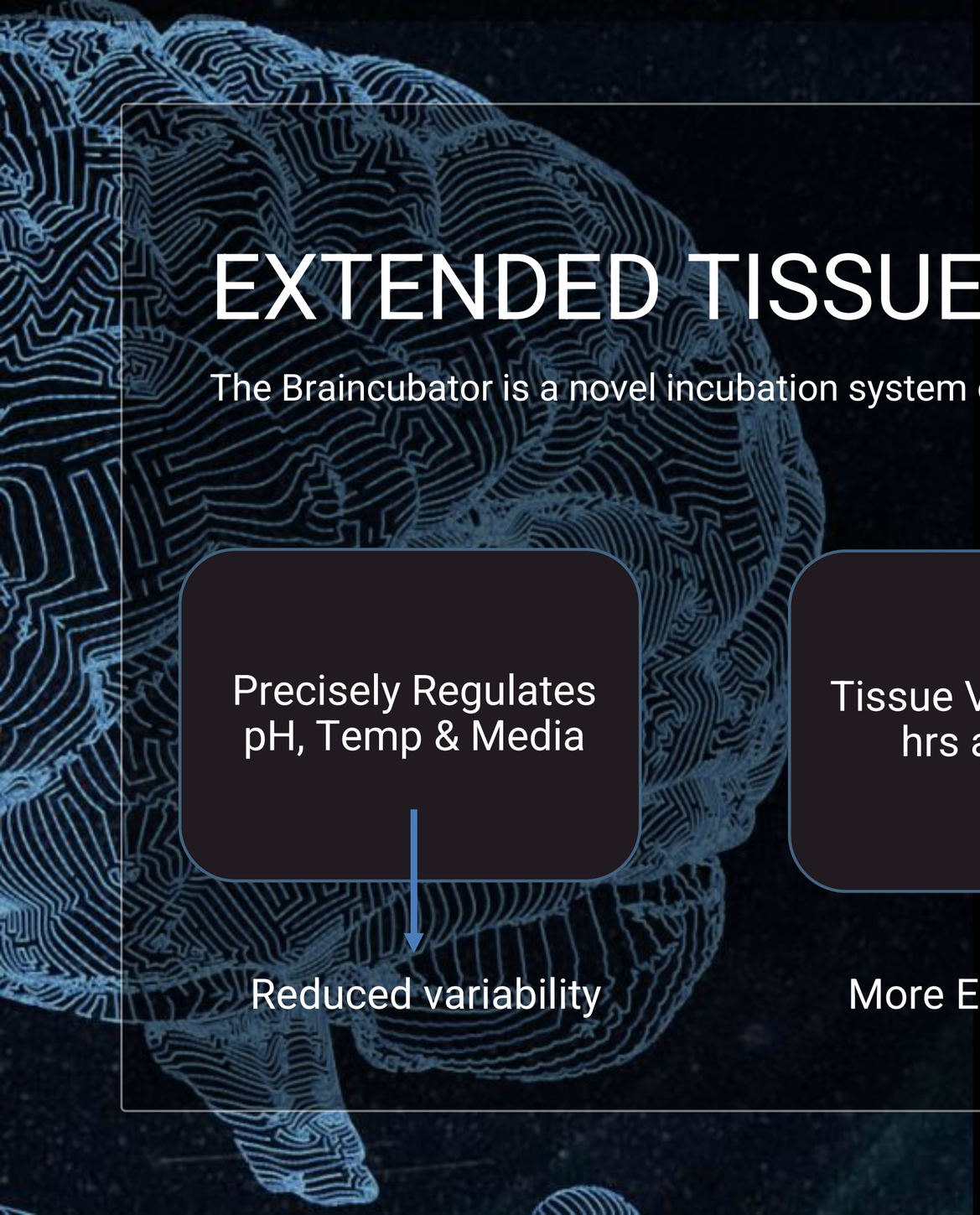
BRAINCUBATOR™

Extending the viability of tissue slices

www.braincubator.com.au



BRAINCUBATOR ™

EXTENDED TISSUE LIFESPAN

The Braincubator is a novel incubation system capable of ***extending the lifespan of thin slice tissue***

Precisely Regulates pH, Temp & Media

↓

Reduced variability

Tissue Viability of 30 hrs and more

↓

More Experiments

Half the Number of Animals Sacrificed

↓

Less Animals

RECOVERY INCUBATION SYSTEM

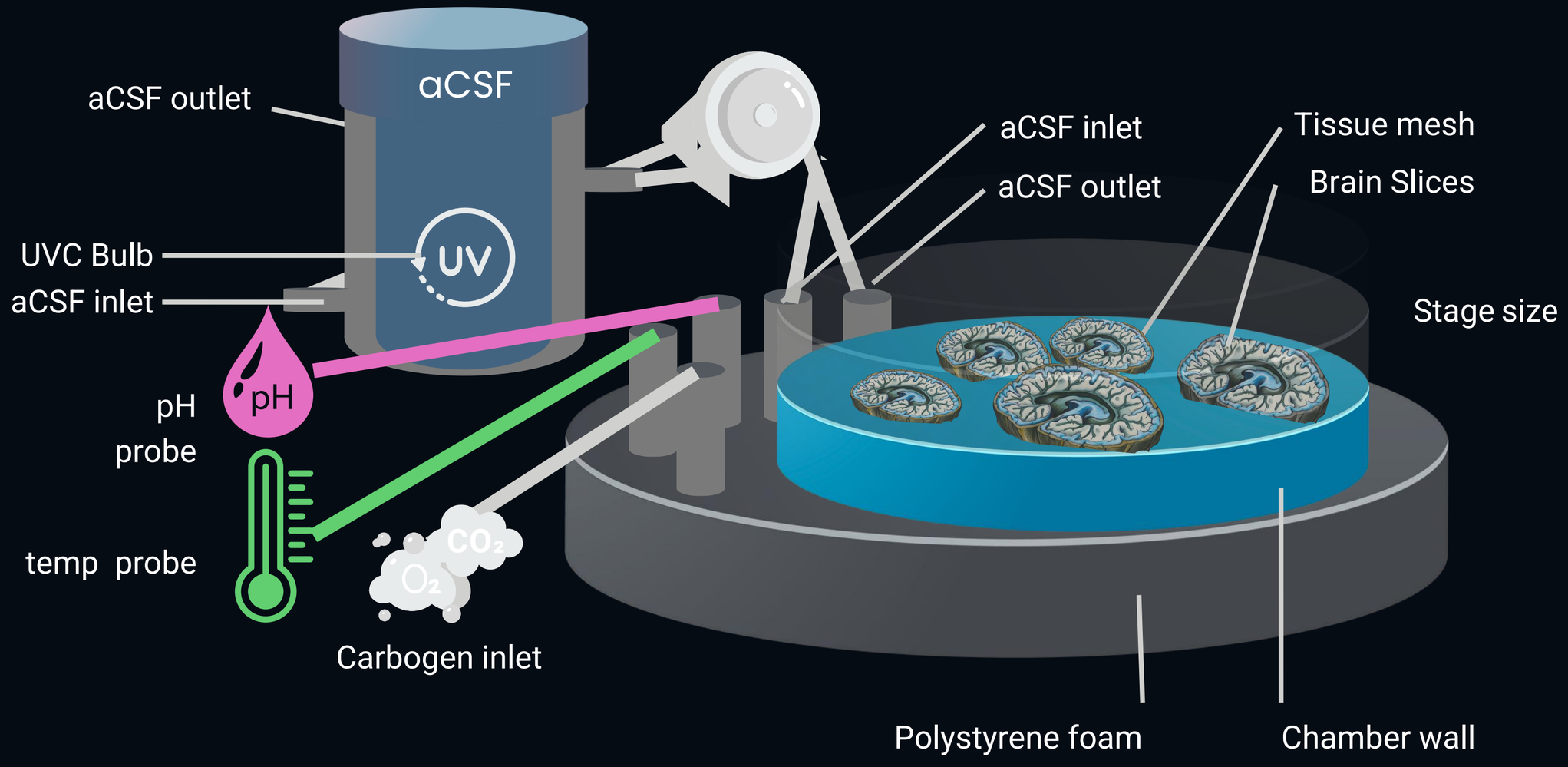
MONITORING & STANDARDISING INCUBATION CONDITIONS

CONTINUOUS aCSF CIRCULATION

pH MONITORING

TEMPERATURE CONTROL

CARBOGEN DIFFUSER



BRAINCUBATOR™ system - Schematic Diagram

BACTERIA COUNTS

STANDARD CONTROL VS BRAINCUBATOR

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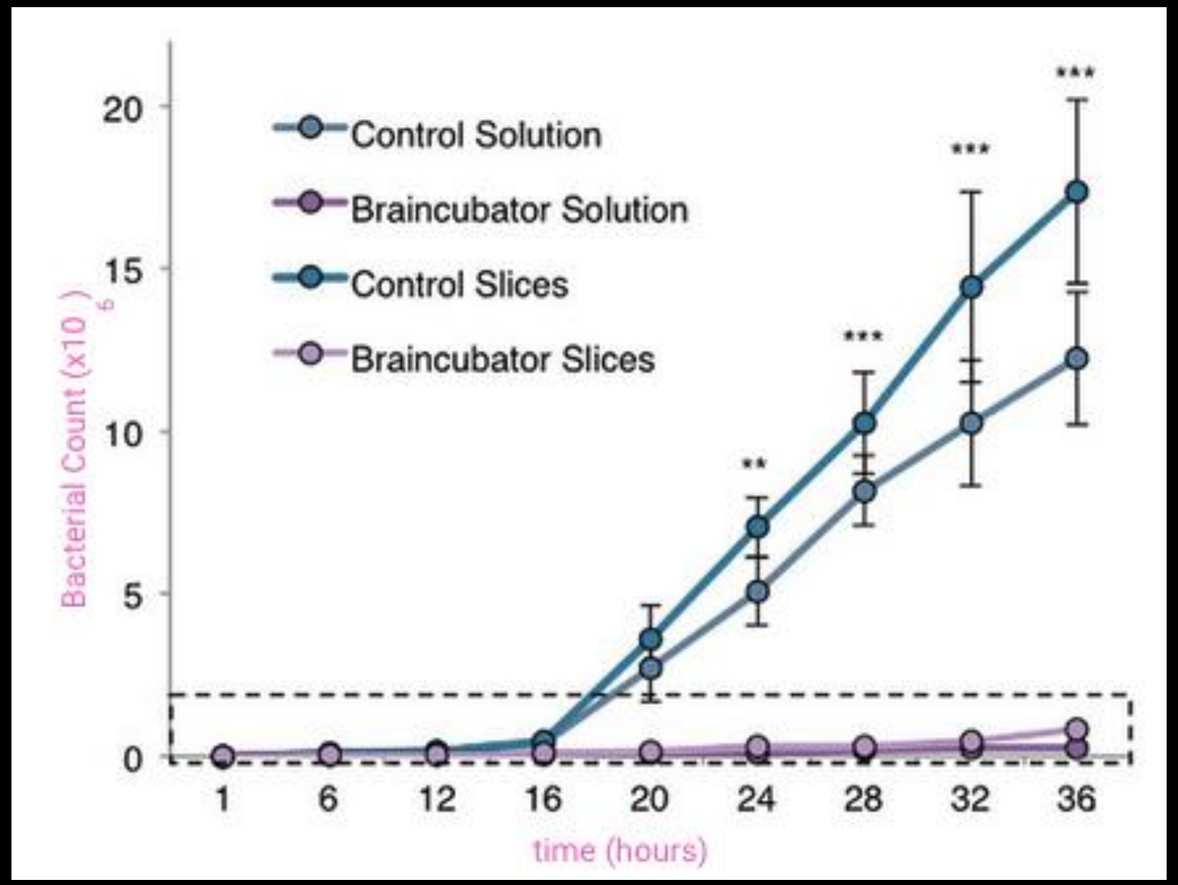


- **Assessment:** *Stenotrophomonas maltophilia* counts tracked at T=0 – T=36 hrs
- **Design:** *S. maltophilia* counts compared at 4hr intervals in Braincubator vs control conditions (solution and slices)
- **Results:** Braincubator-incubated solution or slices ***prevented bacterial growth*** for up to 36 hrs post-excision

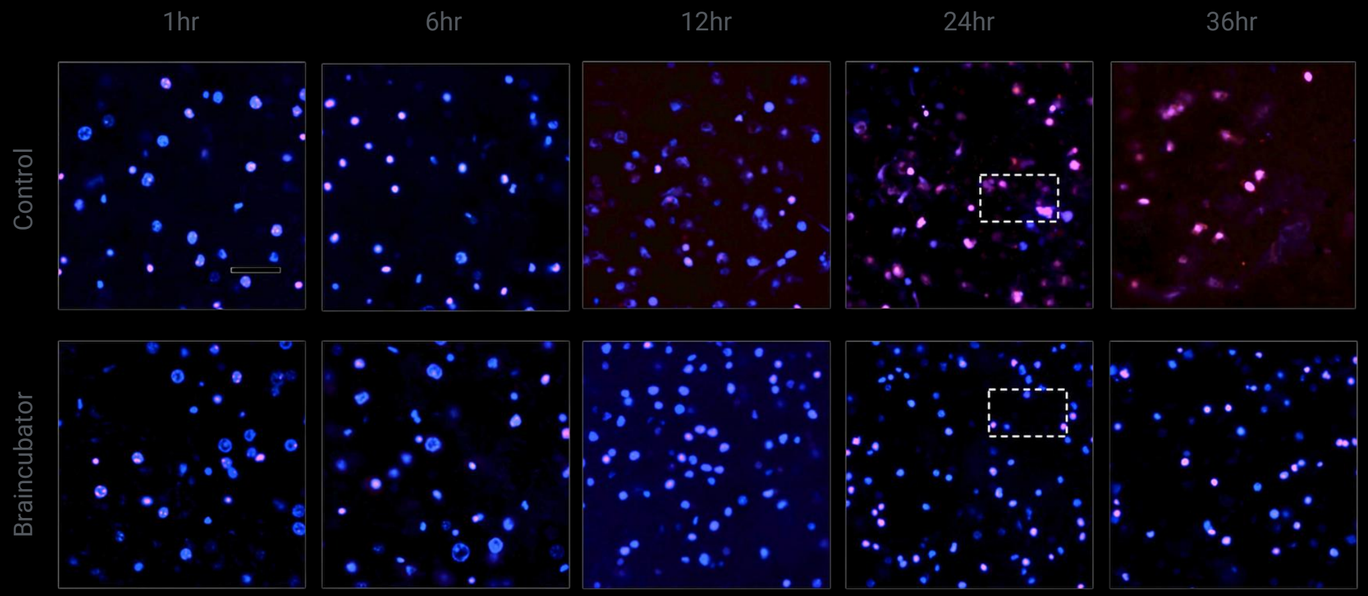
SLICE VIABILITY

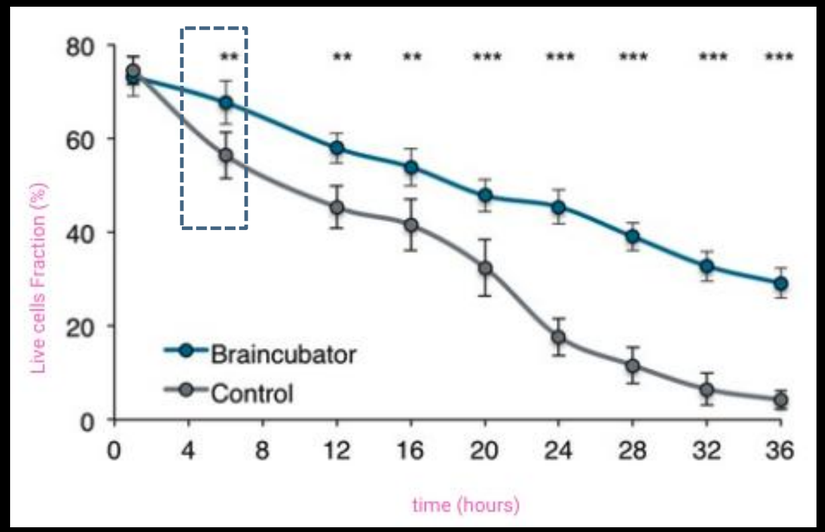
STANDARD CONTROL VS BRAINCUBATOR

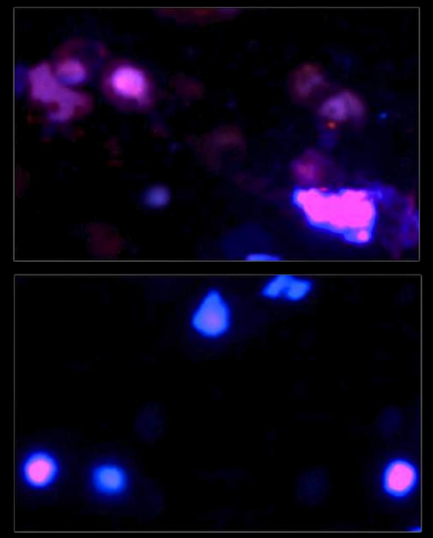
- **Assessment**: Viability of rat cortex slices using PI (dead cells [red]) and DAPI (total cells [blue])

- **Design**: Slices in Braincubator vs. control imaged over 1–36 hours post-sectioning (n=54)

- **Results**: From 6 hours onwards, Braincubator slices showed ***significantly higher viability***







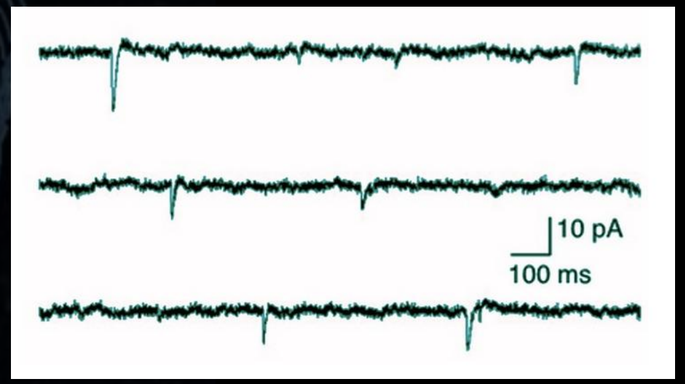
SLICE FUNCTIONALITY

BRAINCUBATOR ™

Stable neuronal **electrophysiology**

Spontaneous **synaptic activity**

Preserved neuronal **morphology**



Sample traces of spontaneous synaptic activity recorded **after 31 hrs in the Braincubator** indicate extended viability of acute brain slices

Buskila Y, Breen PP, Tapson J, van Schaik A, Barton M and Morley J (2014) Extending the viability of acute brain slices. Sci.Rep. 4, 5309; DOI:10.1038/srep05309

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Appendix

SLICE FUNCTIONALITY

BRAINCUBATOR™

Stable neuronal **electrophysiology**

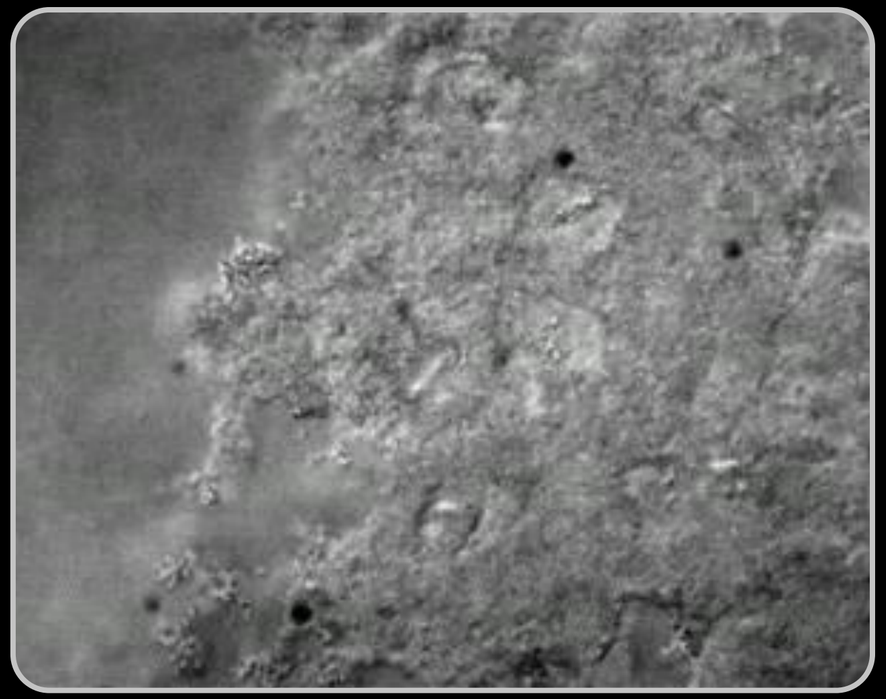
Spontaneous **synaptic activity**

Preserved neuronal **morphology**

	Control < 4 hrs (n = 10)	1 - 3 hrs (n = 12)	5 - 8 hrs (n = 7)	15 - 24 hrs (n = 9)	26 - 41 hrs (n = 9)
Resting membrane potential (mV)	−63 ± 2	−65 ± 2	−64 ± 1	−64 ± 1	−63 ± 1
Input resistance (MΩ)	176 ± 29	165 ± 18	163 ± 27	179 ± 21	145 ± 19
Membrane time constant (ms)	25 ± 4	28 ± 3	24 ± 7	25 ± 2	23 ± 3
Resonance Frequency (Hz)	2 ± 0.3	2 ± 0.2	1.7 ± 0.3	1.5 ± 0.3	1.6 ± 0.3
First spike amplitude (mV)	97 ± 5	97 ± 2	94 ± 4	93 ± 3	92 ± 3
Half-width spike amplitude (ms)	2.4 ± 0.2	2.5 ± 0.2	2.3 ± 0.1	2.6 ± 0.2	2.2 ± 0.1
mEPSC's frequency (Hz)	2.2 ± 0.3	1.75 ± 0.3	2.3 ± 0.4	1.5 ± 0.2	2.2 ± 0.3
mEPSC's frequency (pA)	−7.3 ± 0.5	−6.4 ± 0.5	−7.8 ± 0.5	−5.7 ± 0.4	−8 ± 0.7

Electrophysiological properties of layer V pyramidal neurons were stable across incubation times in the Braincubator vs control samples
No significant different between groups observed (one-way ANOVA)

BRAINCUBATO ™



- **IR-DIC Imaging:** After 30 hours in the Braincubator, slices showed intact neuronal morphology with smooth membranes
- **Presence of Viable Neurons:** Clear visualisation of healthy neurons was maintained.
- **Extended Viability:** Imaging confirmed preserved structural integrity of brain slices over 30 hours.

- **R-DIC Imaging:** Slices in the control chamber for 30 hours showed dead cell morphology (large vacuoles) across the Z-axis (-100 µm).
- **Loss of Viable Neurons:** No intact or viable neurons were observed after 30 hours.
- **Bacterial Presence:** Bacterial movement was visible on the slice surface.