular actin marker and filament actin marker.

# 54. Neuronal network

#### **Belal Shohayeb (Queensland Brain Institute)**

An overlay of 2 sets of hippocampal neurons stained with a monomeric globular actin marker in orange and cyan showing complicated neuronal network.

# 55. Midbrain of a Dragonfly Larva

#### Katie Skeen (University of Adelaide)

The midbrain of an early-stage dragonfly larva. Cyan = DAPI. Magenta = anti-synapsin.

# 56. Imaging the human brainstem

#### Kristie Smith (Neuroscience Research Australia)

The postmortem brainstem of a 65 year old male was imaged with a 7T MRI system, producing a GRE, DWI, and FAC data set that we used to construct the highest resolution MRI Atlas of the Human Brainstem (Paxinos et al., in-press for 2023). This is a single sagittal view section of the brainstem, captured approx. 8mm from the midline of the brain Distinctive features such as the substantia nigra, longitudinal fasciculus of the pons and transverse fibers of the pons are some of the many features clearly visible in this stunning image set.

#### 57. Rainbow paddlepop..err hippocampus

#### Kristie Smith (Neuroscience Research Australia)

This is the hippocampus of an adult mouse expressing Thy-1 YFP and stained with DAPI. The 40 micron tissue section was imaged using a Zeiss confocal microscope with a 20x objective. The image was imported into Image J and the LUT adjusted, resulting in this rainbow hippocampus. The black streak down the right hand side is an artifact - the result of mounting medium flowing over the coverslip. It reminds us even the most beautiful things can have tiny imperfections, and that just makes them even more interesting.

# 58. Cortical fireworks

Kristie Smith (Neuroscience Research Australia)



This is the cortex of a CLARITY cleared brain, from a mouse expressing Thy-1 YFP. A z-stack of the 3mm thick tissue section was captured using a LaVision light-sheet UltraMicroscope with a 10x objective. The z-stack was imported into ImageJ where the LUT was adjusted, with the resulting video displaying cortical pyramidal neurons as a fireworks display.

# 59. Secondary Neurodegeneration in the Basal Ganglia Shannon Stuckey (University of Adelaide)

Double immunofluorescence staining using antibodies against IBA1 (red) and A $\beta$  (green) demonstrated the spatial co-localization of microglial cells around amyloid plaque cores in the basal ganglia of 12-month post-stroke male Sprague-Dawley rats.

# 60. A Nacreous Whorl of Neurodegeneration Lenore Tahara-Eckl (University of Auckland)

The Dementia Prevention Research Clinic (Auckland, New Zealand) investigates early biomarkers and factors influencing the onset and progression of Alzheimer's disease. Diffusion MRI allows us to see the widespread neurodegeneration in the white matter tracts, which is shown prominently in the fornix, hippocampal regions, and corpus callosum of the brain in individuals with Alzheimer's disease compared to older adults.

# 61. Brain Storm

# Ryu Takechi (Curtin University)

GFAP (red) showing activation of astrocytes surrounding the amyloid plaques (green) in hippocampal formation of APP/PS1 Alzheimer's mice.

# 62. Brain Vascular Forrest

# Ryu Takechi (Curtin University)

Spread of brain vascular network, supported by astrocytes

# 63. Visualising the rapid accumulation of Alzheimer's pathology Jennifer Tinston (Monash University)

Amyloid plaques are extracellular aggregates of misfolded protein, and underly the pathology of may neurodegenerative conditions including Alzheimer's disease.

Here, in vivo T2\*- weighted MRI can detect amyloid plaques in a 6-month 5xFAD mouse at different time points. The movie depicts the rapid accumulation of this pathology in the hippocampus over a 30-day period.

# 64. Macrophages to the rescue

# Andres Vidal-Itriago (Macquarie University)

Confocal time lapse of 4-days old transgenic zebrafish larvae, expressing BFP in motorneurons (cyan) and mCherry in macrophages (magenta). Time lapse shows the migration of macrophages to the injury site after performing the axotomy of the pectoral fin innervation.

# 65. Rainbow Danio

# Andres Vidal-Itriago (Macquarie University)

On this masterpiece, we see impressionistic pluripotent stem cells. The merge of the DNA stain in blue (DAPI) and fluorescent antibodies against the pluripotency markers OCT3/4 (green) and SOX-2 (red) creates a colorful representation of the beginning of a cell's fate. The impressionistic touch of this artwork is undeniable.

# 66. iPSCs - impressionistic pluripotent stem cells Florian Walter (Ludwig-Maximilians-Universität München)





An activated newly repopulated microglia (in red) with highly ramified morphology positioned above the granular cell layer in the mouse hippocampus after traumatic brain injury. DAPI labelling shows cell nuclei (blue).

# 67. An activated microglia

# Emily Willis (University of Queensland)

An activated newly repopulated microglia (in red) with highly ramified morphology positioned above the granular cell layer in the mouse hippocampus after traumatic brain injury. DAPI labelling shows cell nuclei (blue).

#### 68. Fate labelled neurons in adult hippocampus Emily Willis (University of Queensland)

Fate labelled neurons and their projections labelled with tdTomato fluorescent reporter (red) with DAPI (blue) in the adult mouse brain.

#### 69. Microglia in the adult hippocampus Emily Willis (University of Queensland)

Microglia in the adult hippocampus, labelled with a tdTomato reporter (red, CX3CR1+) and green (IBA1+) with DAPI (blue) showing the granule cell layer.

# 70. The beauty of zebrafish's main intellect power

Nurul Atiqah Zulazmi (Monash University) Small but magnificent