# The Role of Inhibitory Interneurons in the Prefrontal Cortex of Patients with Post-Stroke Dementia and other Dementias

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## Introduction

- Stroke and age are two major risk factors for the development of dementia. With the ageing population, the incidence of dementia is estimated to increase to over a million people within the UK by 2050[1]
- The 1999 Cognitive Function After Stroke study investigated the role of stroke in the development of dementia. Approximately 30% of the stroke survivors from their cohort went on to develop post-stroke dementia, whilst others remained cognitively normal during their life span[2]
- A major hallmark of post-stroke dementia is executive dysfunction[3]. To explain this finding, ongoing research has been focused on the roles of pre-frontal cortical circuits, involved in executive function, in the development of post-stroke dementia
- Previous studies have identified striking changes in pyramidal neurons within one of these circuits, the **dorsolateral prefrontal cortex (dlPFC)**[4] found in **Brodmann's area 9**

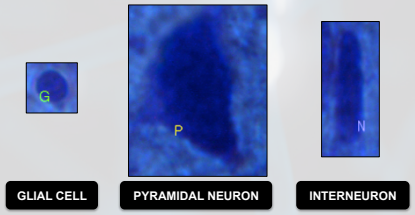



Figure 1 - A glial cell, pyramidal neuron and an interneuron

- In the dlPFC, **pyramidal neurons** create networks of axons sending **excitatory** signals across different brain regions. Mediating them are GABAergic **inhibitory interneurons**
- The balance between these neurons is vital for normal brain function. Stroke-caused lesions can lead to differences in their numbers thus affecting this crucial balance
- **Glial cells** are non-neuronal cells that support and protect neurons

## Objective

This project aims to:

- Investigate the role of the dlPFC in the development of post-stroke dementia, by assessing the densities of **interneurons** and **glial cells**, focusing on **cortical layer V** of the dlPFC

The outcome of this project will complement ongoing research, which aims to explain why one distinct group of stroke patients decline into dementia whilst others remain cognitively stable.

## Methodology

### STUDY SUBJECTS

We studied the brains of approximately 60 subjects, comparing post-stroke dementia (PSD), post-stroke non-demented (PSND), Alzheimer's disease (AD), vascular dementia (VaD), mixed dementia (consisting of AD & VaD pathology) subjects and age-matched controls (about 10 in each group).

### 3D STEREOLOGY

Brain sections of approximately 30μm thickness were analysed, with the operator remaining blinded to case details.

The sample area was mapped out at 2.5x magnification using the Visiopharm Integrator System software, and confirmed under 10x magnification.

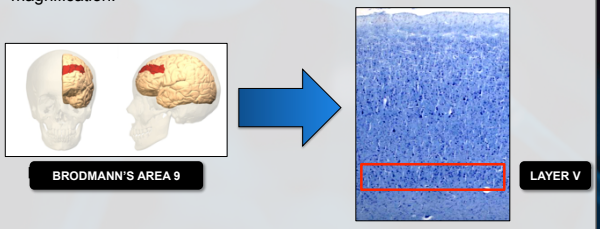



Figure 2 – An area of layer V mapped out from a section of Brodmann's area 9

Using a computer-generated random sampling technique, 20-40 samples were analysed within the sample area, per slide. Each case had 2-3 slides.

Using unbiased 3D stereology, large numbers of neurons and glial cells were sampled, and quantified under 100x magnification. Estimations of interneuronal and glial cell densities were calculated using this formula:

$$Nv = \frac{\sum^{p^-} Q^-}{P.V}$$

Where: Nv = numerical density, $p^-$ = dissector samples, $Q^-$ = number of objects counted, P = total number of dissectors and V = dissector box volume.

### STATISTICAL ANALYSIS

Using the SPSS version 21, parametric and non-parametric tests were used with the level of significance set at $p < 0.05$:

- One-way ANOVA with post hoc tests were performed on normally distributed data
- Mann-Whitney U-tests were performed on non-normally distributed data to establish any significance between groups

Correlations between variables were investigated using Spearman's rank (rho) coefficients.

## Results

| Group | Layer V Mean interneuronal density |
|---|---|
| Controls | 205641.596 |
| PSND | 213195.984 |
| PSD | 220422.640 |
| VaD | 270594.345 |
| Mixed | 226849.093 |
| AD | 210491.002 |

| Group | Layer V Mean glial cell density |
|---|---|
| Controls | 52913.6518 |
| PSND | 67414.1478 |
| PSD | 63116.8336 |
| VaD | 62878.367 |
| Mixed | 73614.846 |
| AD | 61944.4333 |

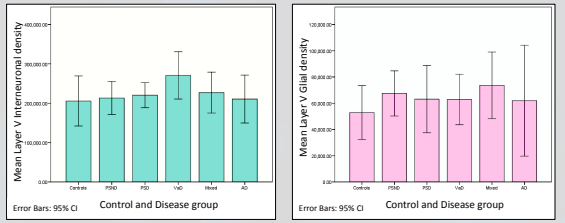
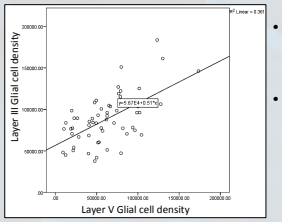



Figure 3 – Mean layer V interneuronal and glial cell densities in each dementia group

- **Interneuronal densities** were found to be non-normally distributed ($p = 0.004$), while **glial cell densities** were found to be normally distributed ($p = 0.059$)
- There were no significant differences in interneuronal and glial cell densities in layer V of the dlPFC between any of the subject groups




- Interestingly, a significant correlation was found between the glial cell densities in layers III and V
- Furthermore, no correlation was detected between fixation times and layer V interneuronal or glial cell densities, suggesting that fixation times did not affect the cell densities

Figure 4 – Correlation between glial cell densities in layers III and V

## Conclusion

- Neither interneuronal nor glial cell densities in layer V of the dlPFC appear to distinguish PSD subjects from their PSND counterparts
- Despite the different pathologies expressed across the demented groups, interneuronal and glial cell densities appeared to remain stable
- This work will continue, analysing remaining cortical levels, with the hope of accurately assessing cell densities in the various dementia groups and elucidating why only a proportion of stroke patients develop dementia

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**References:**
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Background image: "Neurons" (2011) obtained from Flickr, uploaded by Birth into Being (Available from: https://www.flickr.com/photos/birthintobeing/11841180046), accessed 17 October 2014

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