Visualising aluminium in the human brain across complex neurological disorders

Matthew Mold, PhD, MRSB

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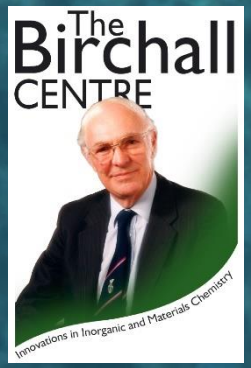
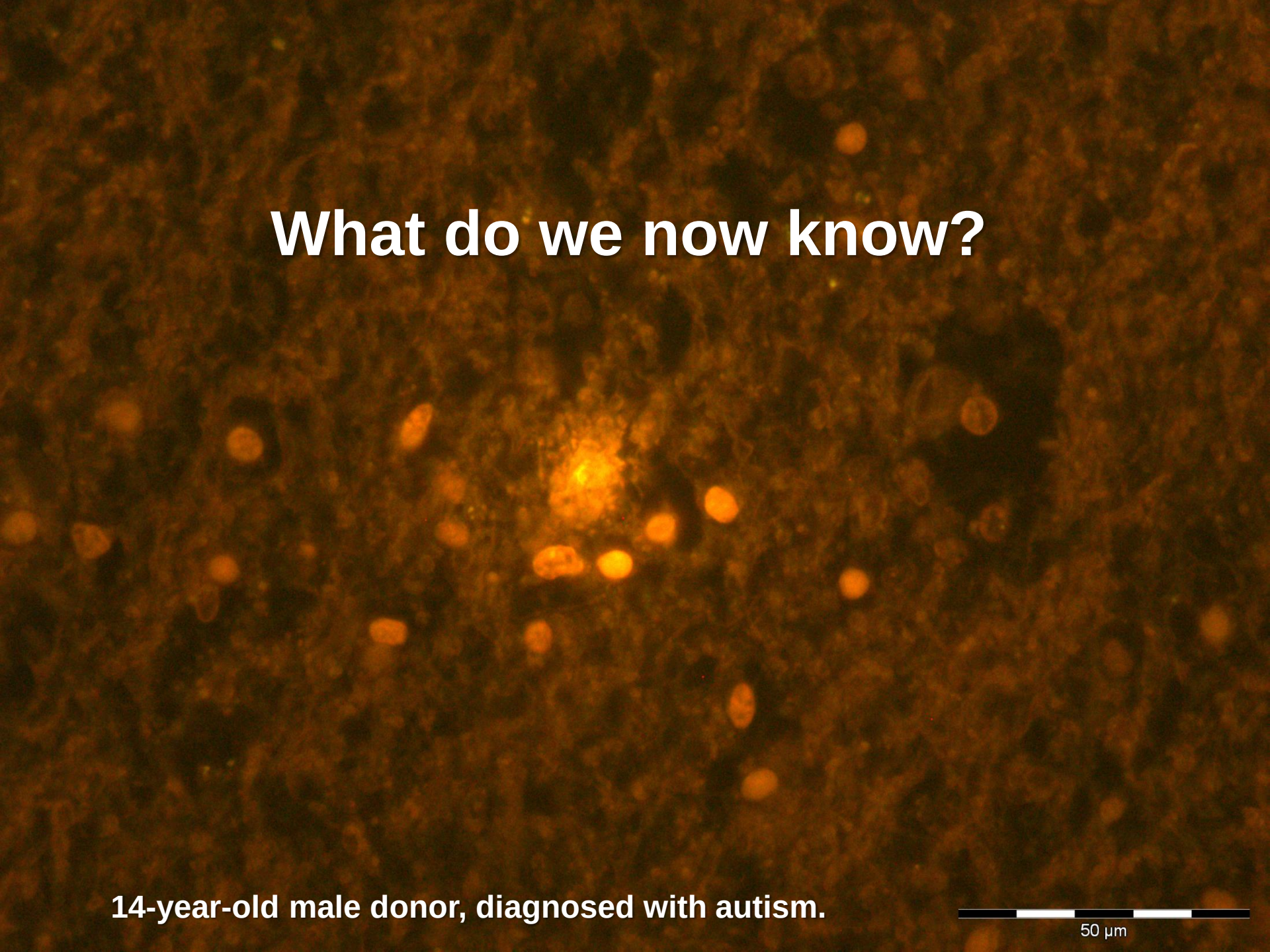


13th Keele Meeting on Aluminium, Mexico 2019

What do we now know?

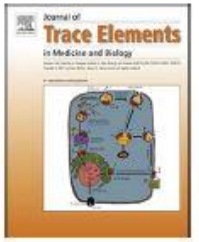


Journal of Trace Elements in Medicine and Biology 46 (2018) 76–82

Contents lists available at ScienceDirect

Journal of Trace Elements in Medicine and Biology

journal homepage: www.elsevier.com/locate/jtemb



Aluminium in brain tissue in autism

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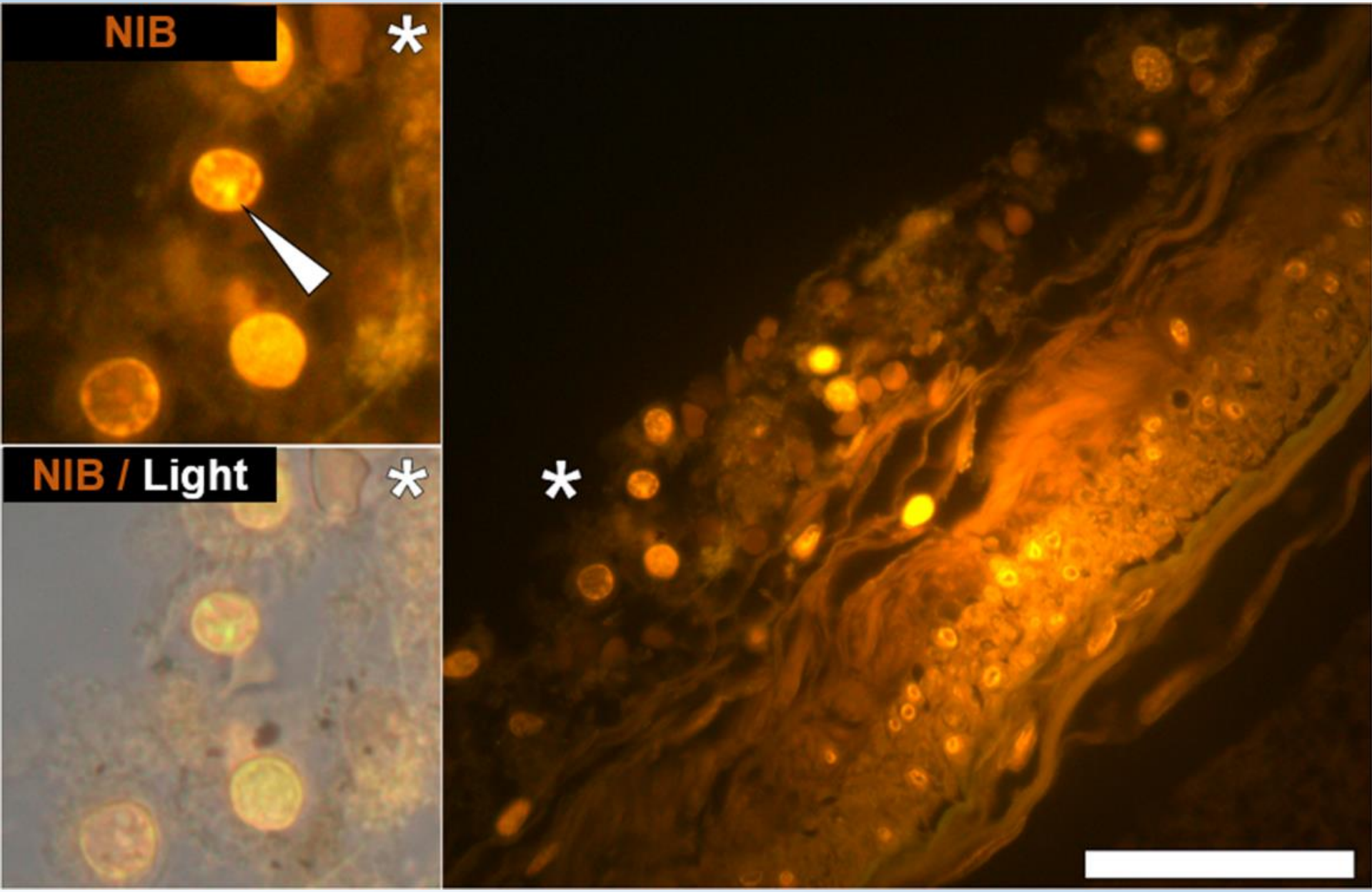
ARTICLE INFO

Keywords:
Human exposure to aluminium
Human brain tissue
Autism spectrum disorder
Transversely heated atomic absorption spectrometry
Aluminium-selective fluorescence microscopy

> 530,000 views on publishers' website.

ABSTRACT

Autism spectrum disorder is a neurodevelopmental disorder of unknown aetiology. It is suggested to involve both genetic susceptibility and environmental factors including in the latter environmental toxins. Human exposure to the environmental toxin aluminium has been linked, if tentatively, to autism spectrum disorder. Herein we have used transversely heated graphite furnace atomic absorption spectrometry to measure, for the first time, the aluminium content of brain tissue from donors with a diagnosis of autism. We have also used an aluminium-selective fluor to identify aluminium in brain tissue using fluorescence microscopy. The aluminium content of brain tissue in autism was consistently high. The mean (standard deviation) aluminium content across all 5 individuals for each lobe were 3.82(5.42), 2.30(2.00), 2.79(4.05) and 3.82(5.17) μg/g dry wt. for the occipital, frontal, temporal and parietal lobes respectively. These are some of the highest values for aluminium in human brain tissue yet recorded and one has to question why, for example, the aluminium content of the occipital lobe of a 15 year old boy would be 8.74 (11.59) μg/g dry wt.? Aluminium-selective fluorescence microscopy was used to identify aluminium in brain tissue in 10 donors. While aluminium was imaged associated with neurones it appeared to be present intracellularly in microglia-like cells and other inflammatory non-neuronal cells in the meninges, vasculature, grey and white matter. The pre-eminence of intracellular aluminium associated with non-neuronal cells was a standout observation in autism brain tissue and may offer clues as to both the origin of the brain aluminium as well as a putative role in autism spectrum disorder.



Aluminium in leptomeningeal membranes (50, M)

International Journal of
Environmental Research and Public Health

MDPI

Article

Aluminium in Brain Tissue in Multiple Sclerosis

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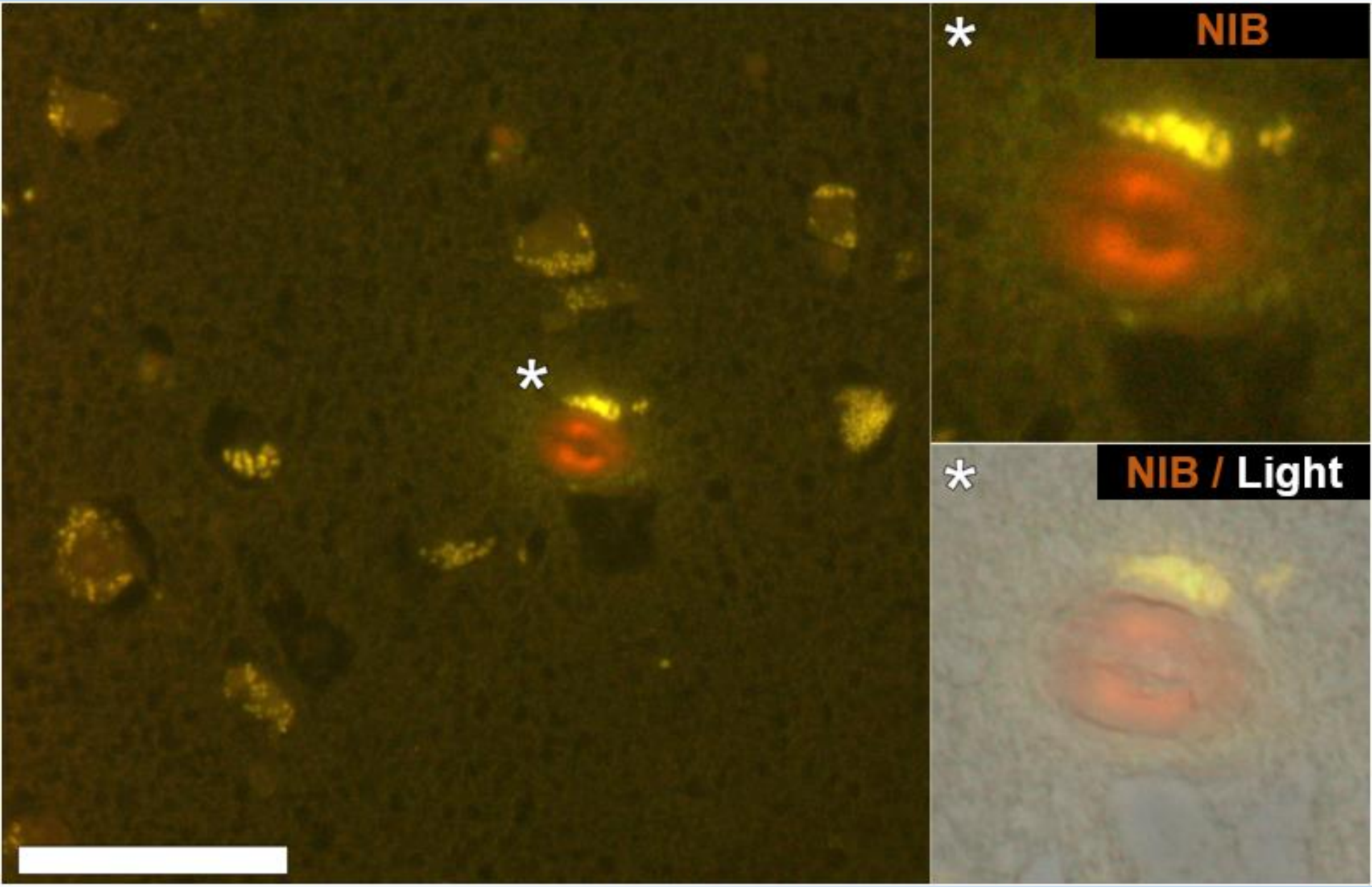
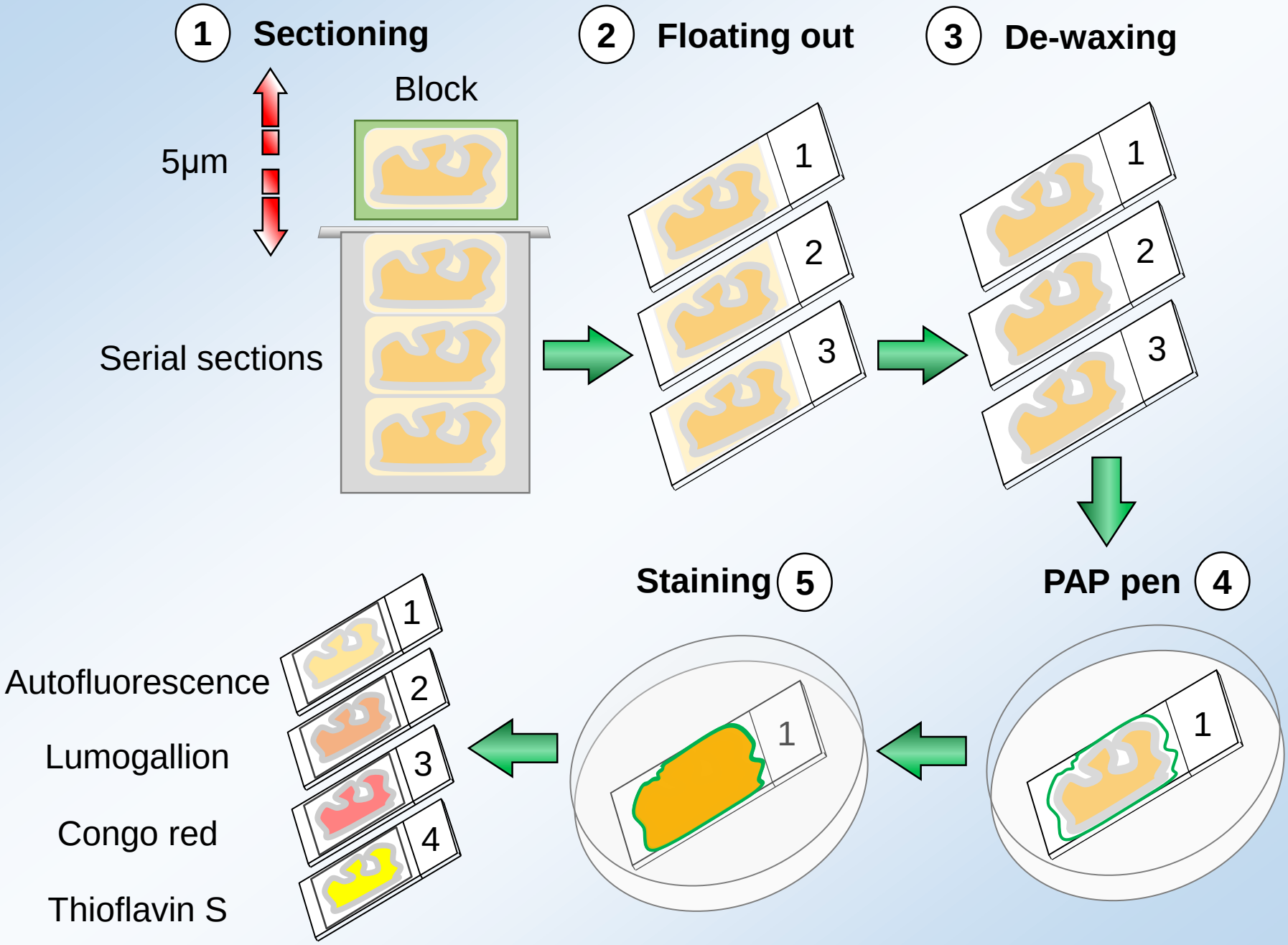
Received: 25 July 2018; Accepted: 15 August 2018; Published: 18 August 2018

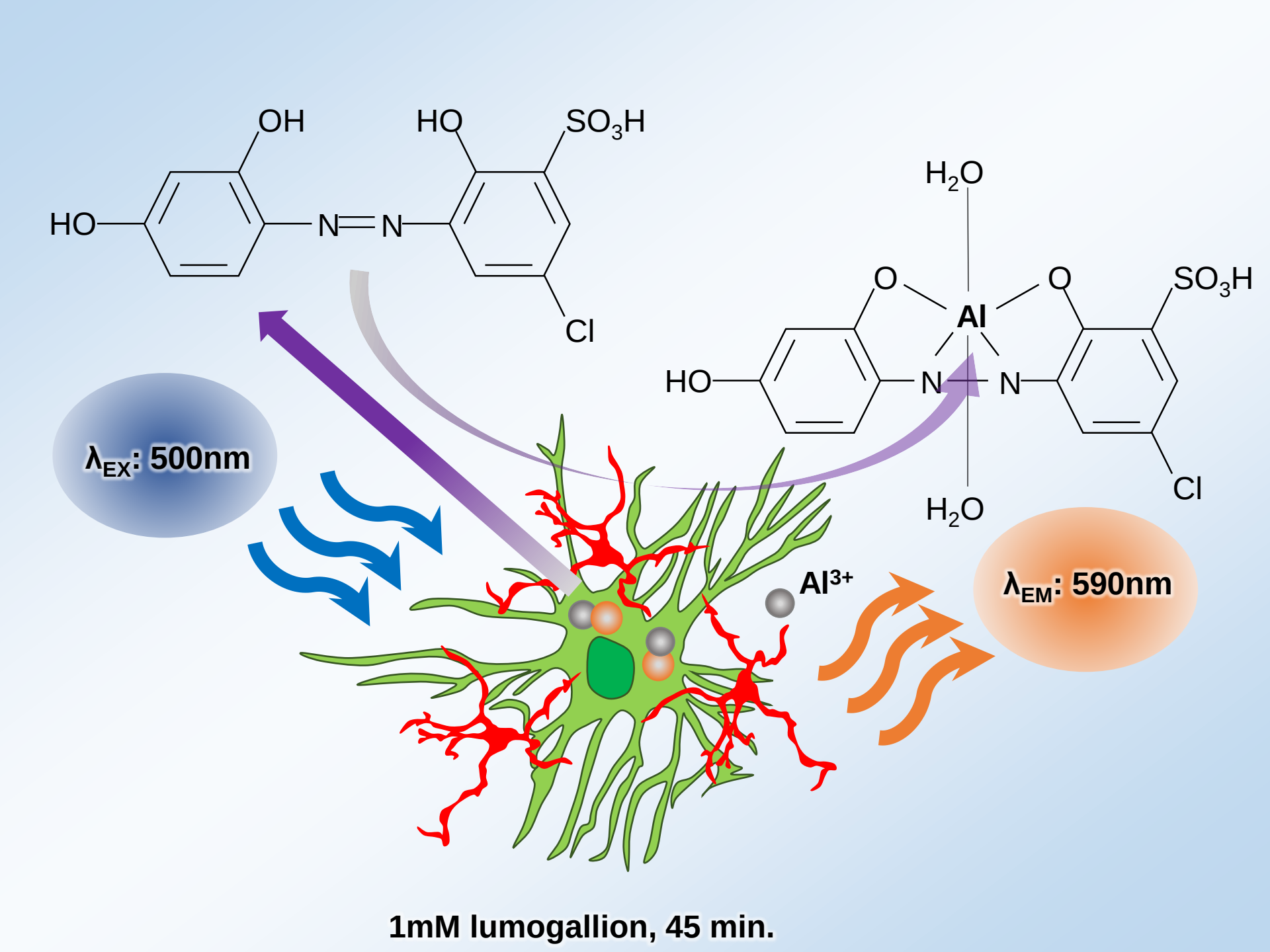
Abstract: Multiple sclerosis (MS) is a devastating and debilitating neurodegenerative disease of unknown cause. A consensus suggests the involvement of both genetic and environmental factors of which the latter may involve human exposure to aluminium. There are no data on the content and distribution of aluminium in human brain tissue in MS. The aluminium content of brain tissue from 14 donors with a diagnosis of MS was determined by transversely heated graphite furnace atomic absorption spectrometry. The location of aluminium in the brain tissue of two donors was investigated by aluminium-specific fluorescence microscopy. The aluminium content of brain tissue in MS was universally high with many tissues bearing concentrations in excess of 10 μg/g dry wt. (10 ppm) and some exceeding 50 ppm. There were no statistically significant relationships between brain lobes, donor age or donor gender. Aluminium-specific fluorescence successfully identified aluminium in brain tissue in both intracellular and extracellular locations. The association of aluminium with corpora amylacea suggests a role for aluminium in neurodegeneration in MS.



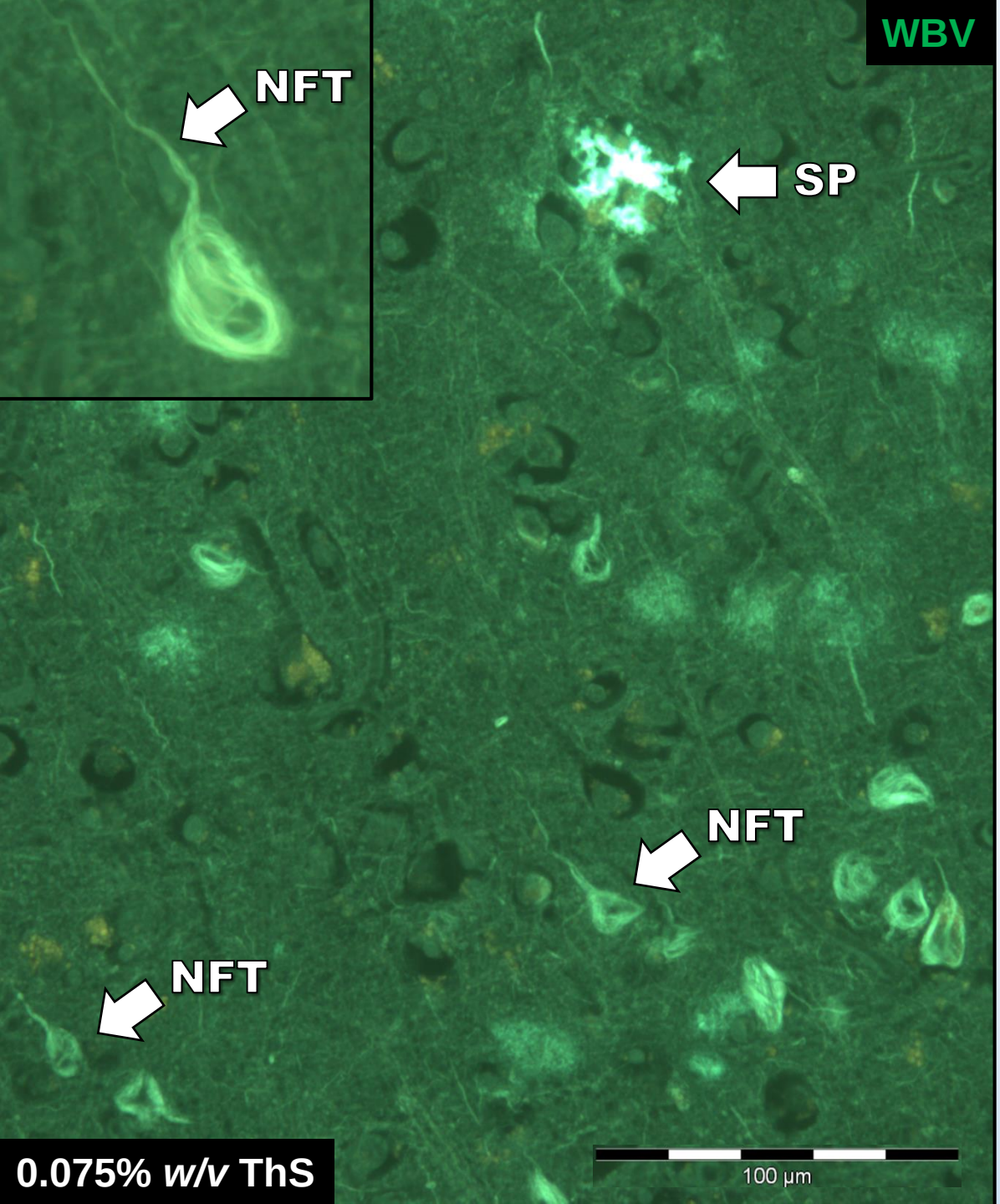
Aluminium in corpora amylacea (48, F)

Methodology


1 Sectioning
2 Floating out
3 De-waxing
Block
5µm
Serial sections
1
2
3
PAP pen 4
Staining 5
Autofluorescence
Lumogallion
Congo red
Thioflavin S
4

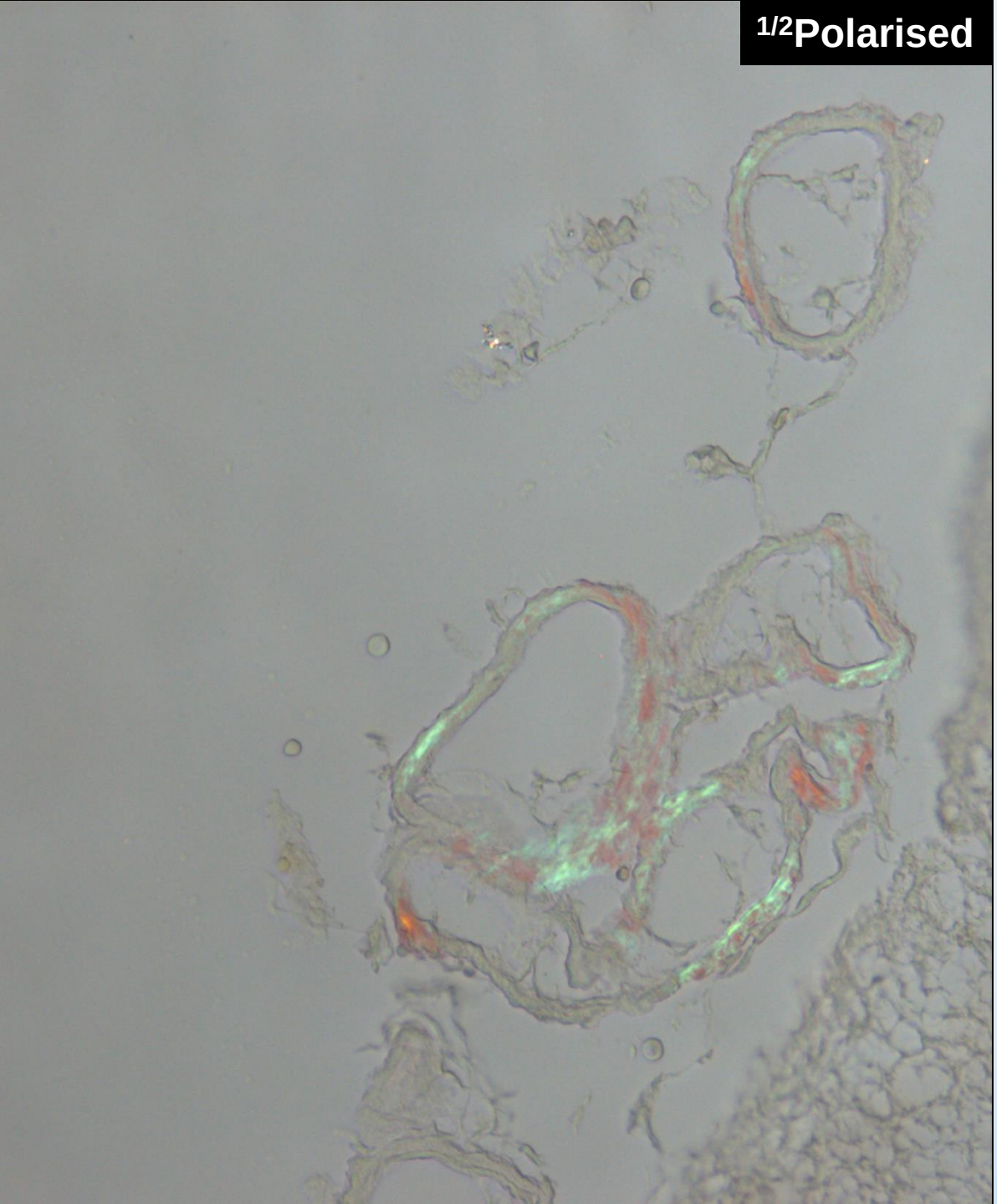


1mM lumogallion, 45 min.



Thioflavin S Staining

- Colombian donor presenting with *PSEN1 E280A* mutation.
- Early onset / familial Alzheimer's disease (fAD).
- Temporal cortex.
- **Thioflavin S** (ThS) staining reveals senile plaques (**SP**) and neurofibrillary tangles (**NFT**).



Congo Red

- Positive amyloid staining revealing Congophilic amyloid angiopathy (**CAA**).

CASE STUDY 1:

Camelford Incident:

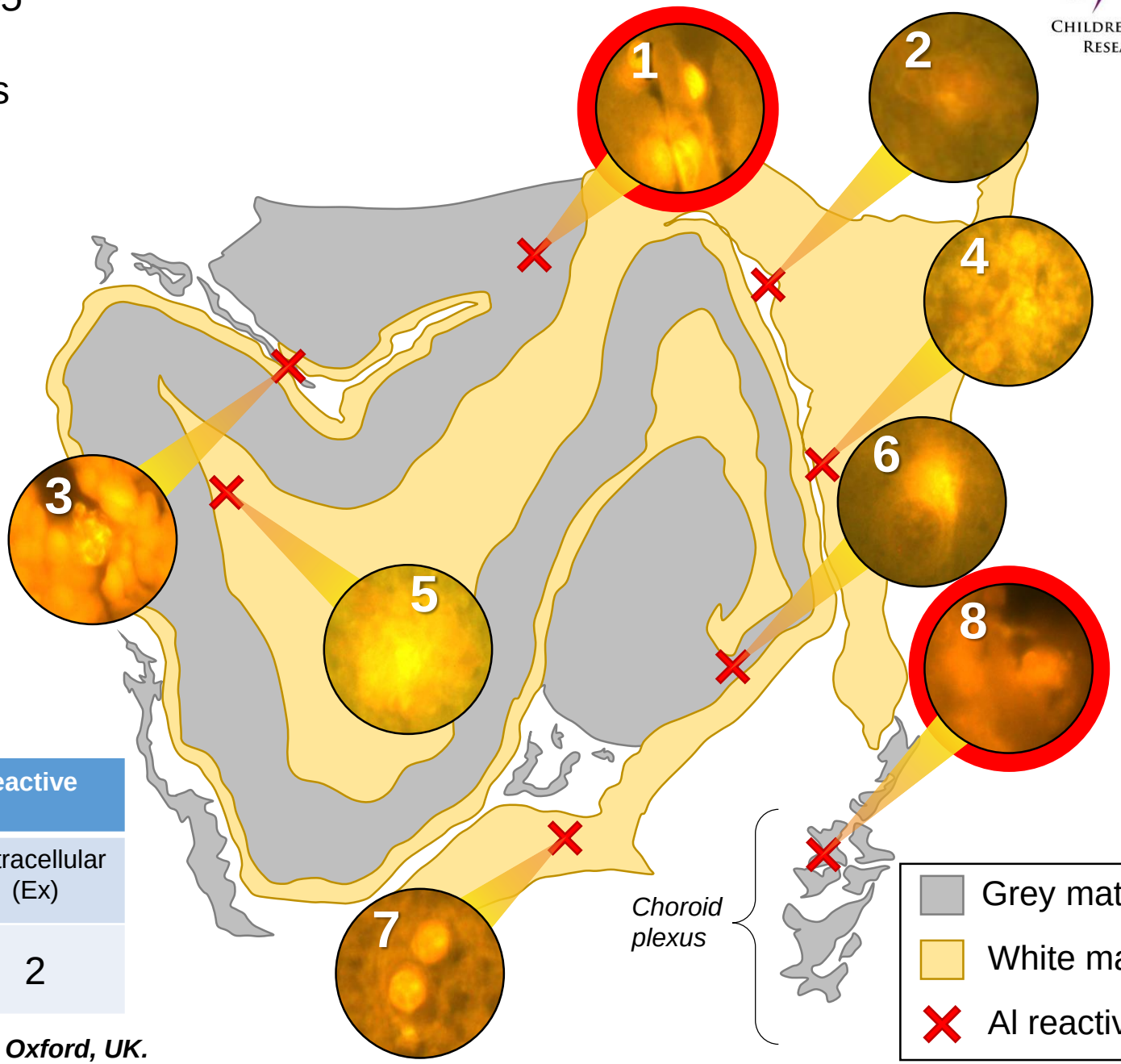
Congophilic / cerebral amyloid angiopathy

- 59-year-old female donor exposed to aluminium in contaminated drinking water.
- Neuropathological examination revealed extensive Congophilic amyloid angiopathy.
- Lumogallion and Congo red staining performed on brain tissue sections.

Patient ID:
NP040/2004-5
Region:
Hippocampus

CASE STUDY 1

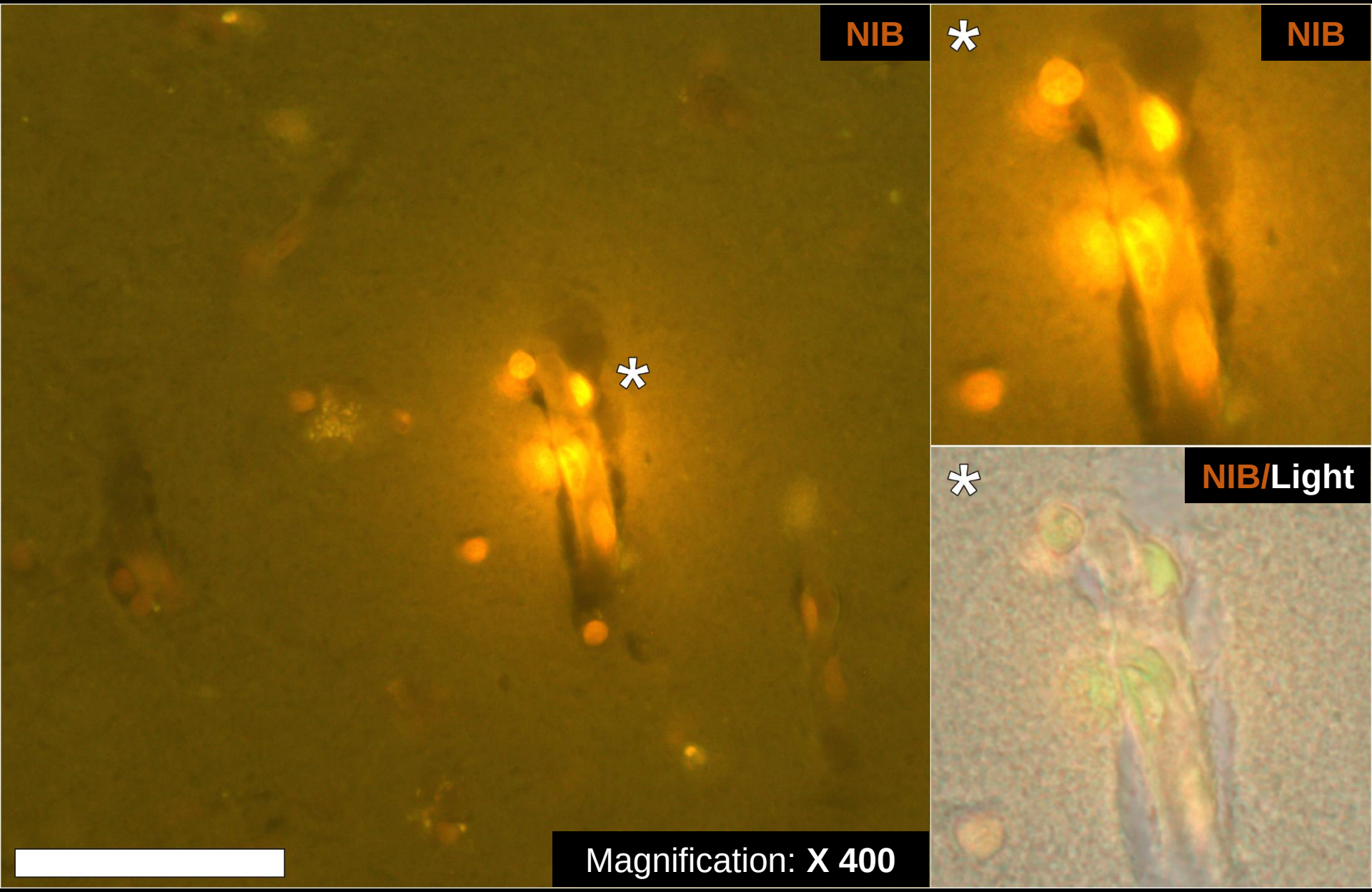
CMSRI
Children's Medical Safety Research Institute



Number of AI reactive regions	
Intracellular (In)	Extracellular (Ex)
6	2

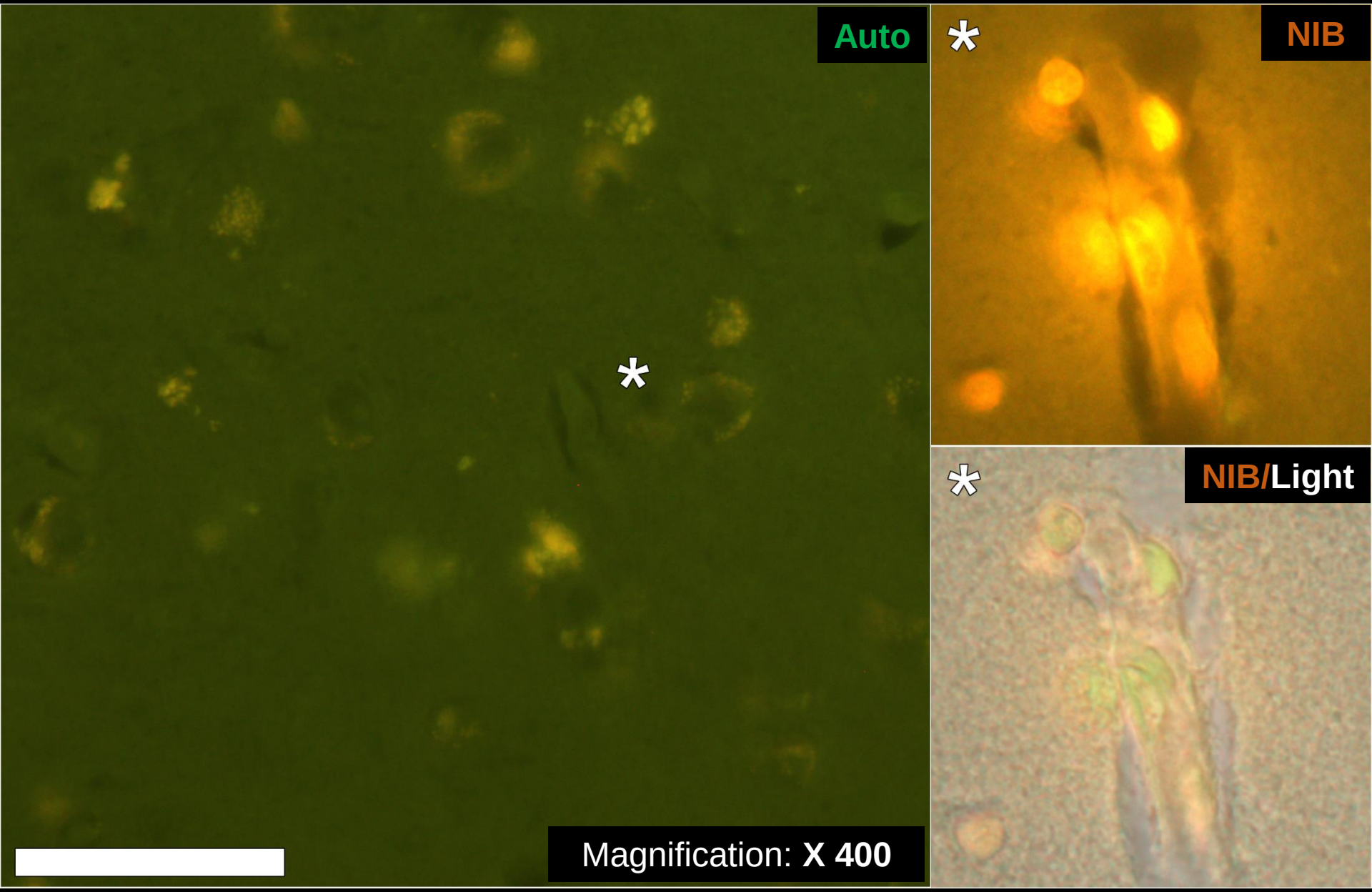
Oxford Brain Bank, Oxford, UK.

- **Hippocampus**



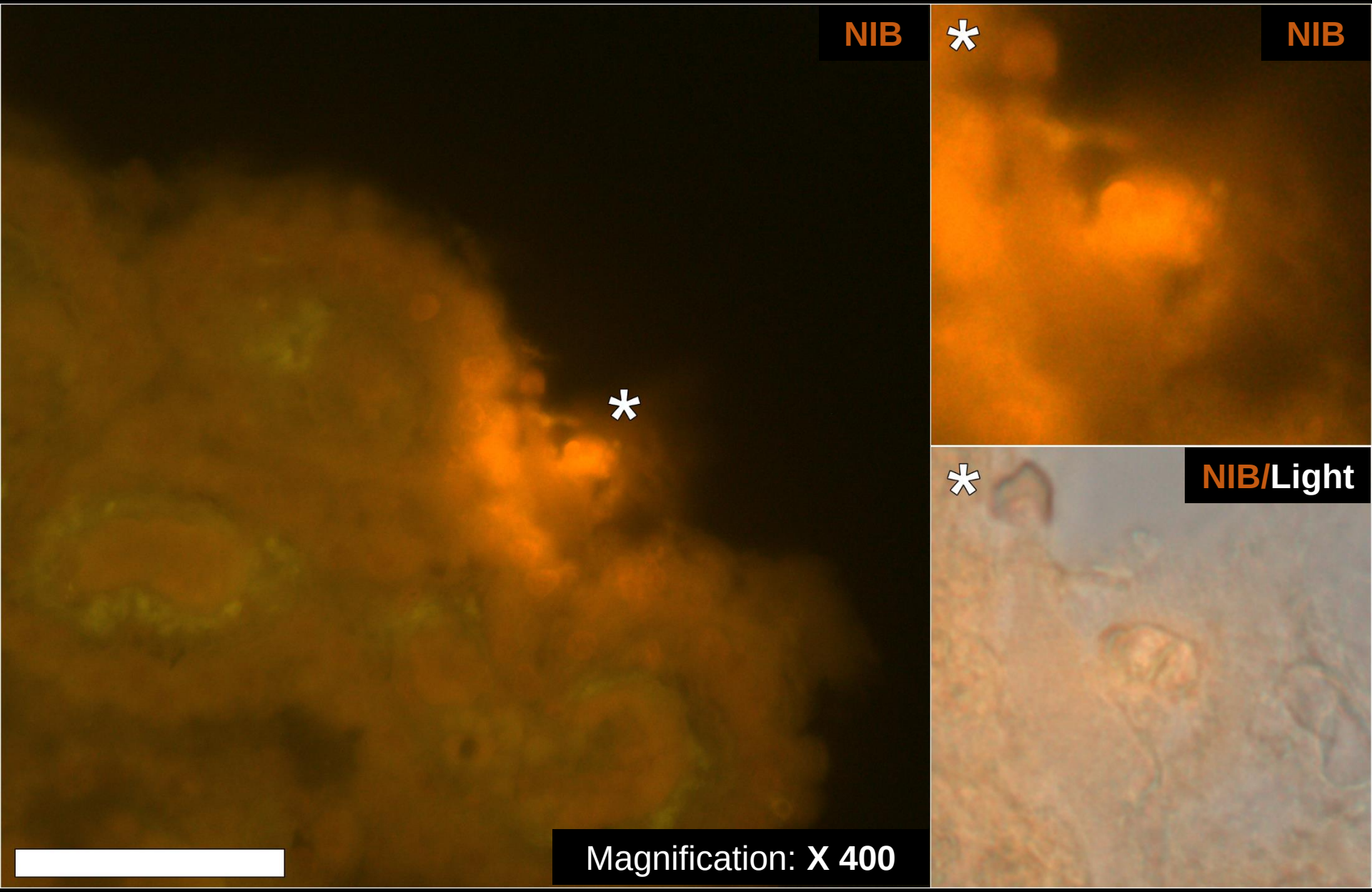
Intracellular aluminium in the vessel wall. (Region: #1)

- **Hippocampus**



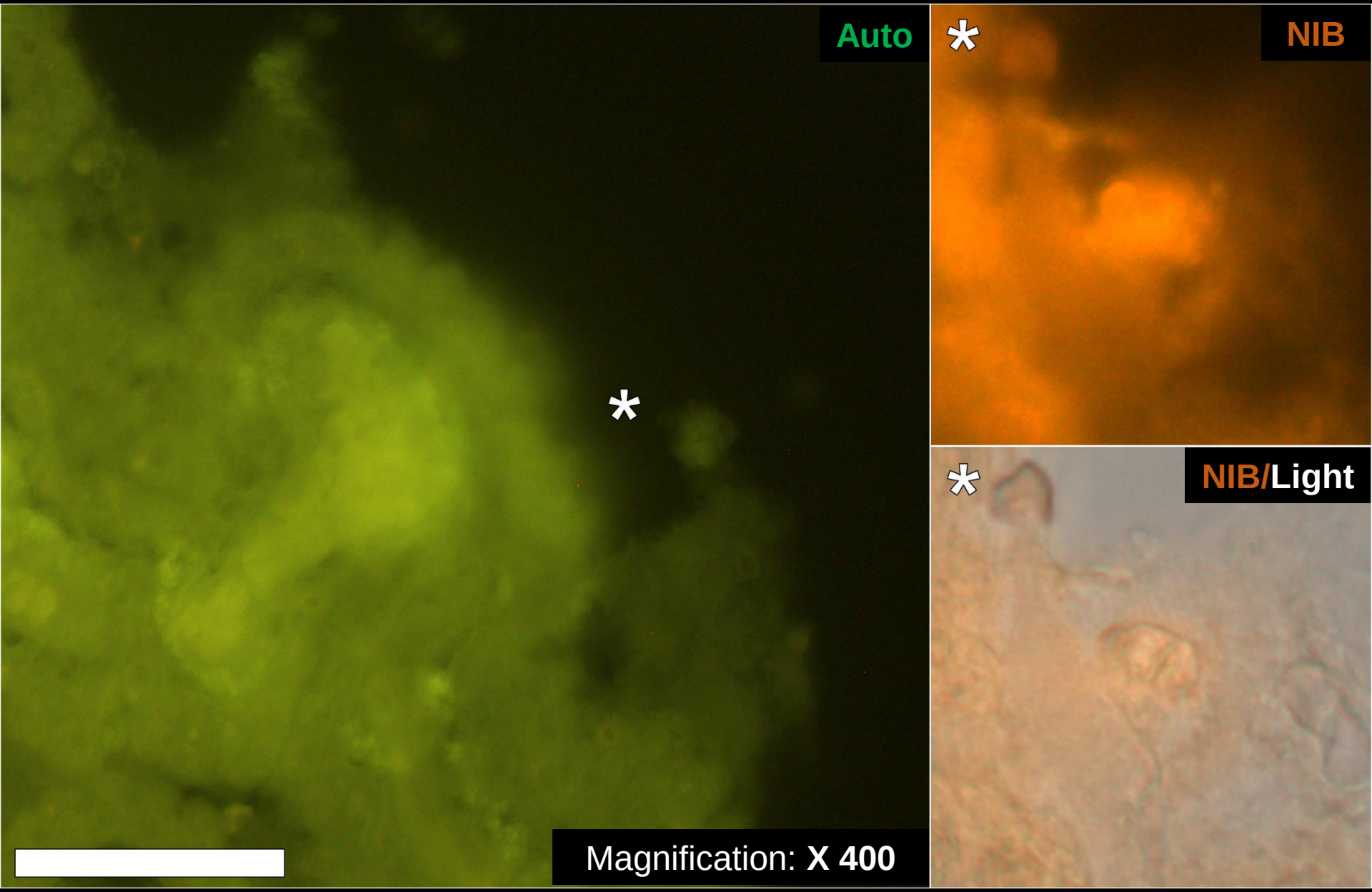
Intracellular aluminium in the vessel wall. (Region: #1)

- **Hippocampus**



Epithelial cells lining the choroid plexus. (Region: #8)

- **Hippocampus**



Epithelial cells lining the choroid plexus. (Region: #8)

Patient ID:
NP040/2004-5
Lobe:
Occipital

CASE STUDY 1

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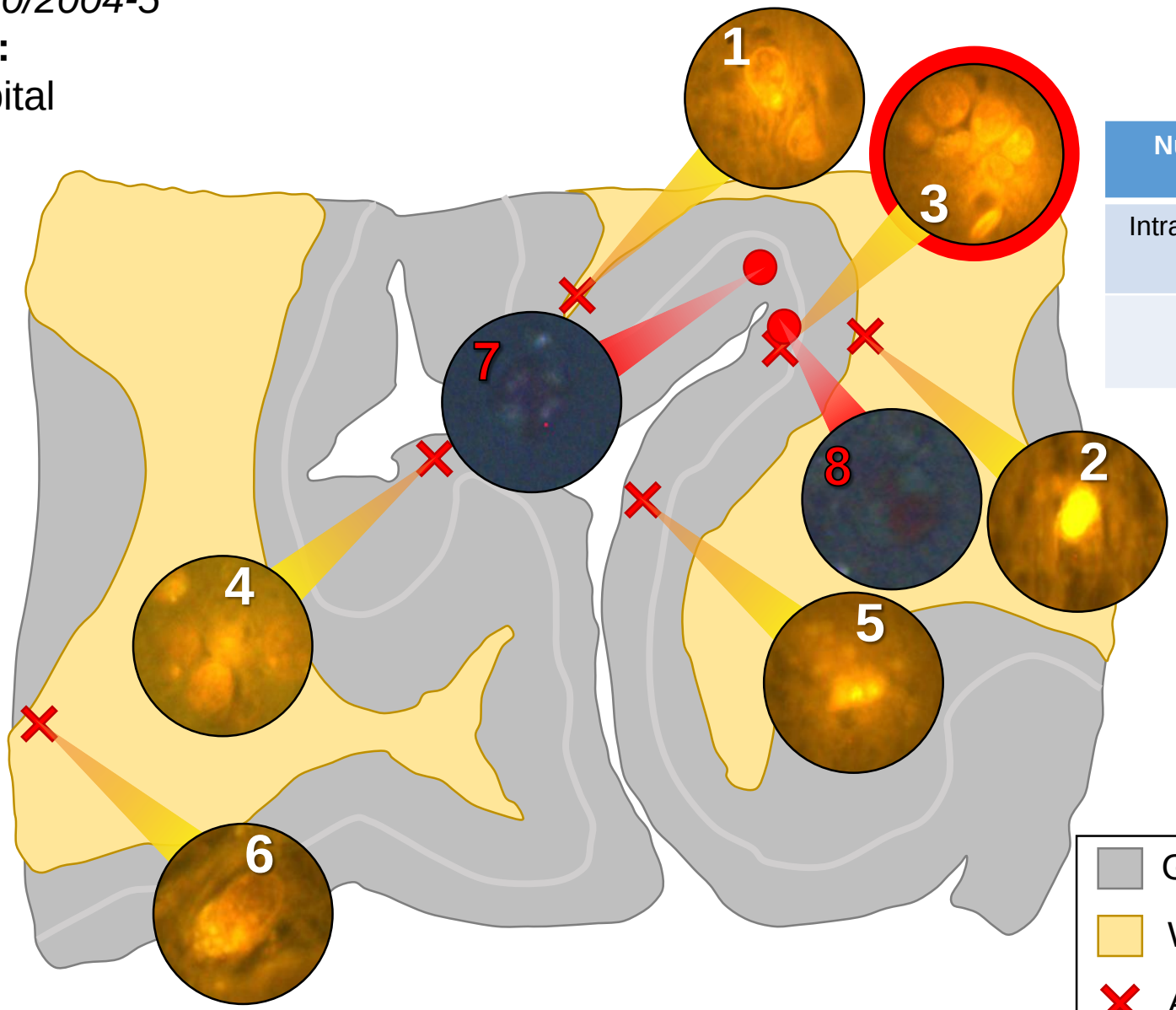
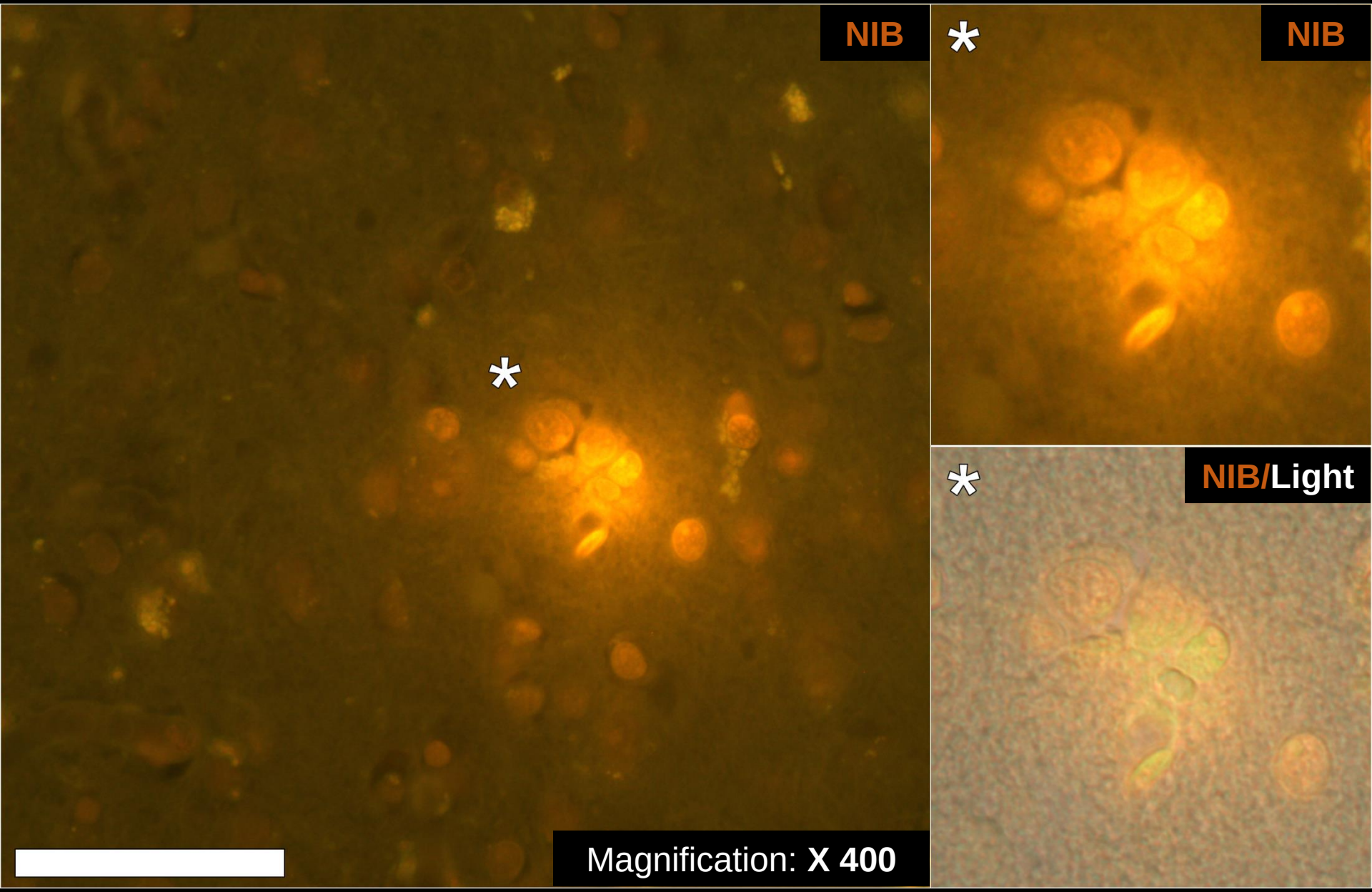


Number of AI reactive regions	
Intracellular (In)	Extracellular (Ex)
6	0

Grey matter (GM)
White matter (WM)
AI reactive
CR positive

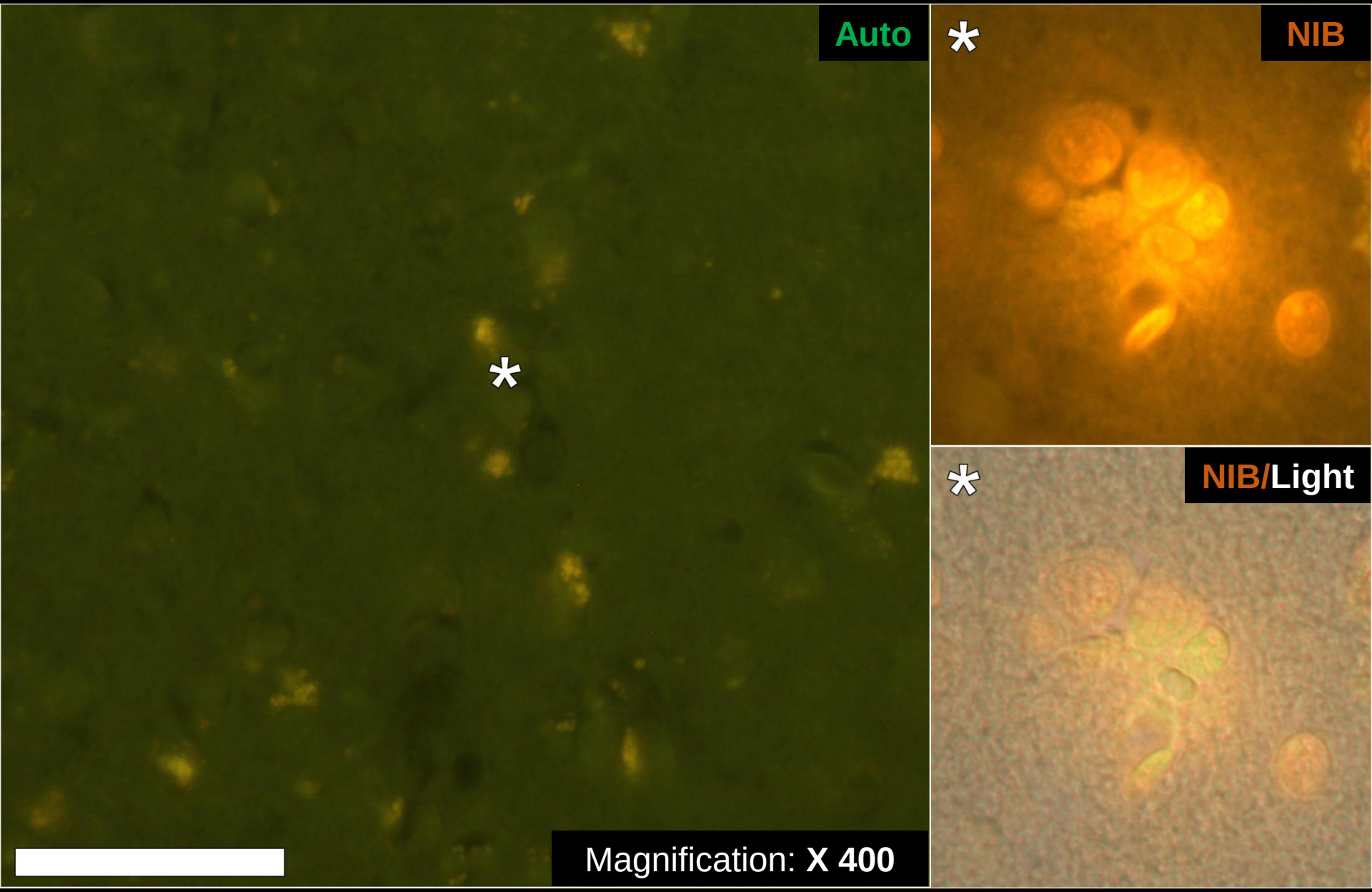
Oxford Brain Bank, Oxford, UK.

- **Occipital cortex**



Aluminium in astrocytes & microglial cells. (Region: #3)

- **Occipital cortex**



Aluminium in astrocytes & microglial cells. (Region: #3)

Patient ID:
NP040/2004-5
Lobe:
Parietal

CASE STUDY 1

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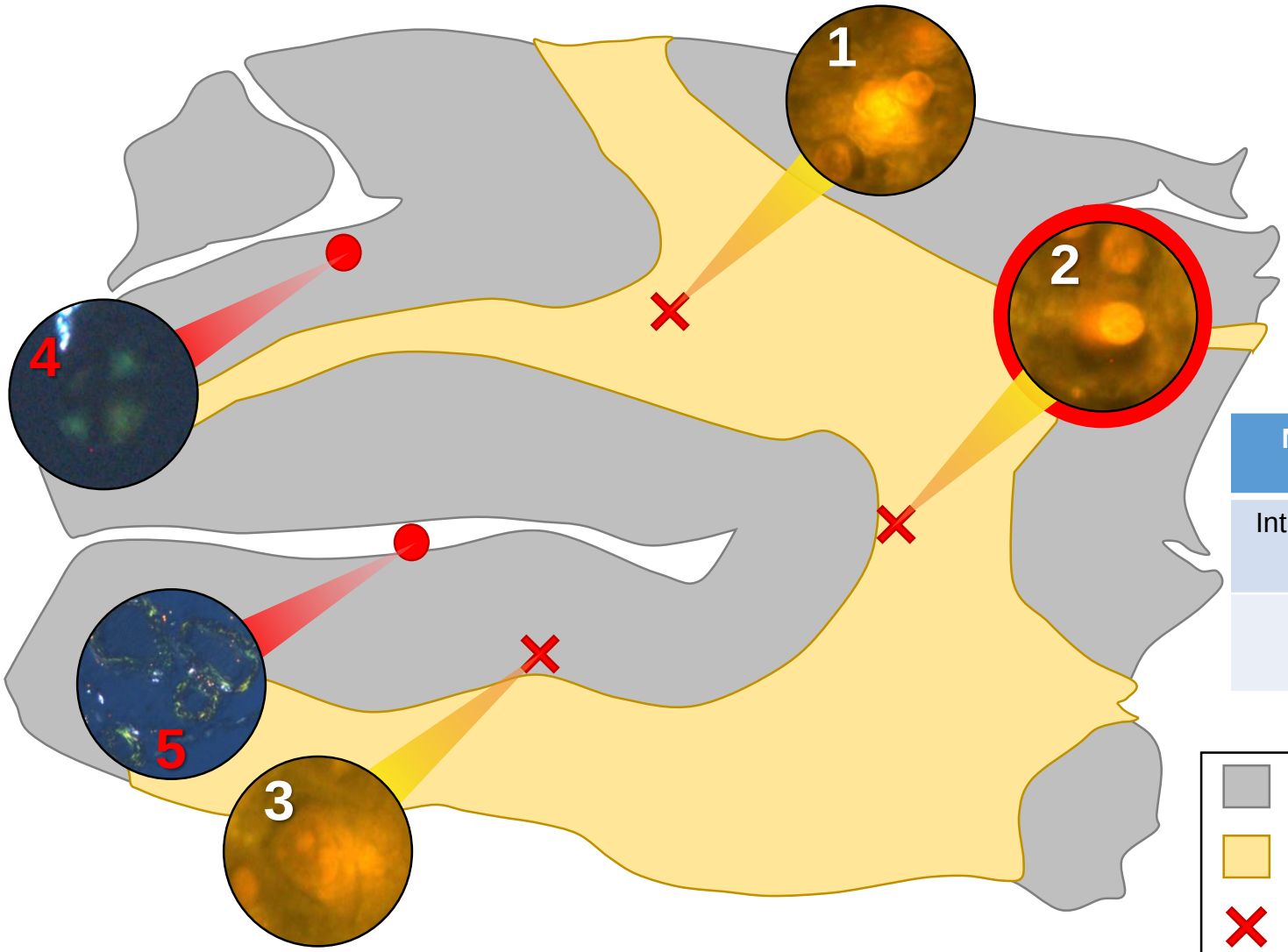
Children's Medical Safety Research Institute

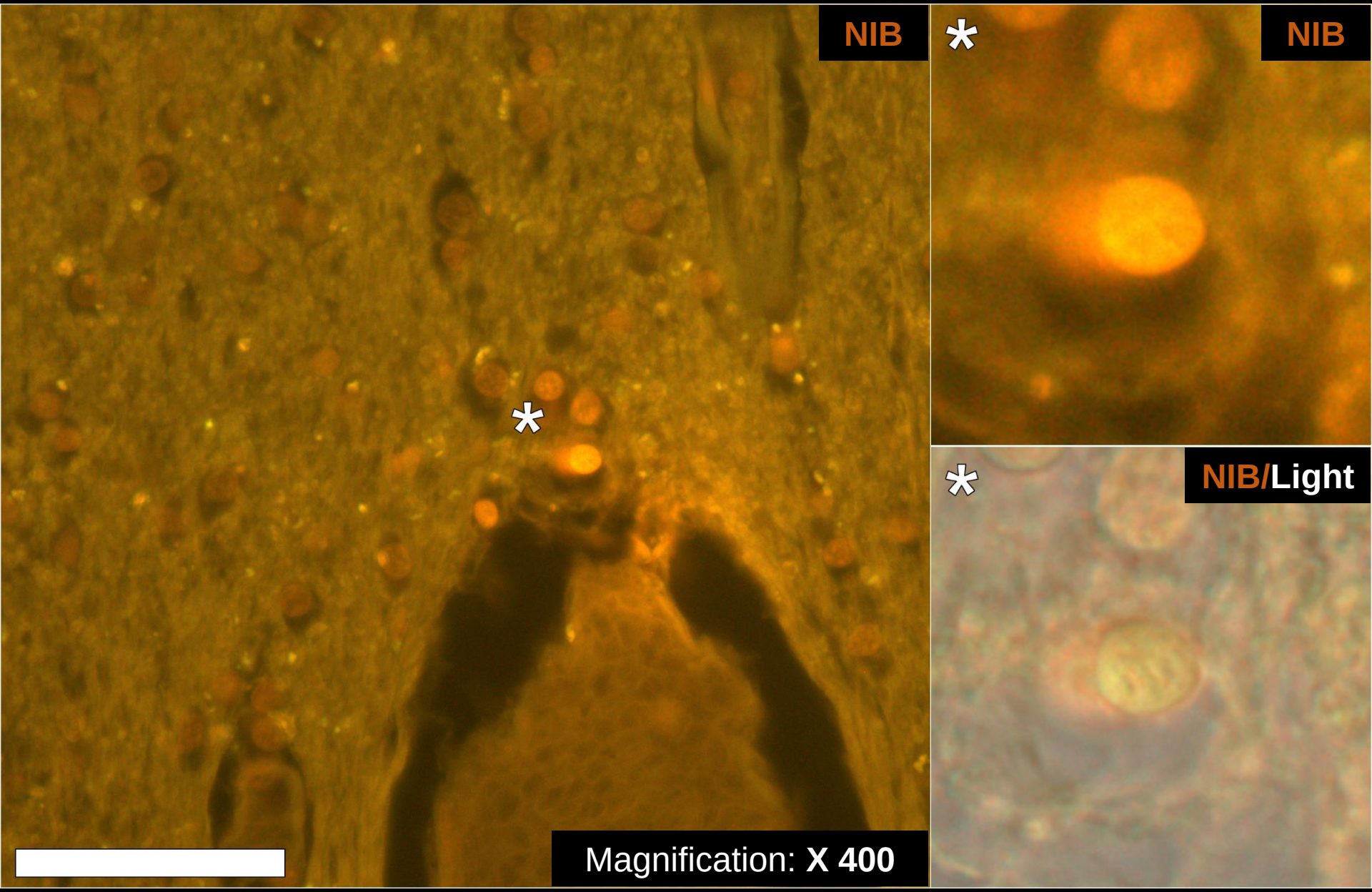


Number of AI reactive regions	
Intracellular (In)	Extracellular (Ex)
3	0

Grey matter (GM)
White matter (WM)
AI reactive
CR positive

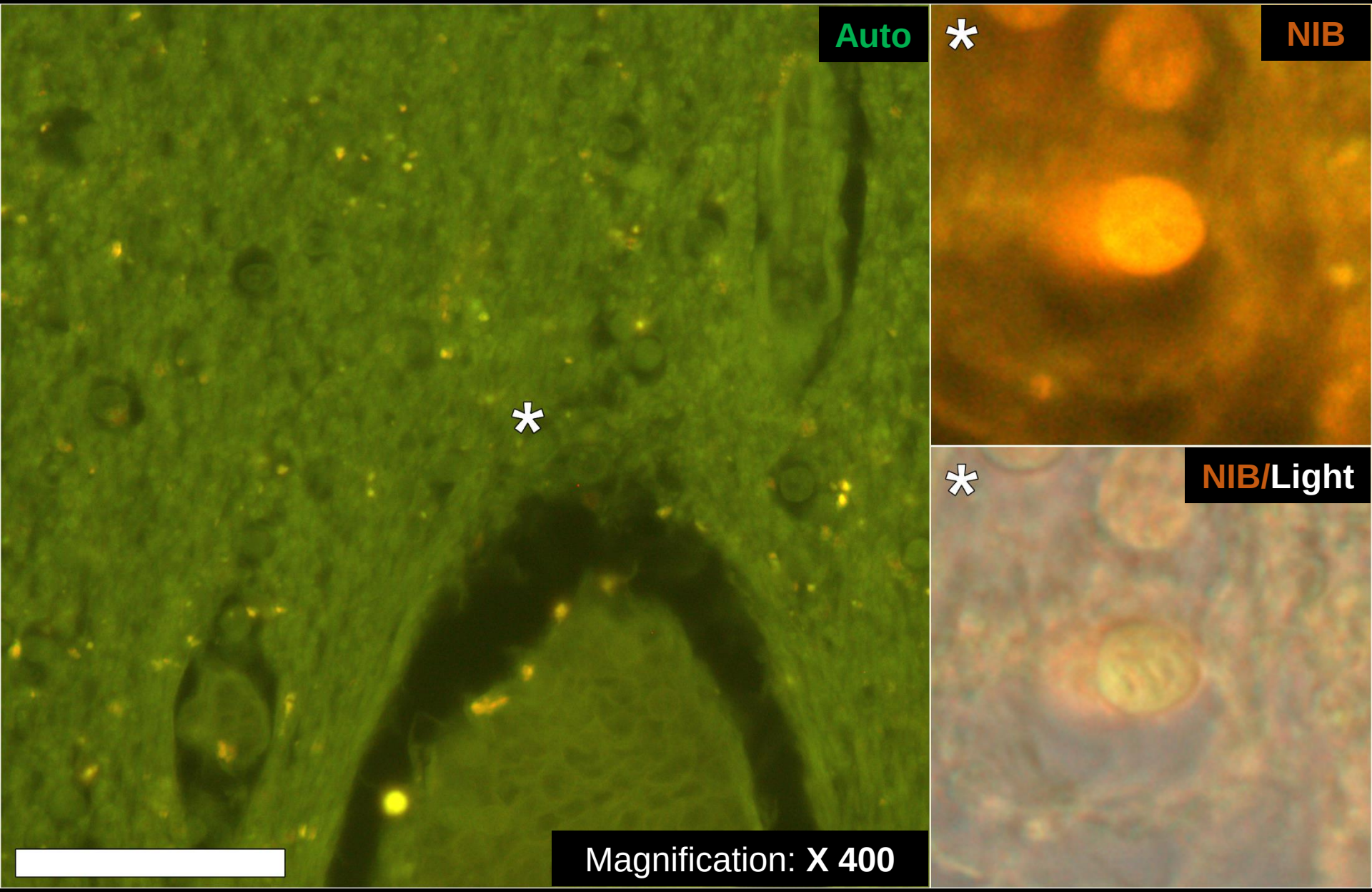
Oxford Brain Bank, Oxford, UK.

- **Parietal lobe**



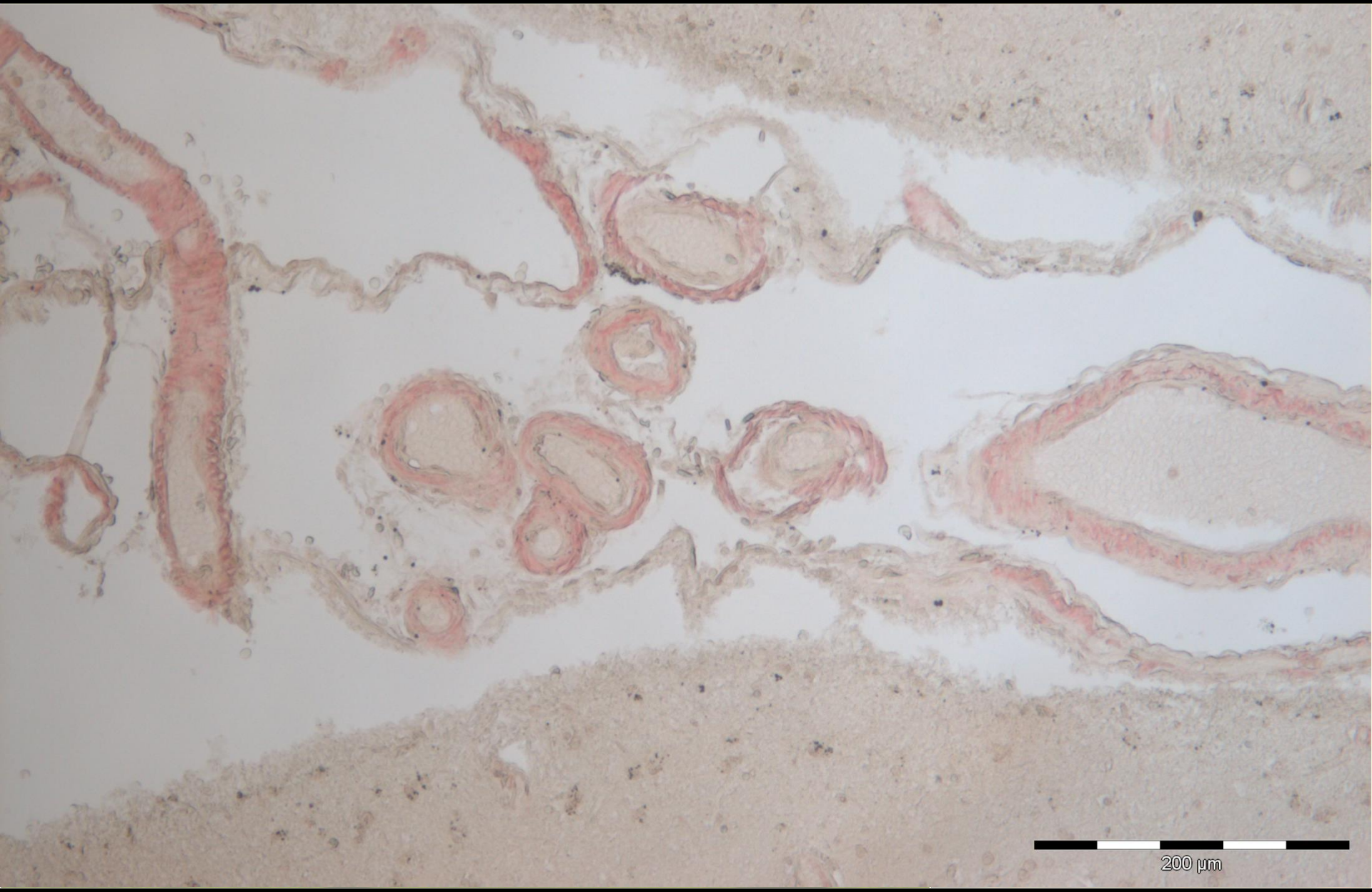
Aluminium in glial cells. (Region: #2)

- **Parietal lobe**



Aluminium in glial cells. (Region: #2)

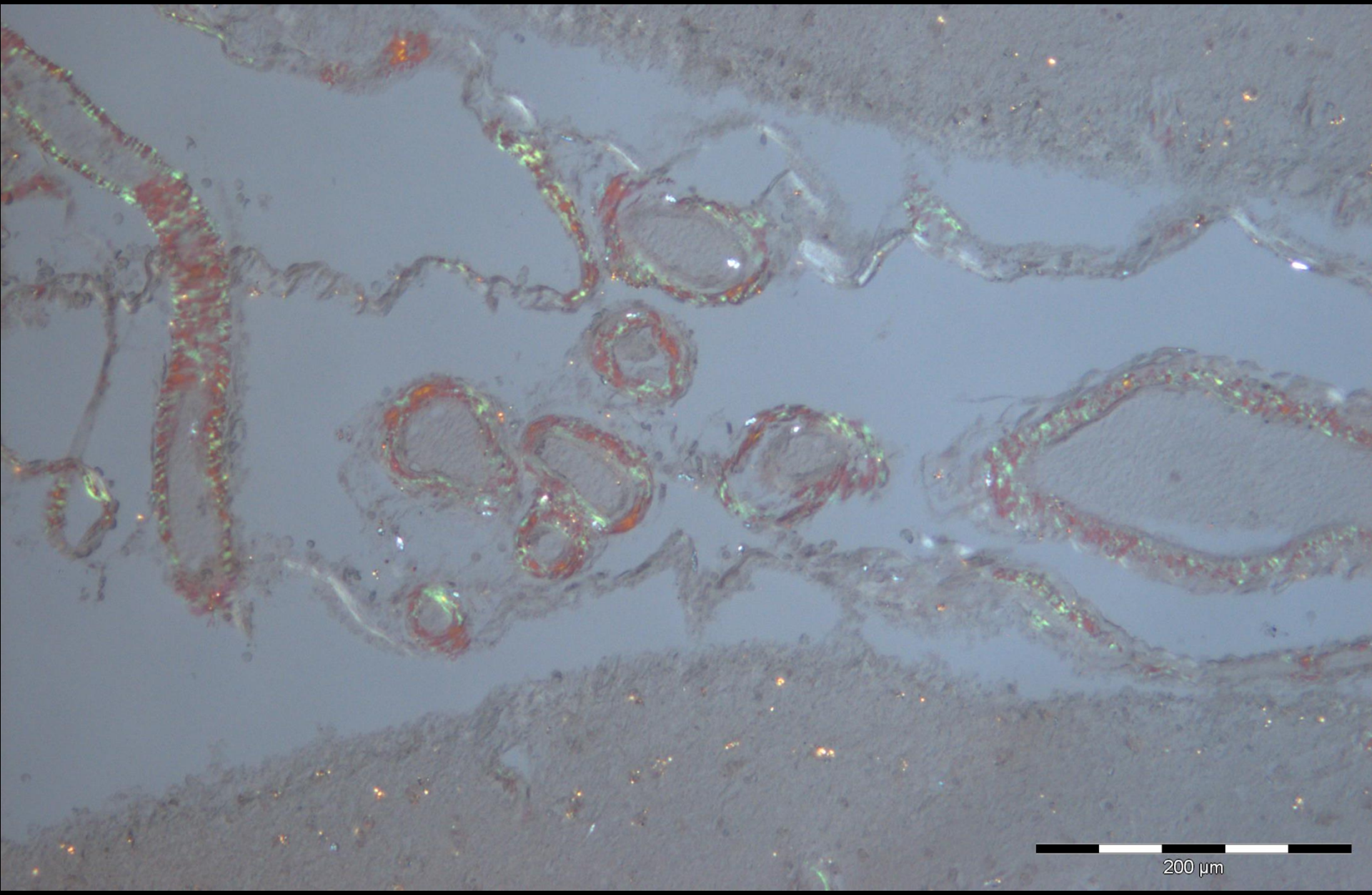
- **Parietal lobe** Light



Aluminium in glial cells. (Region: #2)

- **Parietal lobe**

½ polarised

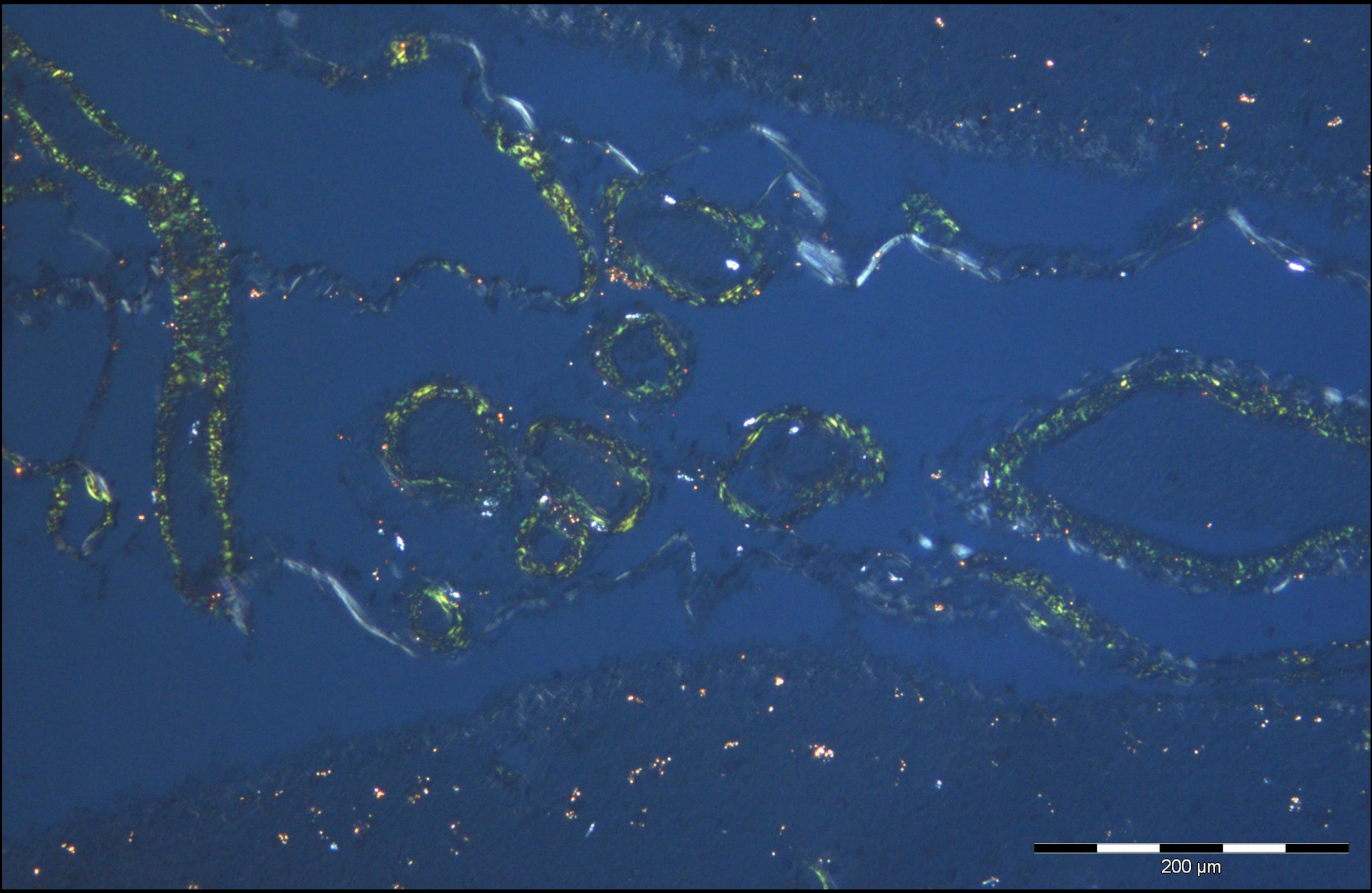


Aluminium in glial cells.

(Region: #2)

- **Parietal lobe**

Polarised



Aluminium in glial cells.

(Region: #2)

Patient ID:
NP040/2004-5
Lobe:
Temporal

CASE STUDY 1

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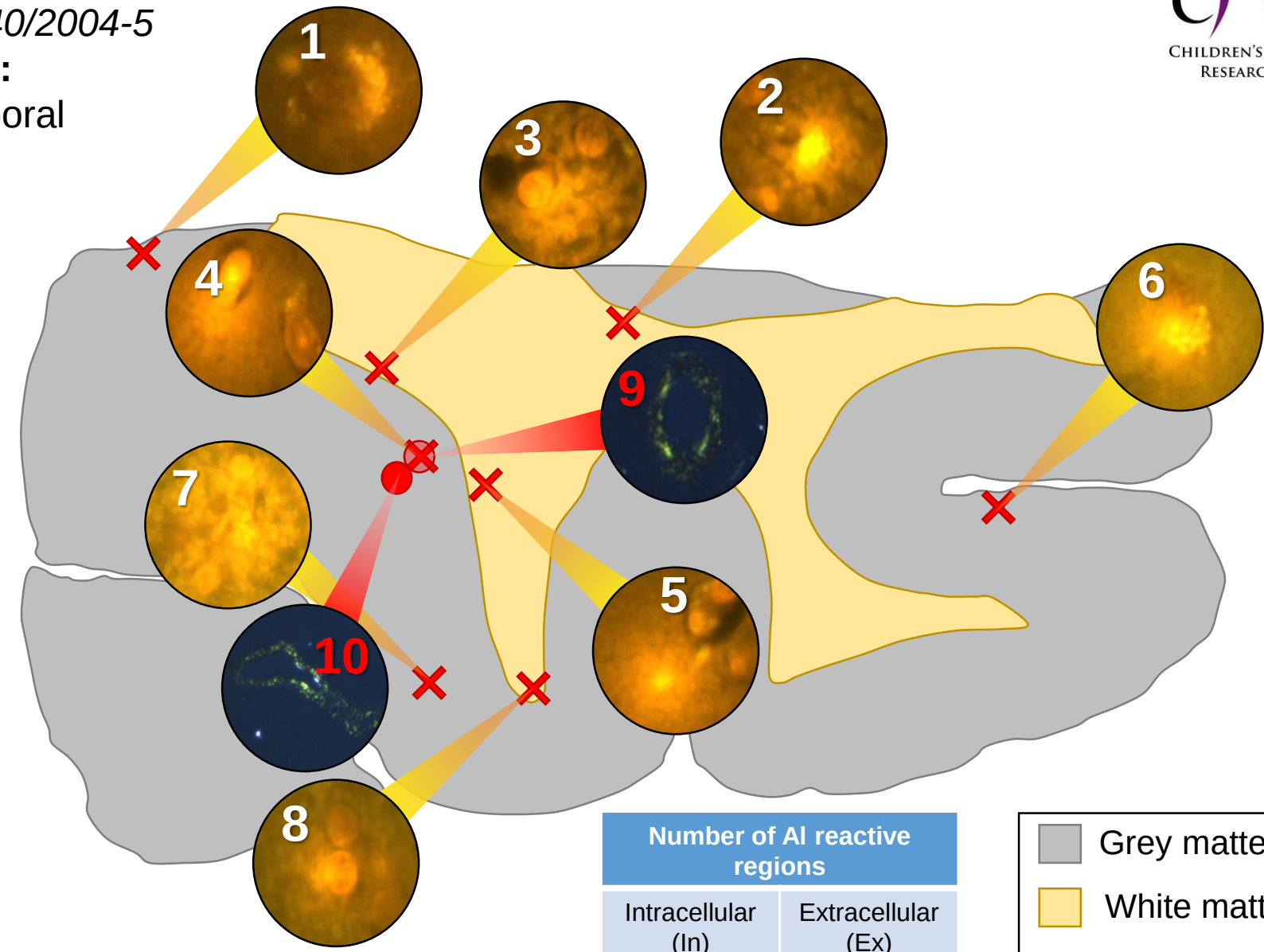
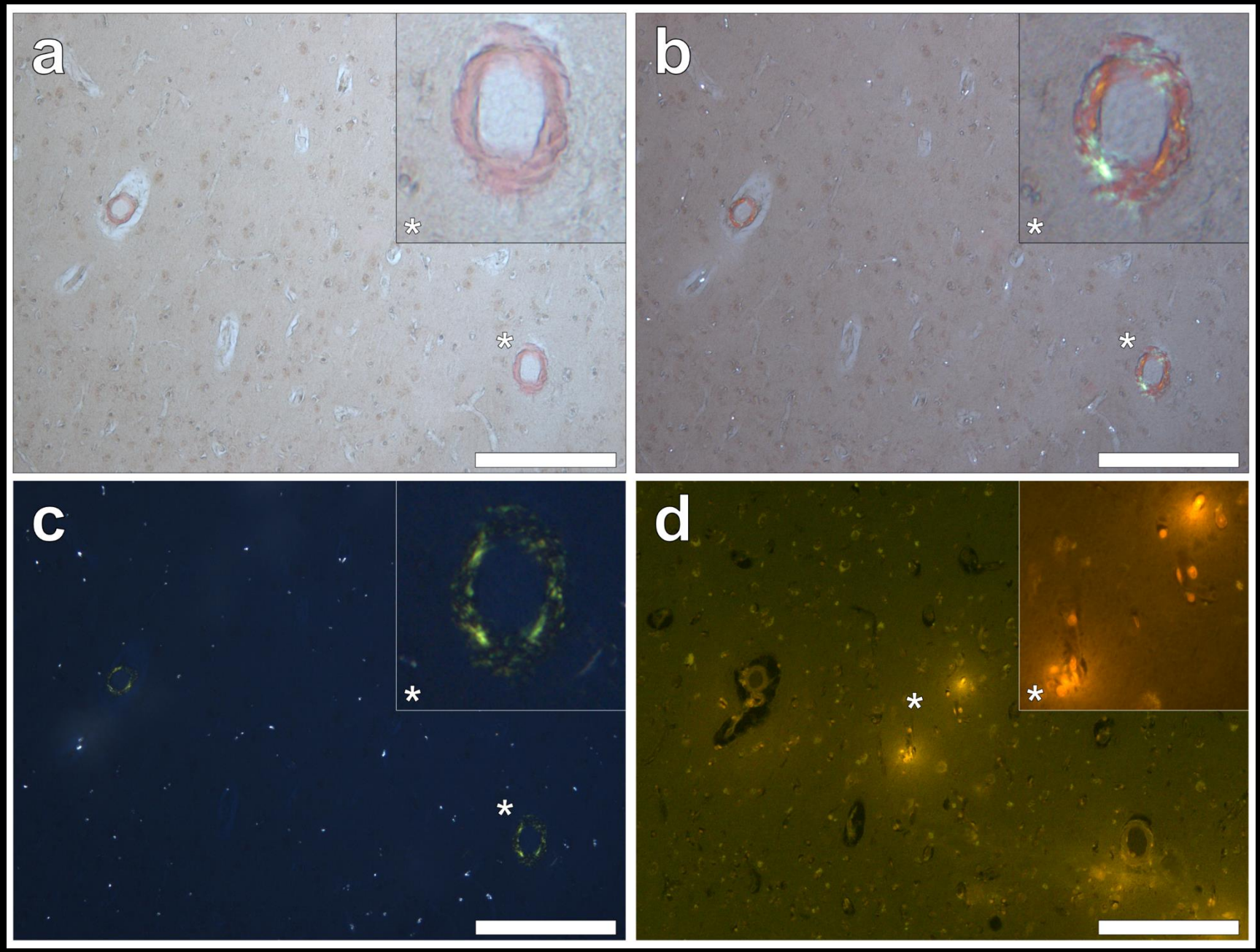


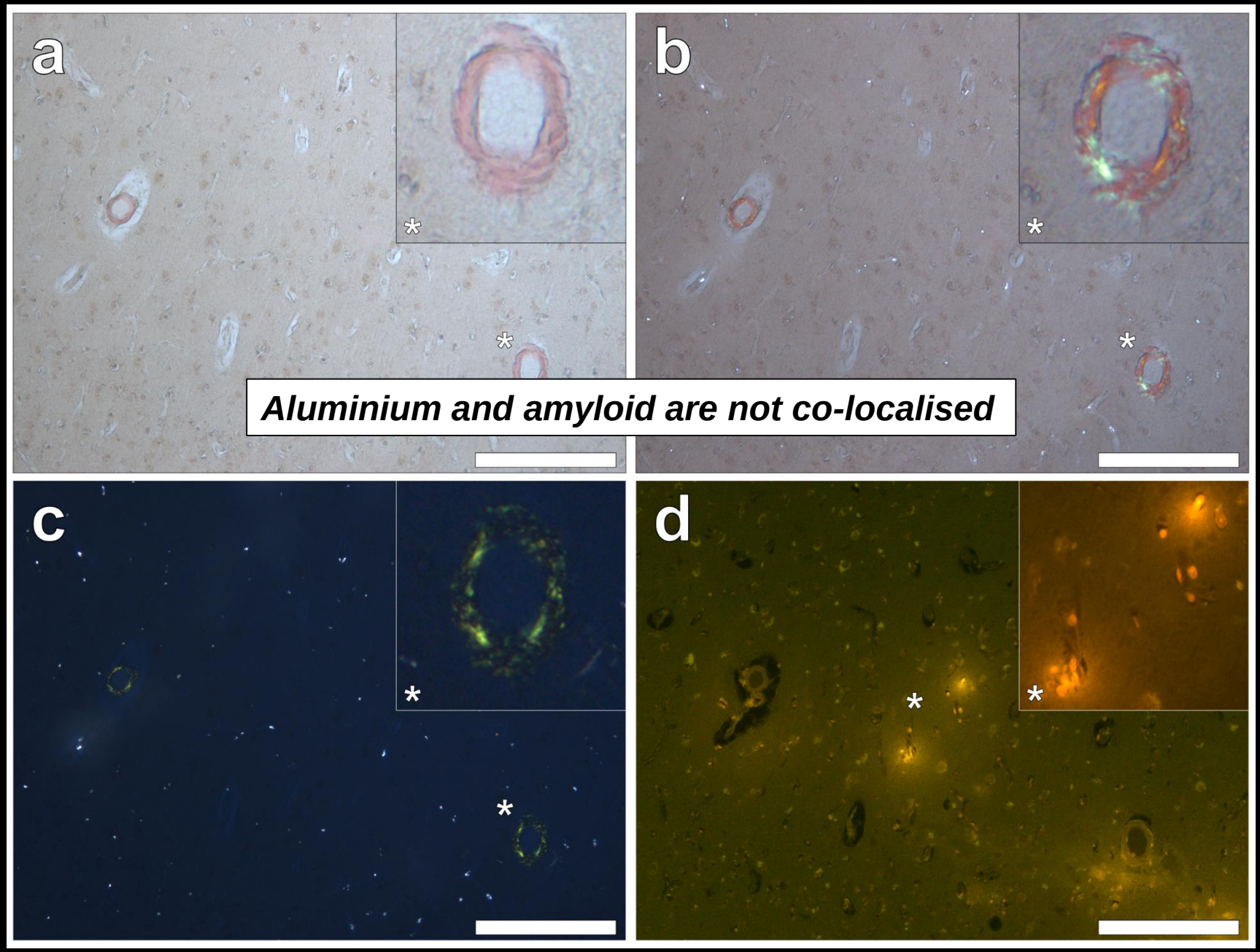
Number of AI reactive regions	
Intracellular (In)	Extracellular (Ex)
6	2

- Grey matter (GM)
- White matter (WM)
- AI reactive
- CR positive

Oxford Brain Bank, Oxford, UK.



a: Congo red, light, (b): ½ polarised, (c): polarised, (d): lumogallion staining.



a: Congo red, light, (b): ½ polarised, (c): polarised, (d): lumogallion staining.

CASE STUDY 2:

Camelford Incident:

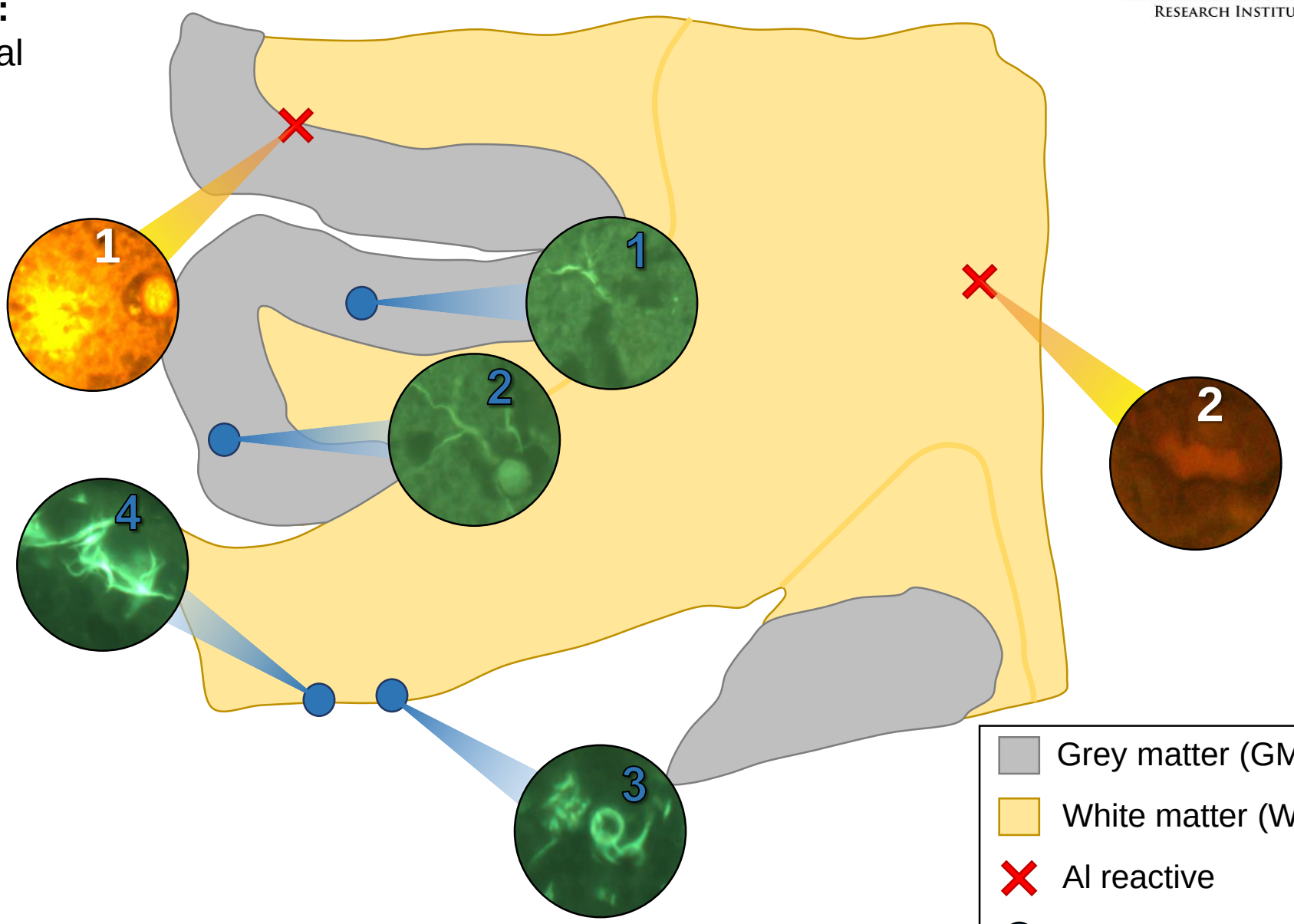
Epilepsy

- 60-year-old male donor exposed to aluminium in contaminated drinking water.
- Neuropathological examination revealed apparent calcification of brain tissue.
- Lumogallion, Congo red and thioflavin S (ThS) staining performed on brain tissue sections.

Patient ID:
PB1286-10
Lobe:
Frontal

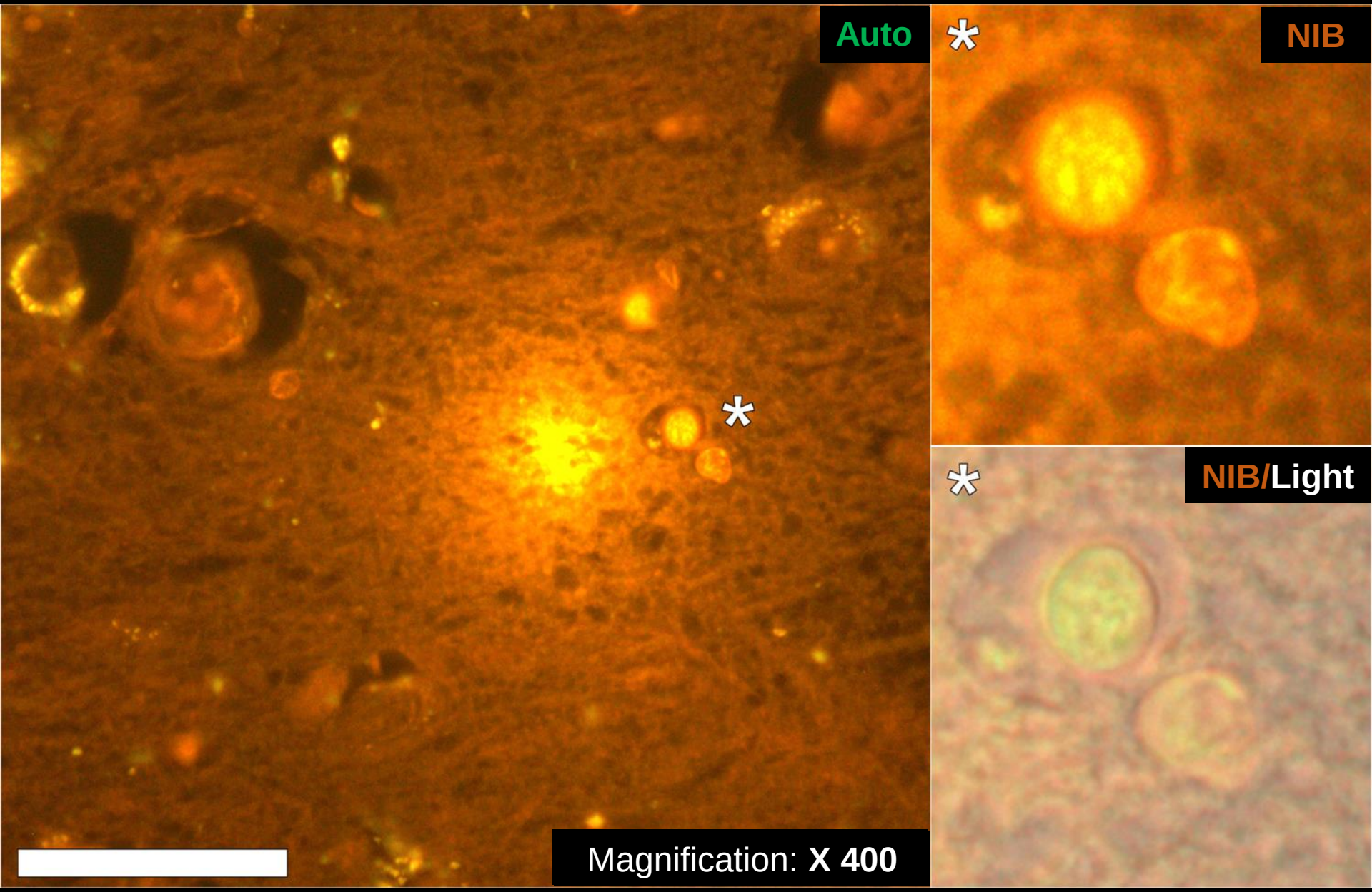
CASE STUDY 2

CMSRI
Children's Medical Safety Research Institute



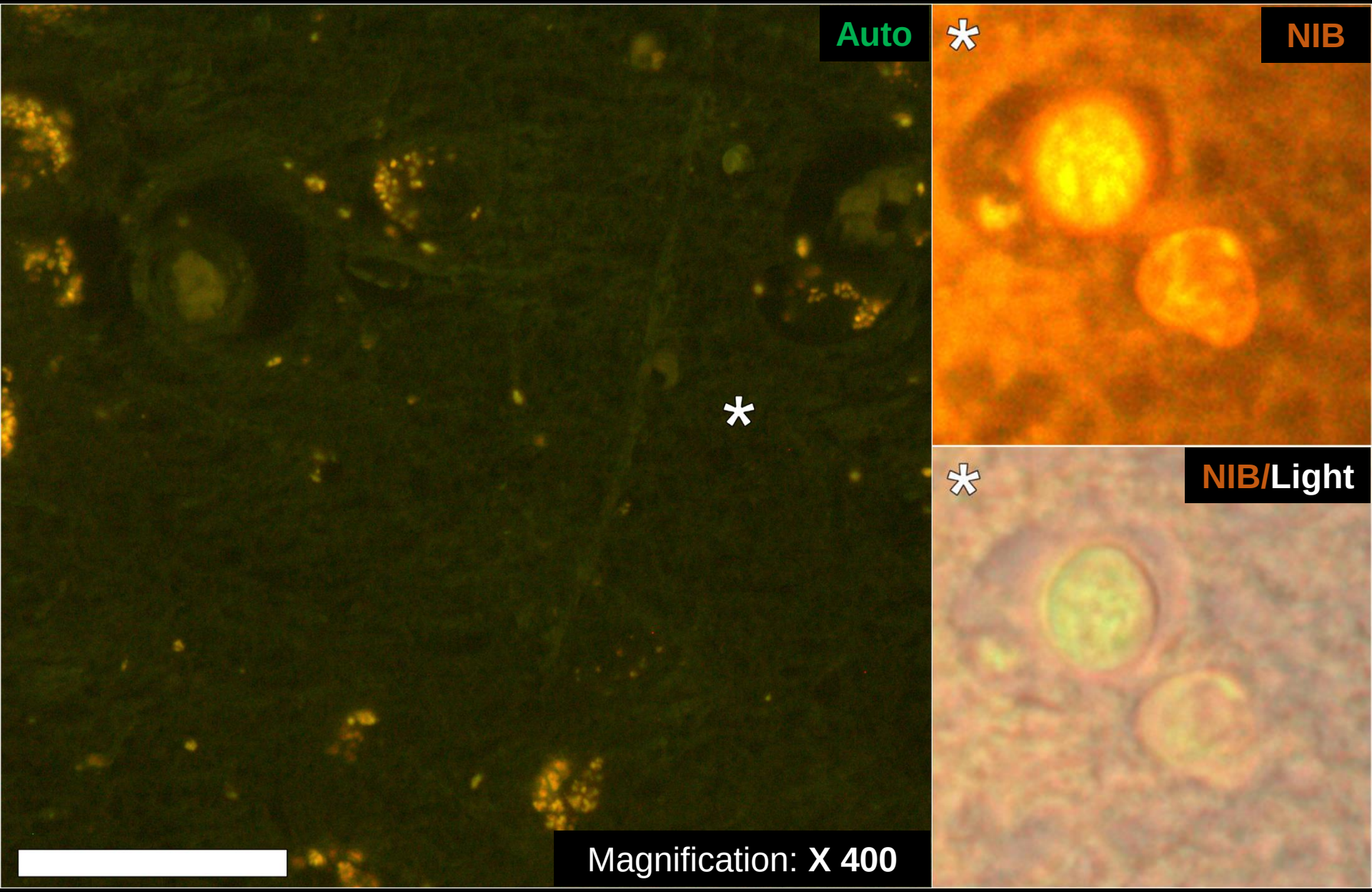
NHS, Plymouth, UK.

- **Frontal cortex**

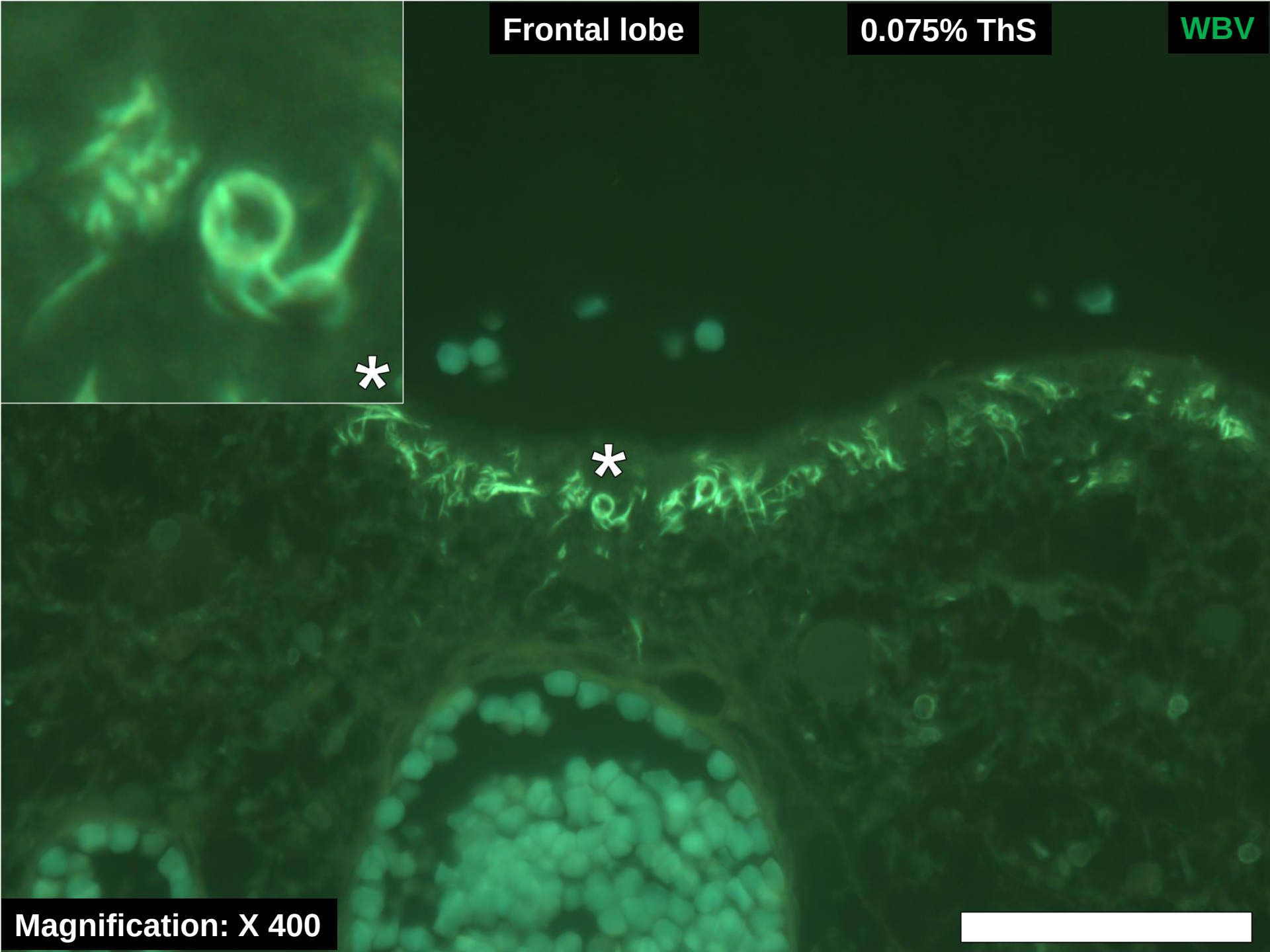


. Intracellular and extracellular aluminium (Region: #1)

- **Frontal cortex**



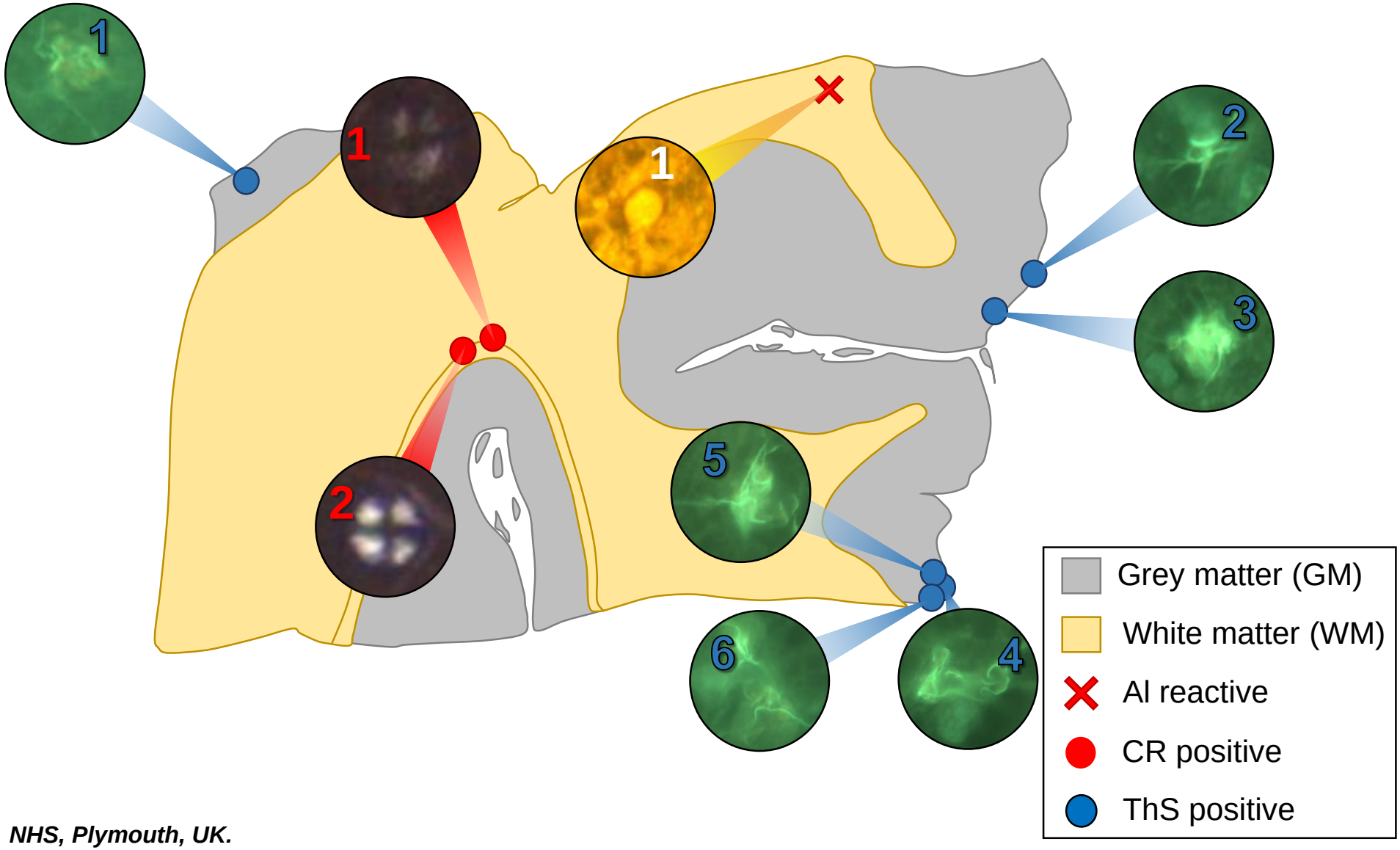
. Intracellular and extracellular aluminium (Region: #1)


Frontal lobe
0.075% ThS
WBV
Magnification: X 400

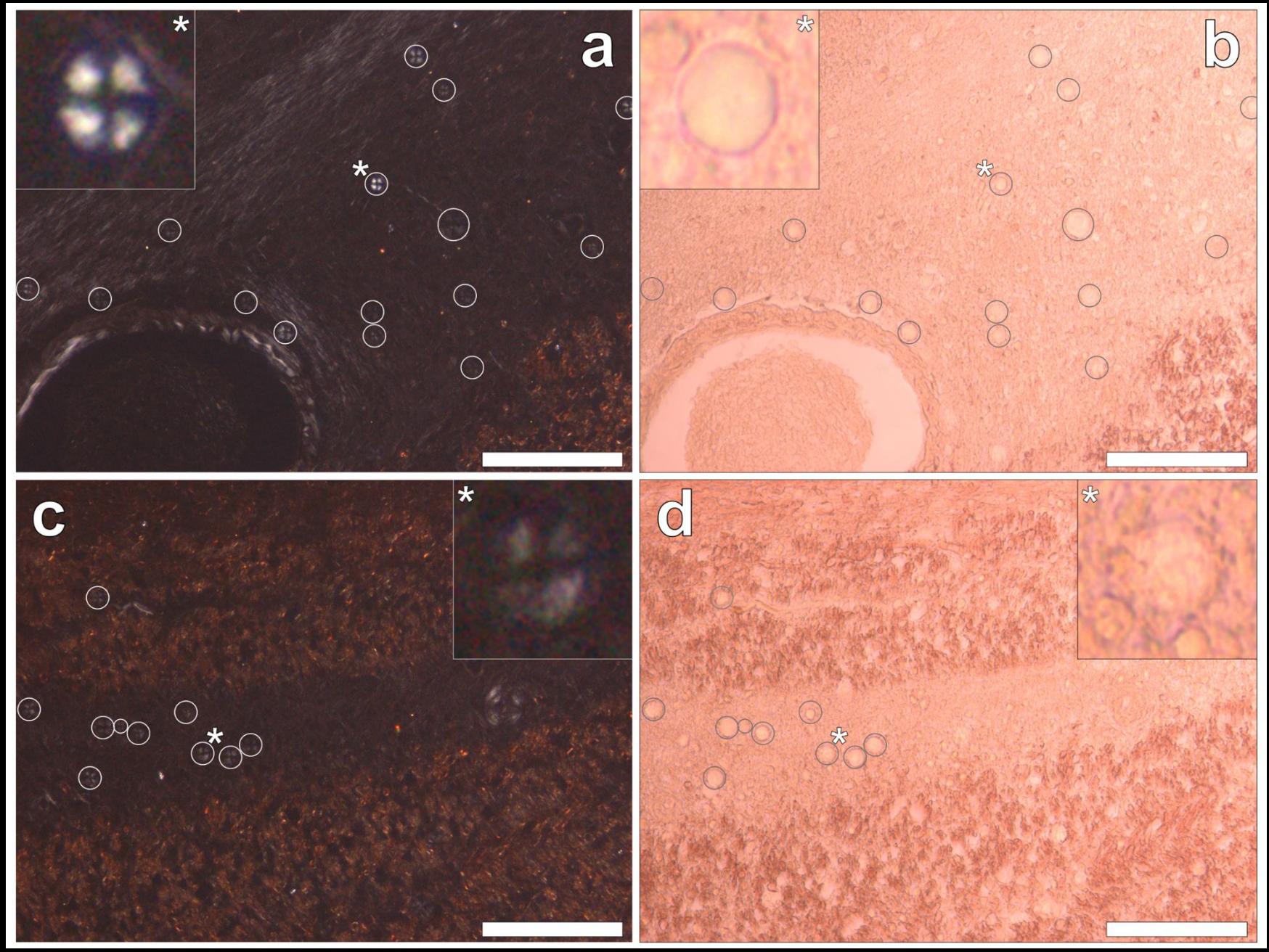
Patient ID:
PB1286-10
Lobe:
Occipital

CASE STUDY 2

CMSRI
Children's Medical Safety Research Institute



NHS, Plymouth, UK.

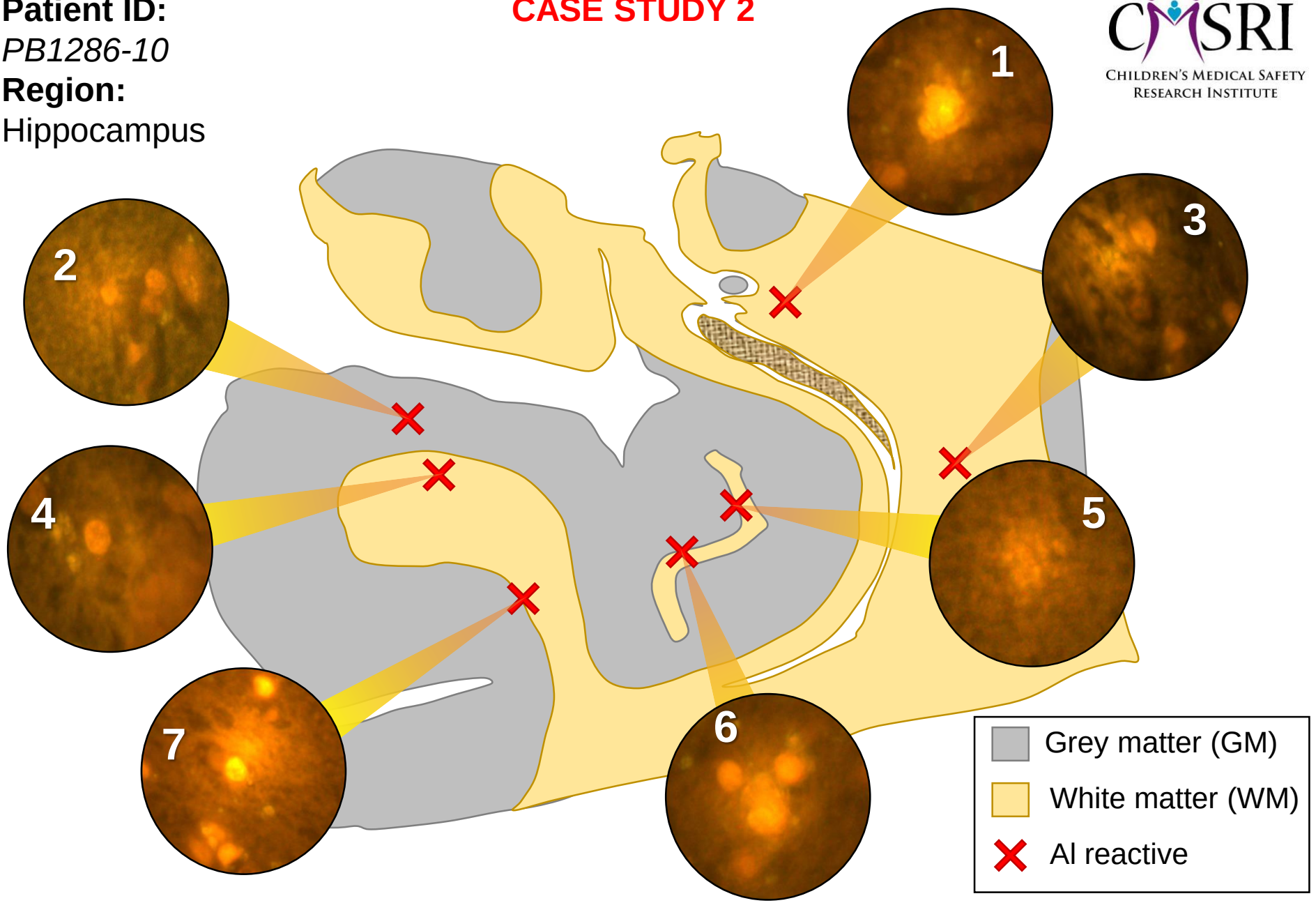


Spherulites in the stria of Gennari (visual cortex) under polarised (a & c) and bright field (b & d) illumination.

Patient ID:
PB1286-10
Region:
Hippocampus

CASE STUDY 2

CMSRI
Children's Medical Safety Research Institute

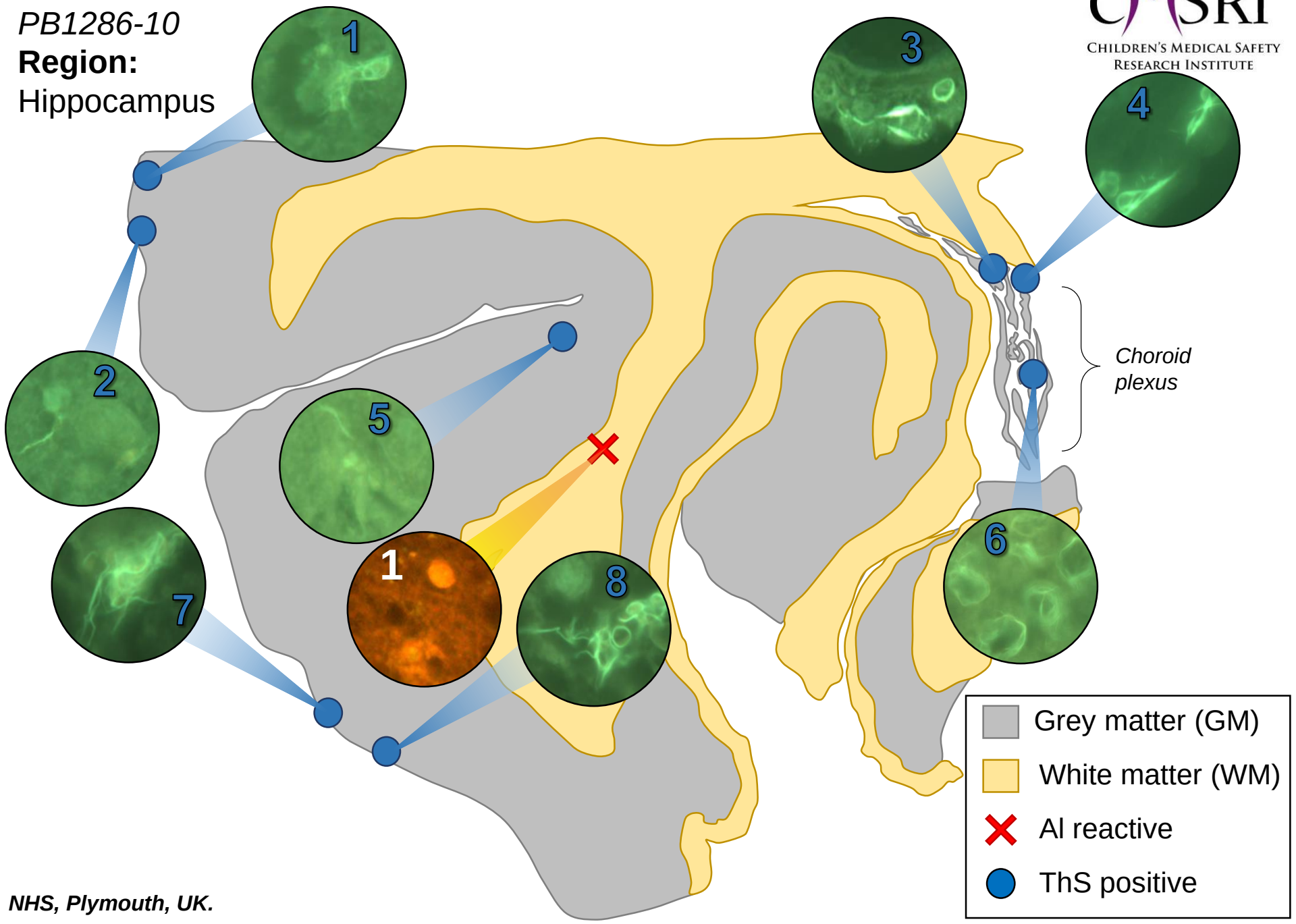


NHS, Plymouth, UK.

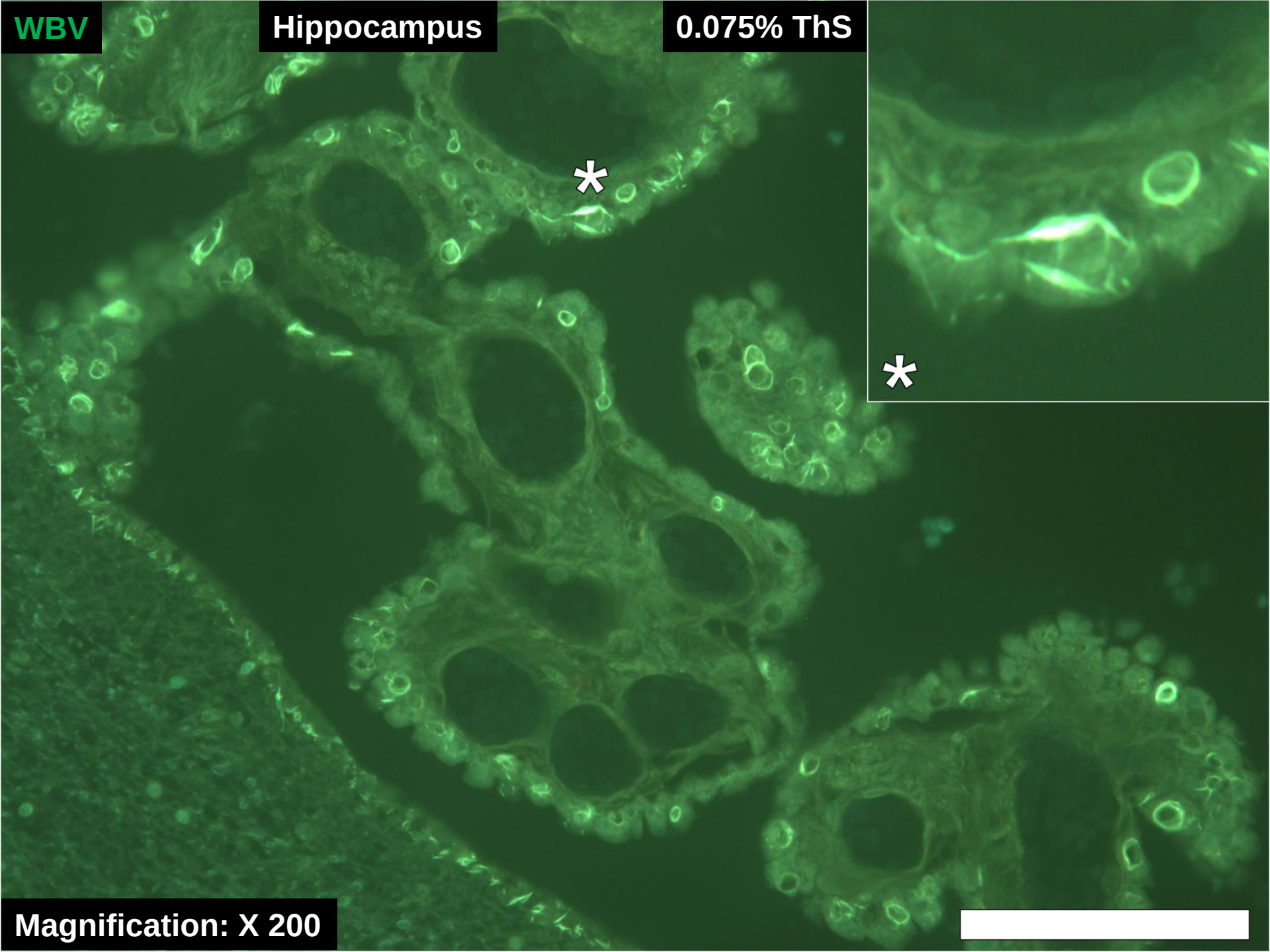
Patient ID:
PB1286-10
Region:
Hippocampus

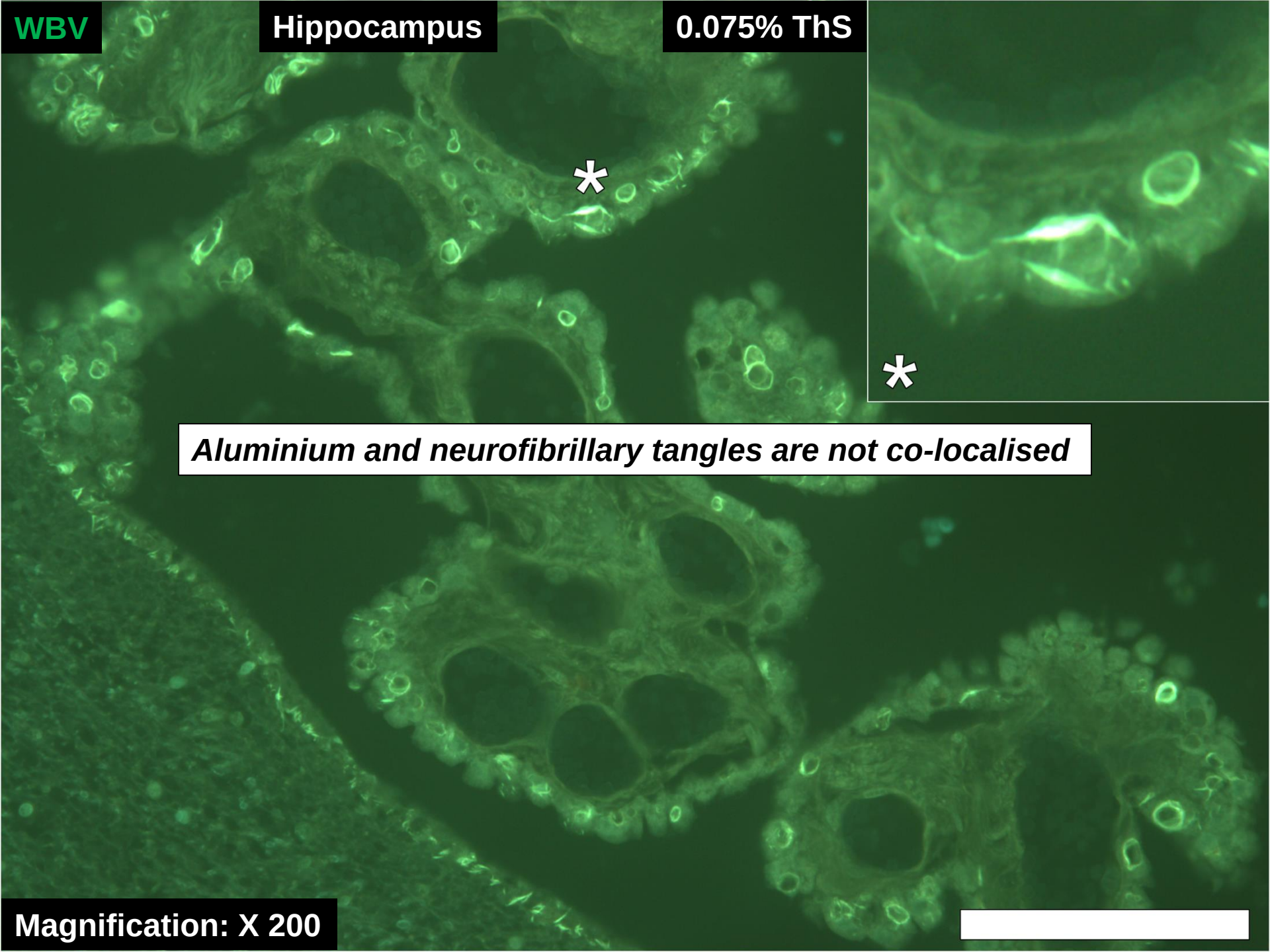
CASE STUDY 2

CMSRI
Children's Medical Safety Research Institute



NHS, Plymouth, UK.


WBV
Hippocampus
0.075% ThS
Magnification: X 200


WBV
Hippocampus
0.075% ThS
*
*
Aluminium and neurofibrillary tangles are not co-localised
Magnification: X 200

Conclusions

- Aluminium was predominantly intracellular in both cases and found mainly in non-neuronal cells.
- **CASE STUDY 1:** Severe CAA with no evidence for co-localisation of amyloid with aluminium.
- Presence of aluminium in the choroid plexus supports inflammation.
- **CASE STUDY 2:** Extensive NFT deposition in cortical regions with apparent calcifications noted in the visual cortex.
- Intracellular aluminium in microglia, astrocytes and lymphocytes supports possibility of aluminium being carried into the brain.

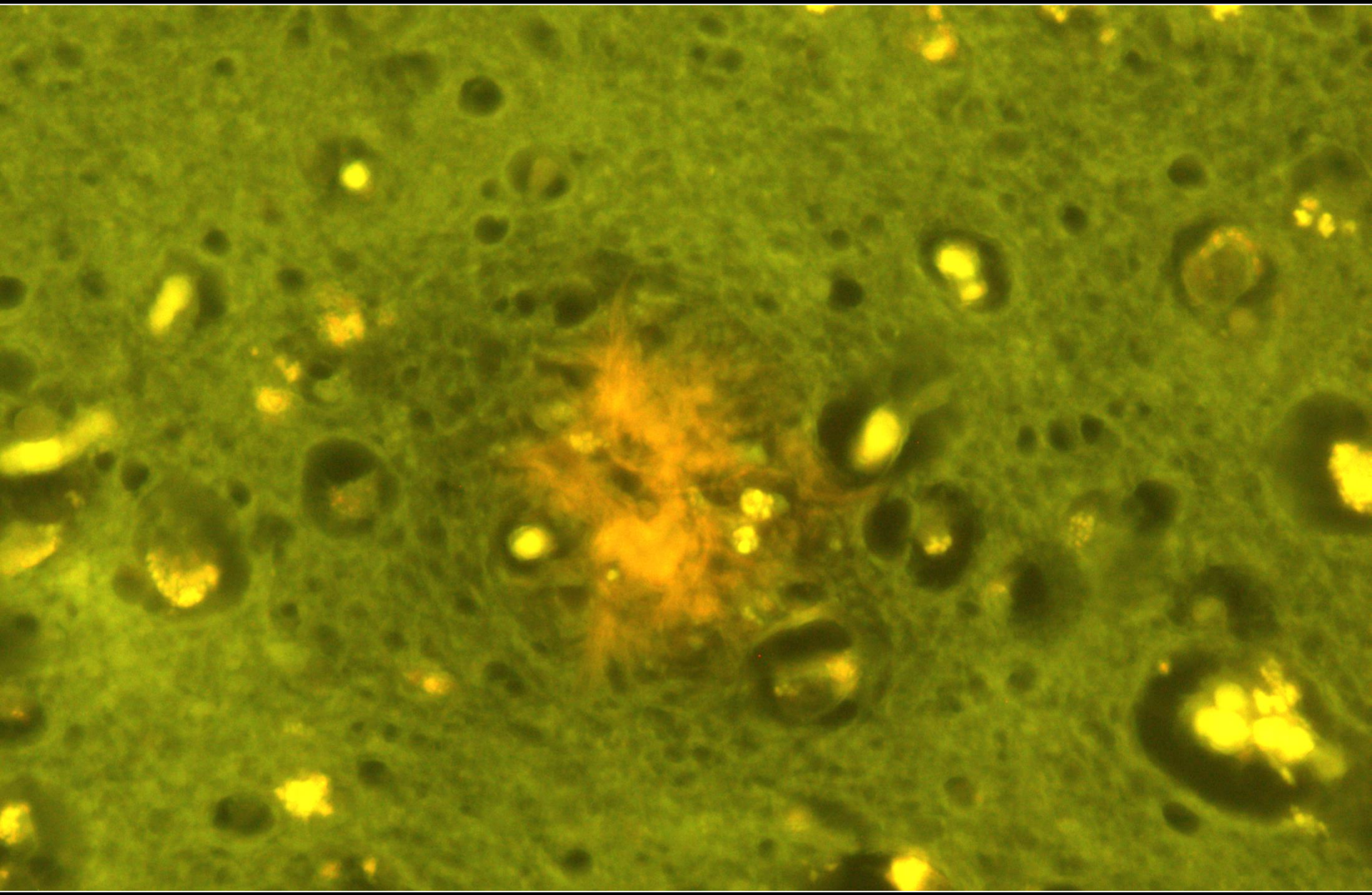
FUTURE WORK:

Colombian donor tissues:

Familial Alzheimer's disease (PSEN1 E280A)

- **Frontal lobe**

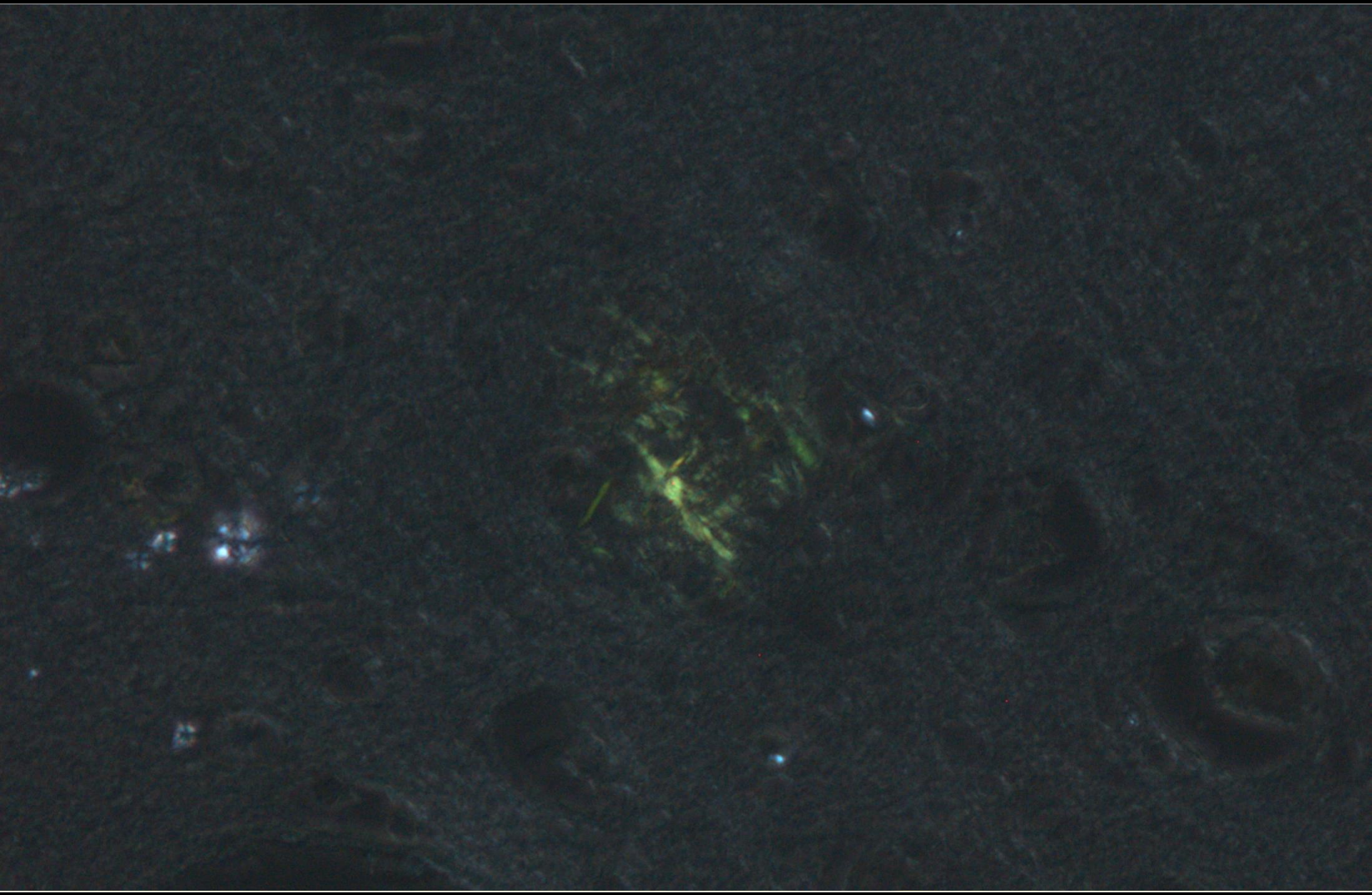
NIB / Lumo



Cortical senile plaque.

- **Frontal lobe**

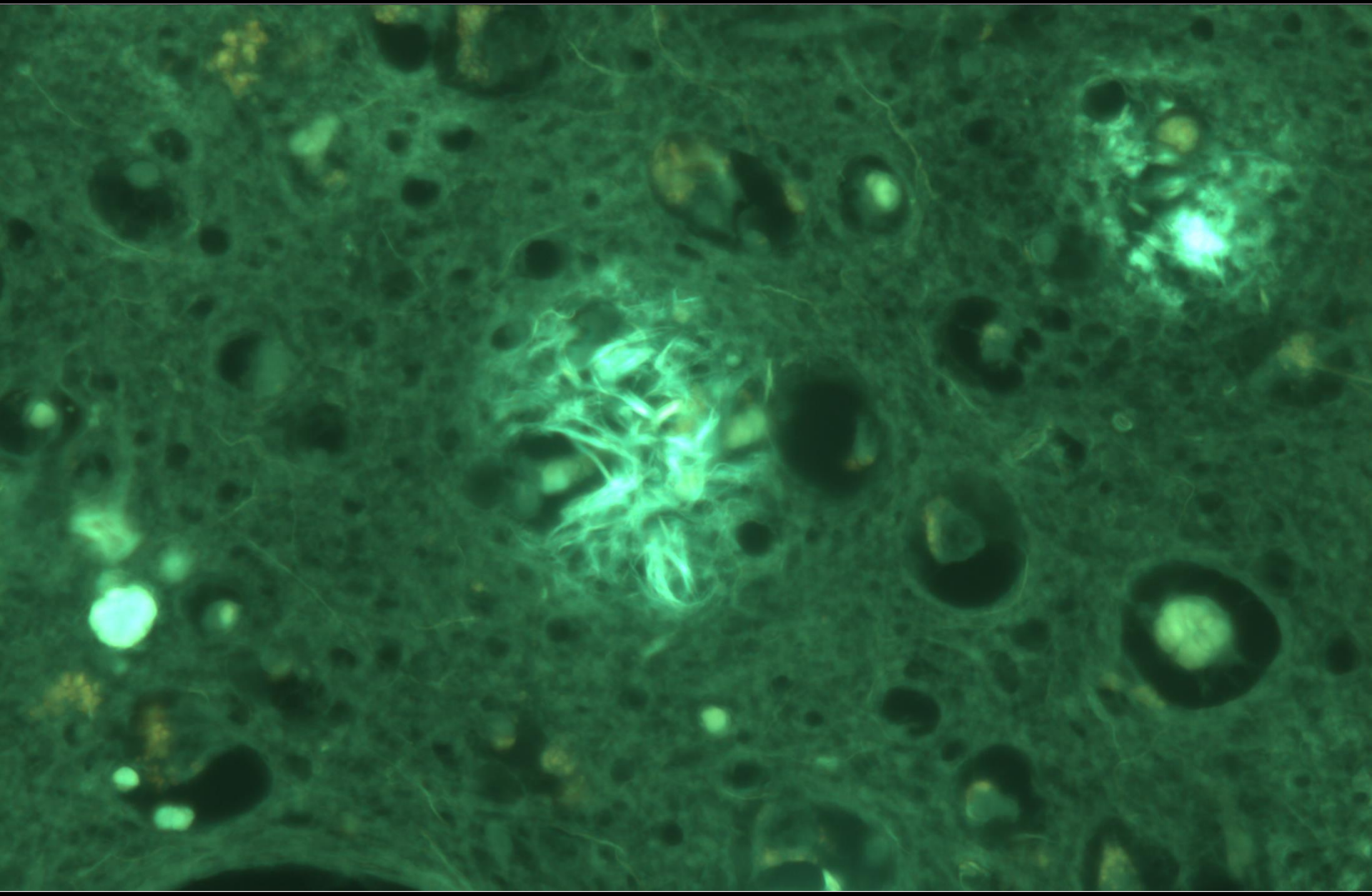
Polarised



Cortical senile plaque.

- **Frontal lobe**

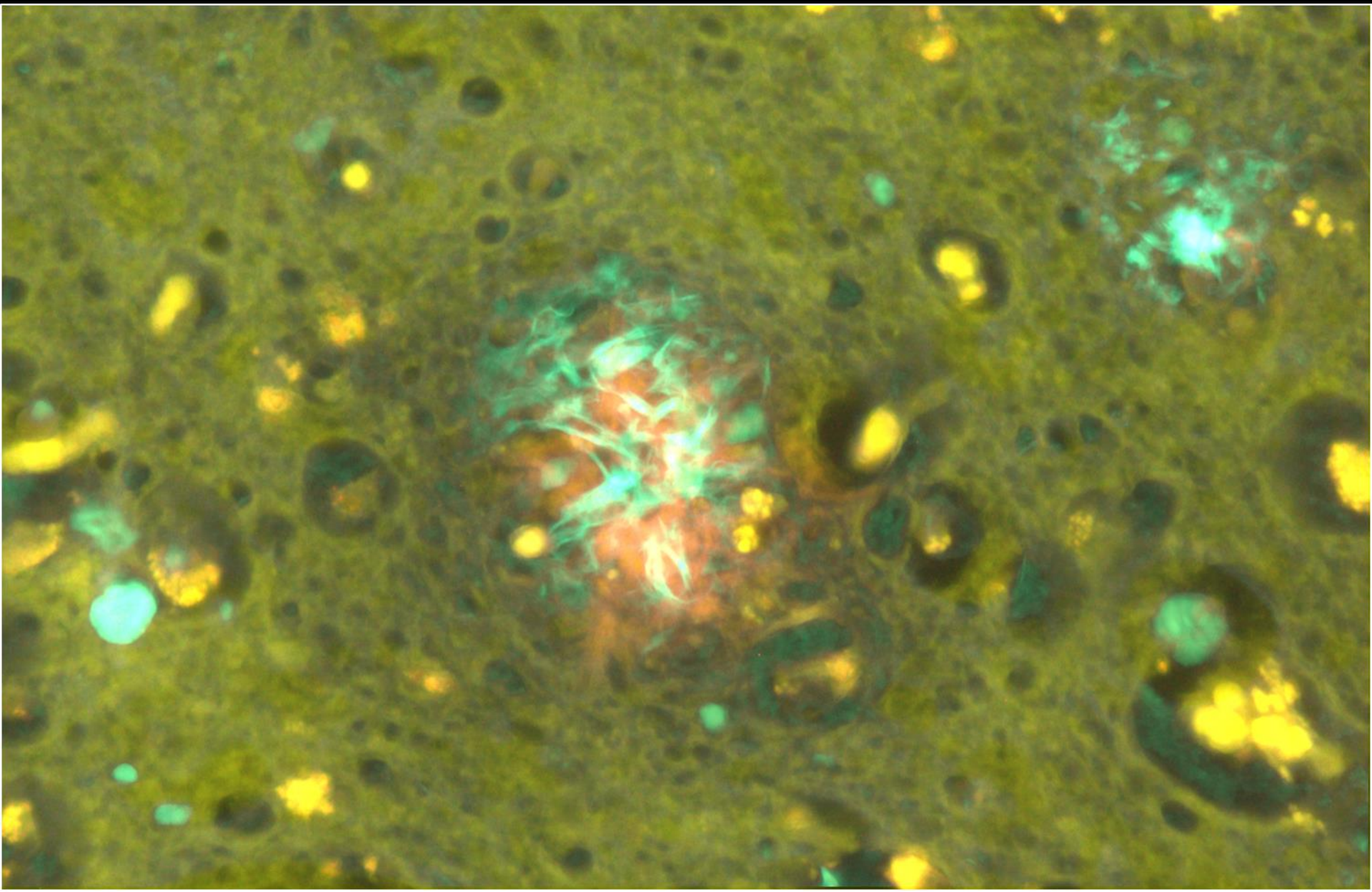
WBV / ThS



Cortical senile plaque.

- **Frontal lobe**

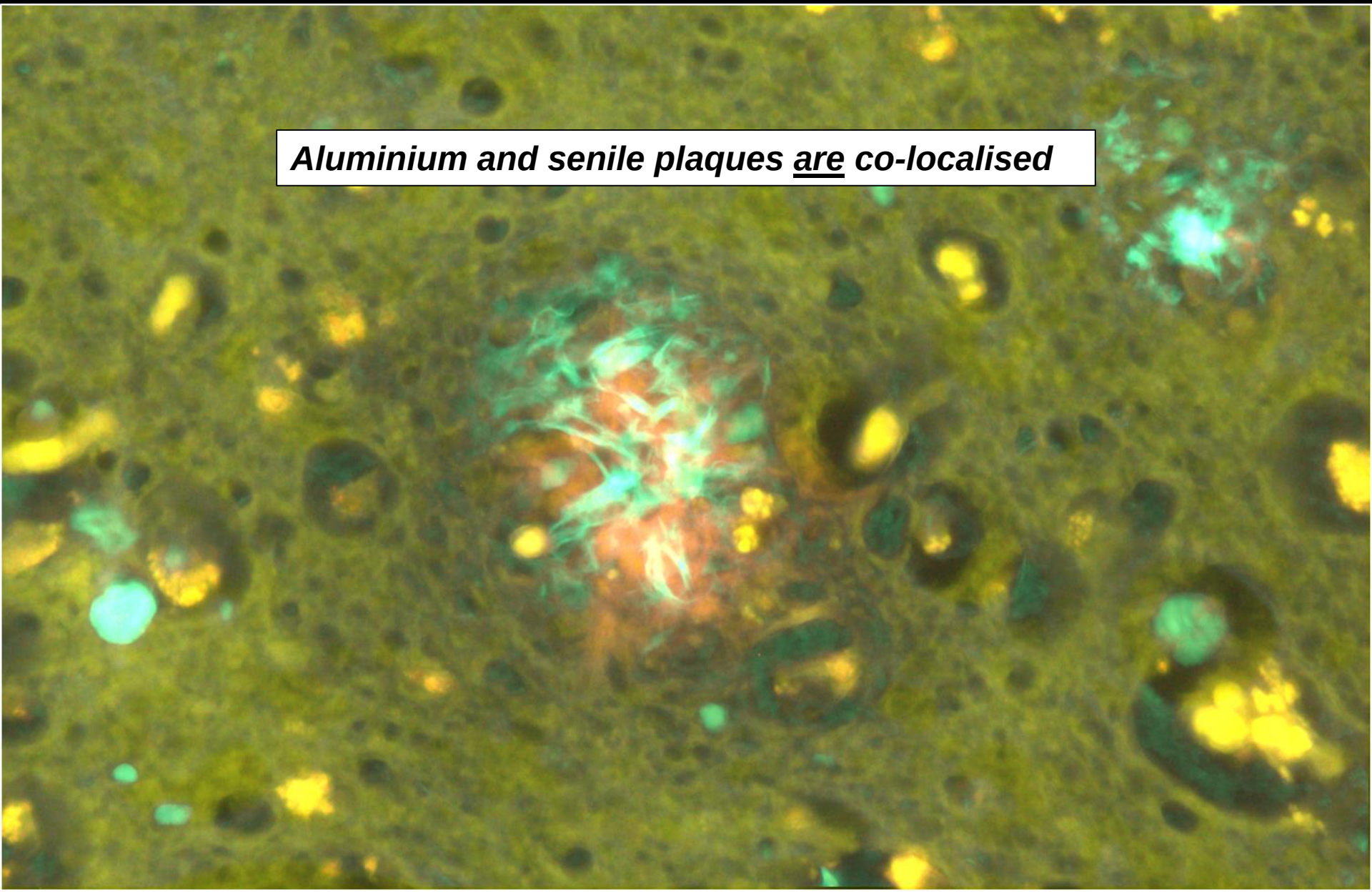
NIB / Lumo Polarised WBV / ThS



Cortical senile plaque.

- **Frontal lobe**

NIB / Lumo Polarised WBV / ThS



Cortical senile plaque.

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Acknowledgements

Funding:

- **Children's Medical Safety Research Institute (CMSRI)**

Bioinorganic chemistry of Al and Si research group:

- **Prof. Christopher Exley**
- **Jason Cottle** (3rd year medical student)
- Dr Emma Shardlow
- Isabel Rodriguez (Ph.D. candidate)

Collaborators and brain banks:

- **Dr David Hilton & Phil Edwards** (NHS, Plymouth, UK)
- **Dr Andrew King** (Kings College Hospital, London, UK)
- Oxford Brain Bank, Oxford, UK
- MS Society Tissue Bank & MRC Brain Banks, London, UK

University of Zaragoza:

- **Prof. Lluís Luján**
- Javier Asin (Ph.D. candidate)