# Imaging of neuroinflammation in dementia: a review

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Dementia, Inflammation, Neuroimaging, Alzheimer Disease, TSPO protein

## Abstract

We are still very limited in management strategies for dementia, and establishing effective disease modifying therapies based on amyloid or tau remains elusive. Neuroinflammation has been increasingly implicated as a pathological mechanism in dementia and demonstration that it is a key event accelerating cognitive or functional decline would inform novel therapeutic approaches, and may aid diagnosis. Much research has therefore been done to develop technology capable of imaging neuroinflammation in-vivo. The authors performed a systematic search of the literature and found twenty-eight studies that used in vivo neuroimaging of one or more markers of neuroinflammation on human patients with dementia. The majority of the studies used PET imaging of the TSPO microglial marker and found increased neuroinflammation in at least one neuroanatomical region in dementia patients, most usually Alzheimer's disease, relative to controls, but the published evidence to date does not indicate whether the regional distribution of neuroinflammation differs between dementia types or even whether it is reproducible within a single dementia type between individuals. It is less clear that neuroinflammation is increased relative to controls in Mild Cognitive Impairment (MCI) than it is for dementia, and therefore it is unclear whether neuroinflammation is part of the pathogenesis in early stages of dementia. Despite its great potential, this review demonstrates that imaging of neuroinflammation has not thus far clearly established brain inflammation as an early pathological event. Further studies are required, including those of different dementia subtypes at early stages, and newer, more sensitive, PET imaging probes need to be developed.

# Imaging of neuroinflammation in dementia: a review

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## Introduction

Dementia is a chronic condition in which progressive cognitive impairment leads to functional disability. Alzheimer's disease (AD) is the most frequent cause of dementia, but other common types include vascular dementia, 'mixed' dementia, frontotemporal dementia (FTLD), dementia with Lewy Bodies (DLB), and Parkinson's Disease Dementia (PDD)[1]. Our understanding of the aetiology and pathogenesis of dementia is constantly increasing[2], but we are still very limited in our ability to accurately differentiate between dementia subtypes at early stages of the disease. This in turn limits our ability to give accurate diagnostic and prognostic information[3], particularly in the situation of overlapping phenotypes[4], and to develop treatments that prevent the progression of dementia when it is mild.

As well as tau and amyloid pathology, neuroinflammation is a disease process that has been increasingly implicated as a pathological mechanism in dementia[5]. Unlike neurological conditions such as multiple sclerosis and autoimmune encephalitis in which there is an established pathogenic role of the adaptive immune system, neuroinflammation in dementia is thought to be mediated predominantly by aberrant activation of the brain's innate immune system, namely microglia[5]. Microglia are CNS resident macrophages derived from haematopoietic stem cells whose processes rapidly converge on damaged cortex in vitro[6]. Ligands to the Peripheral Benzodiazepine Receptor (PBR, also known as the '18kDa Translocator Protein' or 'TSPO') found on activated microglia amplify apoptosis of rat neurones in vitro[7] and delivery of microglia inhibitory factor (MIF) significantly reduces microglial activation and the volume of damage caused by fibrillary Abeta in aged primate cortex[8]. Part of the neurodegeneration in dementia might be mediated through microglial release of pro-inflammatory cytokines such as interleukin-1B[9].

However, neuroinflammatory mechanisms are complex and incompletely understood, and neuroinflammation might also be neuroprotective in dementia under certain conditions and stages of the disease. It has been suggested that microgliosis in AD might initially be neuroprotective (through the degradation of amyloid plaques) but that impaired clearance of amyloid due to age or genetic predisposition leads to amyloid accumulation and an exaggerated microglial response that is ultimately neurodegenerative[10]. Astrocytes are the most numerous and diverse glial cells in the CNS and they phagocytose and degrade amyloid plaques in cultured mouse models of Alzheimer's disease[11], suggesting that, as with microglial activation, reactive astrogliosis in dementia might initially be a protective response to the primary neuropathological insult of amyloid plaque formation[12]. Furthermore, in-vivo imaging of TSPO in rats after ethanol-induced neuronal insults suggests that activated astrocytes might limit the expansion of microglial-induced scarring[13].

The above studies are invariably in-vitro, post-mortem or animal studies and predominantly in models of Alzheimer's disease rather than other forms of dementia. The ability to measure neuroinflammation in humans in vivo might therefore provide novel insights into the neuropathology of numerous types of dementia, the development of novel preventative therapies as well as enabling early diagnosis and prognosis of this extremely important disease.

Attempts have been made to measure serum and CSF biomarkers of neuroinflammation, but these have proven unable to provide sufficiently detailed information regarding the extent and distribution of neuroinflammation in vivo. Non-invasive imaging of neuroinflammation has therefore been heralded as a potential method of providing such information. Structural neuroimaging studies using e.g. CT, MRI have made limited progress to date, but functional neuroimaging technologies using e.g. PET[14] enable the imaging of specific molecules associated with inflammation and thus, potentially,

detailed information regarding the neuroanatomical distribution of specific biomarkers of inflammation.

Much research has therefore been done to develop technology capable of imaging neuroinflammation in-vivo [15]. However, efforts to develop PET ligands capable of quantifying regional neuroinflammation are hampered by our incomplete understanding of its pathophysiology. As mentioned above, the TSPO is thought to be a marker of activated microglia. It is a phylogenetically conserved receptor present on the outer mitochondrial membrane that has been implicated in cholesterol transport, cell replication and apoptosis; immunohistochemical studies have demonstrated minimal expression of TSPO in ependymal cells of healthy brain and significantly upregulated TSPO expression in microglia, macrophages and astrocytes of diseased brain[16]. Most of the plethora of PET ligands targeting putative inflammatory targets that have hitherto been used in animals or humans (see Supplementary File) have labelled the TSPO on activated microglia[16]. Table 1 is a summary of the techniques that have been used to image neuroinflammation in humans to date; whilst most target the TSPO, several ligands are available that target alternative putative indices of neuroinflammation including monoamine oxidase B (MAO-B) in activated astrocytes, peripheral macrophages that are thought to have infiltrated the CNS and even metabolite levels such as arachidonic acid or N-acetylaspartate that were thought to be non-specific markers of neuroinflammation. Increasingly, many studies have also investigated inflammatory neuroimaging in human dementia in vivo[17]. However, no comprehensive review has been recently published on this important topic.

The authors therefore performed a systematic search of the literature for all studies that imaged neuroinflammation in human patients with dementia in order to investigate whether neuroinflammation imaging might potentially be of use in the clinical management of dementia.

## Methods

In order to investigate the techniques used in neuroinflammation imaging, the literature was initially searched for studies that imaged neuroinflammation in-vivo in any neurological or psychiatric disorder in either humans or animal models. Thirty-three distinct imaging methodologies utilising PET, SPECT or MRI in either animals or humans were found and are listed in the Supplementary File;Table 1 contains the sixteen imaging methodologies that have been used in humans to date.

In order to investigate the imaging of neuroinflammation in human patients with dementia, on 7/9/2014 the databases AMED, EMBASE, HMIC, MEDLINE, PsycINFO, BNI, CINAHL, HEALTH BUSINESS ELITE were searched for articles with the following terms:

((dementia OR alzheimer* OR mild cognitive impairment OR Parkinson* OR front* OR vascular) AND (PET OR SPECT OR imaging OR neuroimaging) AND (inflammation OR PK11195 OR DAB OR PBR OR TSPO OR DPA*)).af

There were 15,720 results that reduced to 2128 after excluding duplicates. References from this electronic search, as well as reference lists of reviews and important papers in the field, were screened on the basis of relevance of title and abstract to the review. This yielded 100 potentially included references. Eight of these references were only published in abstract/poster form – the authors of all of these references were contacted twice but only three authors replied explaining a full length article had not been published, so all 8 of these references were excluded because the full length article could not be obtained or was not yet published. After further screening on the basis of full text, 28 of these 92 references were included in the review and are included in Table 2. In order to be included, studies had to use in vivo neuroimaging of one or more markers of neuroinflammation on human patients with dementia, which included Mild Cognitive Impairment (MCI) as well as AD, vascular dementia, mixed dementia, DLB, FTLD or PDD.

Table 1

| Ligand | Imaging Modality | Mechanism of imaging neuroinflammation |
|---|---|---|
| PK11195 | PET, SPECT | Measures TSPO |
| Ro5-4864 | PET | Measures TSPO |
| PBR28 | PET | Measures TSPO |
| FEDAA1106 | PET | Measures TSPO |
| DAA1106 | PET | Measures TSPO |
| AC-5216 | PET | Measures TSPO |
| Vincopetine | PET | Measures TSPO |
| FEPPA | PET | Measures TSPO |
| DEP (Deprenyl) | PET | Selective irreversible MAO-B antagonist that labels activated astrocytes |
| DED (Deuteriodeprenyl) | PET | Selective irreversible MAO-B antagonist that labels activated astrocytes |
| Arachidonic acid | PET | Neuroinflammation is characterised by phospholipase A2 activation, and thus arachidonic acid metabolism, in glia and neurones |
| TMSX | PET | Measures adenosine A2A receptors (a putative regulator of inflammation) |
| | MRS | Measurement of myoinositol, N-acetylaspartate and choline (metabolites proposed to be elevated in neuroinflammation) |
| Free-water MRI | MRI | A diffusion MRI processing analysis that can calculate extracellular volume, which is sensitive to neuroinflammation, and tissue fractional anisotropy, which is sensitive to axonal degeneration. Mainly images neuroinflammation of white matter. |
| USPIO=ultra small particle iron oxide e.g. SHU555C | MRI | Labels peripheral macrophages that have infiltrated the CNS |
| Gadolinium-DTPA | MRI | Images BBB breakdown, a putative 'marker' of inflammation |

Table 2

| Author (Year) | Number of participants in the following groups: | Imaging Ligand(s) | Neuranatomical areas with significantly increased inflammation in dementia group relative to controls | Other findings |
|---|---|---|---|---|
| Groom (1995) | Controls: 1<br>AD: 8<br>Small unilateral gliomas: 7 | [11C](R)PK11195<br>[18F]FDG | None | None reported |
| Cagnin (2001) | Controls: 15<br>AD: 8<br>MCI: 1 | [11C](R)PK11195<br>[18F]FDG | Inferior and middle temporal gyri, fusiform gyri, left parahippocampal gyrus, left amygdala, left posterior cingulate, inferior parietal lobes, putamen and right pallidum in AD relative to controls. Fusiform gyri, inferior temporal gyri and left parahippocampus in MCI relative to controls. | Increase inflammation in the left inferior temporal lobe discriminated AD relative to controls with a sensitivity of 75% and no false categorisations of controls. Areas with high inflammation showed the highest rate of atrophy over 12-24 months in AD. |
| Versijpt (2003) | Controls: 9<br>AD: 10 | [123I]PK11195<br>99mTc-ECD | Prefrontal cortices, lateral frontal cortices, left orbitofrontal cortex, right mesotemporal cortex. | Specific regionally increased inflammation was associated with specific cognitive impairments e.g. picture recognition task and right superior frontal cortex, orientation task and left lateral frontal, right superior frontal and left parietal cortex, verbal fluency and left parietal inferior cortex. |
| Cagnin (2004) | Controls: 8<br>FTLD: 5 | [11C](R)PK11195 | Putamen, left dorsolateral prefrontal cortex, right hippocampus, right parahippocampus. | None reported |
| Kropholler (2005) | Controls: 6<br>AD: 2<br>MCI: 5 | [11C](R)PK11195 | Intervention and control groups not statistically compared in article for this purpose | None reported |
| Anderson (2007) | Controls: 12<br>AD: 4<br>Huntington's: 11 | [11C](R)PK11195 | Intervention and control groups not statistically compared in article for this purpose | None reported |
| Kropholler (2007) | Controls: 20<br>AD: 9<br>MCI: 10 | [11C](R)PK11195 | None | None reported |
| Schuitemaker (2007) | Controls: 20<br>AD: 9 | [11C](R)PK11195 | Intervention and control groups not statistically compared in article for this purpose | None reported |
| Turkheimer (2007) | Controls: 18<br>AD: 4<br>Huntington's: 3 | [11C](R)PK11195 | Intervention and control groups not statistically compared in article for this purpose | None reported |
| Boellaard (2008) | Controls: 9<br>AD: 9 | [11C](R)PK11195 | Intervention and control groups not statistically compared in article for this purpose | None reported |
| Edison (2008) | Controls: 24<br>AD: 13 | [11C](R)PK11195<br>[11C]PIB | Frontal, temporal, parietal, occipital and cingulate cortices. | Inflammation in posterior cingulate gyri, parietal cortices and frontal cortices correlated with MMSE scores. Inflammation did not correlate with PiB binding. |
| Esposito (2008) | Controls: 9<br>AD: 8 | [1-11C]AA<br>[15O]water | 78 of 90 hemispheric grey-matter regions widespread in the neocortex, hippocampus and amygdala. The only regions without increased inflammation were the caudate, pallidum, posterior cingulate, right anterior and middle cingulate, left superior occipital cortex, left superior and medial temporal poles and left thalamus i.e. mainly subcortical. | None reported |
| Tomasi (2008) | Controls: 10<br>AD: 10 | [11C](R)PK11195 | Cerebellum, lateral occipital lobe, anterior cingulate gyrus, posterior cingulate gyrus, frontal lobe middle frontal gyrus, posterior temporal lobe, parietal lobe, thalamus, superior parietal gyrus and occipital lobe lingual gyrus. | None reported |
| Yasuno (2008) | Controls: 10<br>AD: 10 | [11C]DAA1106 | Dorsal and medial prefrontal cortices, lateral temporal cortices, parietal cortices, occipital cortices, anterior cingulate cortices, striatum and cerebellum. | No correlation between MMSE and inflammation in any volume of interest. |

| | | | | |
|---|---|---|---|---|
| Okello (2009) | Controls: 24<br>AD: 22<br>Amnestic MCI: 14 | [11C](R)PK11195<br>[11C]PIB | 11C-PiB positive MCI patients had increased inflammation in frontal cortices relative to controls. Data not given for inflammation in AD relative to controls. | No correlation between regional inflammation and PiB in MCI. No correlation between age of onset or MMSE score and inflammation in AD-PiB positive patients. Correlation between lower MMSE and increased inflammation in the anterior cingulate and temporal cortices in AD-PiB negative patients. |
| Wiley (2009) | Controls: 5<br>AD: 6<br>MCI: 6 | [11C](R)PK11195<br>[11C]PIB | None | No difference in inflammation between PiB positive or negative patients. |
| Gulyas (2011) | Controls: 12<br>AD: 6 | [11C]-vincopetine | None | None reported |
| Santillo (2011) | Controls: 11<br>AD: 9 | 11C-DED [11C]PIB | Frontal, parietal, temporal and medial temporal lobes. | Correlation between inflammation and PiB binding in occipital cortex. |
| Yokokura (2011) | Controls: 10<br>AD: 11 | [11C](R)PK11195<br>[11C]PIB [18F]FDG | Medial frontal and parietal cortices and left temporal cortex. | Negative correlation between MMSE and inflammation in the left anterior cingulate cortex, left precuneus, left hippocampus and left middle frontal cortex. Negative correlation between inflammation and PiB binding in posterior cingulate cortices. |
| Carter (2012) | Controls: 14<br>AD: 7<br>MCI: 8 | 11C-DED [11C]PIB<br>18F-FDG | Frontal and parietal cortices in MCI relative to controls. Right occipital cortex in MCI PiB+ patients relative to controls/MCI PiB- patients/AD.<br>Decreased inflammation in right hippocampus in AD relative to MCI and controls. | No regional correlation between the 3 PET tracers. |
| Yaqub (2012) | Controls: 17<br>AD: 8<br>MCI: 9 | [11C](R)PK11195 | None | None reported |
| Yasuno (2012) | Controls: 10<br>AD: 10<br>MCI: 7 | [11C]DAA1106 | Cerebellum, medial prefrontal cortices, parietal cortices, lateral temporal cortices, anterior cingulate cortices and striatum in MCI and AD relative to controls. No regional difference between MCI and AD. | No correlation between MMSE and regional inflammation. MCI patients with global inflammation 0.5 standard deviations greater than the control mean developed dementia within 5 years (75% AD, 25% DLB). |
| Edison (2013) | Controls: 24<br>PDD: 11<br>Parkinson's: 8 | [11C](R)PK11195<br>[11C]PIB [18F]FDG | Anterior and posterior cingulate, frontal, temporal, parietal, occipital cortices and striatum in PDD relative to controls. | Negative correlation between inflammation and MMSE in tempero-parietal, occipital and frontal cortices. |
| Iannaccone (2013) | Controls: 11<br>DLB: 6<br>Parkinson's: 6 | [11C](R)PK11195 | Basal ganglia, substantia nigra, frontal lateral cortices, parietal lateral cortices, temporal lateral cortices, anterior and posterior cingulate cortices, occipital medial cortices, occipital lateral cortices, temporal poles and precuneus in DLB relative to controls. | None reported |
| Kreisl (2013) | Controls: 13<br>AD: 19<br>MCI: 10 | [11C]PBR28<br>[11C]PIB | Prefrontal cortices, sensorimotor cortices, inferior parietal cortices, superior parietal cortices, middle and inferior temporal cortices, posterior cingulate cortices, occipital cortices, hippocampi and entorhinal cortices in AD relative to controls. Differences were greatest in inferior parietal lobules, middle and inferior temporal cortices and precuneus, but no difference was seen in white matter, thalamus, striatum or cerebellum. No difference in inflammation in any region between MCI and controls. | MMSE, Clinical Dementia Rating Scale Sum of Boxes, Logical Memory Intermediate (Wechsler Memory Scale Third Edition), Trail Making part B and Block Design (Wechsler Adult Intelligence Scale Third Edition) scores negatively correlated with inflammation, particularly in inferior parietal lobules, in MCI and AD. Inflammation in parietal cortex and striatum negatively correlated with age of onset in MCI and AD. Correlation between inflammation and PiB in inferior parietal lobules, superior temporal cortices, precuneus, hippocampi and parahippocampal gyri, in AD and MCI, after partial volume correction. |
| Schuitemaker (2013) | Controls: 21<br>AD: 19<br>MCI: 10 | [11C](R)PK11195 | Voxel-wise statistical parametric mapping showed small clusters of increased inflammation in occipital lobes in AD relative to controls. Region of interest analysis showed no significant differences in inflammation between groups (including between MCI and control patients). | No significant difference in regional inflammation between MCI patients who progressed to dementia and MCI patients who did not. Inflammation did not correlate with MMSE, New York University paragraph recall test, Rey's Auditory Verbal Learning Test or Trail A and B tests. |
| Varrone (2013) | Controls: 7<br>AD: 9 | [18F]FEDAA1106 | None | None reported |
| Yoder (2013) | Controls: 18<br>AD: 7<br>MCI: 7 | [11C]PBR28 | Intervention and control groups not statistically compared in article for this purpose | PBR28 binding is sensitive to TSPO genotype. |

Results

After screening on the basis of full text, 28 studies (see Table 2) were considered to meet the inclusion criteria and used in vivo neuroimaging of one or more markers of neuroinflammation on human patients with dementia, which was taken to include MCI as well as Alzheimer's disease, vascular dementia, mixed dementia, Lewy body dementia, frontotemporal dementia or Parkinson's Disease Dementia.

Twenty-seven of these studies used PET and only one study[18] used SPECT as the imaging modality to measure neuroinflammation.

Seven ligands were used; 19 studies used PK11195[18-36], two studies used DAA1106[37-38], one study used vincopetine[39], two studies used DED[40-41], two studies used PBR28[42-43], one study used FEDAA1106[44] and one study used arachidonic acid (AA)[45]. Five of these seven ligands (PK11195, DAA1106, vincopetine, PBR28, FEDAA1106) bind the TSPO on activated microglia, one (DED) binds to MAO-B in activated astrocytes and one (AA) images a metabolic by-product of microglial-induced neuroinflammation[45]. No study was found that used two ligands together to measure neuroinflammation.

The mean number of control participants was 13.1 (range 1-24) and the mean number of participants in a dementia group was 8.9 (range 1-22).

All 28 studies had a cross-sectional design to at least part of the trial; 25 of the 28 studies were purely cross-sectional whilst one study had both cross-sectional and case-control components[20] and two studies had both cross-sectional and cohort components[36, 38].

All 28 studies included healthy control participants while 14 studies had comparator groups with Alzheimer's disease[18-19, 23, 25-29, 32, 37, 39-40, 44-45], 11 studies had both Alzheimer's disease and MCI comparator groups[20, 22, 24, 30, 31, 33, 36, 38, 41, 42, 43], one study had FTLD as the comparator[21], one study had PDD[34] and one had DLB[35]. Six of the 28 included studies investigated neuroimaging methodologies and did not compare inflammatory ligand binding between dementia and control patients[22, 23, 25-27, 43]; their results are thus not pertinent to the question of whether inflammation neuroimaging might potentially be of use in the clinical treatment of dementia. The present review therefore found 22 studies which compared inflammatory ligand binding (and thus neuroinflammation) between control and dementia patients. A minority (6) of these 22 studies did not find a statistically significant difference in inflammation between control and dementia patients in at least one region of the brain[19, 24, 31, 33, 39, 44]; each of these 6 'negative' cross-sectional studies compared control patients with Alzheimer's disease +/- MCI patients, therefore no 'negative' cross-sectional studies were found that investigated other dementia subtypes. The majority (16) of the 22 included studies which compared neuroinflammation between control and dementia patients found a statistically significant difference in inflammation between control and dementia patients in at least one brain region[18, 20, 21, 28, 29, 30, 32, 34-38, 40, 41, 42, 45]. Unexpectedly, one of these 16 cross-sectional studies reported decreased inflammation in the right hippocampus in AD relative to controls[41], but the remainder found increased neuroinflammation in dementia patients.

Nine included studies compared MCI with healthy age-matched controls[20, 24, 30, 31, 33, 36, 38, 41, 42]. Three of these studies[24, 31, 33] found no statistically significant difference in neuroinflammation in AD or MCI relative to age-matched controls, two of these studies found increased neuroinflammation in AD (but not MCI) relative to controls[36, 42], two of these studies found increased neuroinflammation in AD and MCI relative to controls[20, 38], one study found increased neuroinflammation in MCI relative to controls but did not give data for AD relative to MCI

or controls[30] and one study found increased neuroinflammation in MCI relative to both AD and controls[41].

One study[20] used discriminant function analysis to show, retrospectively, that increased inflammation in the left inferior temporal lobe discriminated AD patients from age-matched controls with a sensitivity of 75% and with no false categorisations of controls.

One study[20] found that neuroanatomical regions with high inflammation demonstrated the highest rate of atrophy over 1-2 years, whilst Yasuno et al (2012)[38] found that 4 out of the 5 MCI patients who were followed up had whole measured region neuroinflammation more than 0.5 standard deviations greater than the control mean and all 4 developed dementia within 5 years. However, Schuitemaker et al (2013)[36] did not find significant differences in neuroinflammation between MCI patients who progressed to dementia and MCI patients who did not.

Several of the included studies investigated whether regional neuroinflammation correlated with dementia severity, as measured by either cognition[18, 28, 30, 32, 34, 36-38, 42] or age of dementia onset[30, 42]. Three of these studies found no statistically significant association between MMSE and ligand binding in any neuroanatomical region in Alzheimer's disease[36-38]. Five studies found statistically significant correlations between standardised cognitive questionnaire scores and regional neuroinflammation in Alzheimer's disease[18, 28, 30, 32, 42]. The neuroanatomical regions demonstrating an association between inflammation and cognition varied between studies but included frontal cortex[18, 28, 32], parietal cortex[18, 28, 42], temporal cortex[30], cingulate cortex[28, 30, 32], precuneus[32] and hippocampus [32]. Furthermore, one study found a statistically significant association between cognition and neuroinflammation in the frontal, parietal, temporal and occipital cortex in Parkinson's Disease Dementia[34]. No studies were found by this review investigating an association between cognition and neuroinflammation in any other dementia type.

Only two studies assessed whether regional neuroinflammation correlated with age of dementia onset in Alzheimer's disease. Okello et al (2009)[30] found no correlation between duration of symptoms and neuroinflammation in AD patients who were amyloid positive, whilst Kreisl et al (2013)[42] found that early onset (<65) AD patients had increased global neuroinflammation relative to late-onset (>65) AD patients. Kreisl et al (2013)[42] also found that neuroinflammation in the parietal cortex and striatum correlated with lower age of dementia onset in AD patients.

Seven studies were found that used multiple PET ligands to image not just neuroinflammation but also amyloid load (e.g. Pittsburgh Compound B [PiB] binding). Four of these seven studies found no statistically significant correlation between inflammatory ligand binding and PiB binding in Alzheimer's disease[28, 30, 31, 41]. Three of these seven studies[32, 40, 42] found inflammatory ligand binding to positively correlate with PiB binding (and thus, presumably, amyloid load) in at least one neuroanatomical region (the parietal cortex[42], temporal cortex[42], occipital cortex [40], cingulate cortex[32], hippocampus[42] and precuneus[42]) in Alzheimer's disease.

## Discussion

The present review contributes to the existing literature on neuroinflammation imaging, and thus is of interest not just for dementia but also for the diverse and increasing number of disorders that have a putative inflammatory component; this includes depression[46], delirium[47] and schizophrenia[48].

The majority of the studies to date of in vivo imaging of neuroinflammation in dementia have used PET imaging of the TSPO microglial marker. TSPO is an attractive imaging target because if it were found to be correlated with the development of dementia in vivo it would be relatively simpler to attempt to modify this single molecular target than if multiple separate markers of

neuroinflammation were each shown to be partly involved. However, the almost exclusive focus on TSPO is a weakness within the current imaging literature since microglial activation is unlikely to represent the totality of the neuroinflammatory response in dementia. Indeed, astrocytes have been implicated in the neuroinflammation of dementia[5] but only two[40,41] of the twenty-eight studies included in the present review used radioligands targeting a putative marker of astrocyte activation, namely MAO-B. Whilst the majority of PET ligands found to have been used in the imaging of neuroinflammation in any neuropsychiatric disorder in humans also target the TSPO (see Table 1), ligands are available from animal studies that target alternative putative indices of neuroinflammation e.g. COX-1, MPO, macrophage infiltration and even metabolite levels such as arachidonic acid or N-acetylaspartate (see Supplementary File). Future studies should therefore investigate the utility of such alternative ligands, either in isolation or in combination with TSPO-targeting ligands, in dementia patients.

An additional cause for concern by the current focus on TSPO imaging in the literature is the finding that the rs6971 TSPO single nucleotide polymorphism alters binding affinity of second generation radioligands for TSPO between healthy individuals independently of the degree of inflammation. Indeed, in vivo uptake in healthy individuals of the PBR28 radioligand for TSPO was 40% greater in homozygotes than heterozygotes for the high affinity TSPO protein [49]. However, only one[43] of the six[37-39,42-44] studies using second generation TSPO radioligands that were included in the present review measured participant TSPO binding affinity genotype. Furthermore, a possible role for the rs6971 TSPO single nucleotide polymorphism as a modifier of neuroinflammation in dementia has not been extensively investigated, despite in vitro evidence that high affinity rs6971 TSPO homozygotes have significantly elevated levels of the neurosteroid precursor, pregnenolone, in peripheral cells compared to low affinity homozygotes or heterozygotes[50]. It is therefore imperative that future imaging studies control for the relative proportions of low and high affinity TSPO participants[43].

Whilst a minority of the included studies did not find a statistically significant difference in inflammation between control and dementia patients in at least one region of the brain[19, 24, 31, 33, 39, 44], the studies included in this review were not adequately powered to conclude that there is no significant difference in neuroinflammation between dementia and controls. Furthermore, the majority (15) of included studies found increased neuroinflammation in at least one neuroanatomical region in dementia patients relative to controls. Alzheimer's disease was the most commonly studied dementia type, with increased inflammation in virtually all neuroanatomical locations, both cortical (frontal/parietal/temporal/occipital/cingulated/hippocampal/entorhinal/fusiform gyri/parahippocampal gyri) and subcortical (amygdala, pallidum, thalamus, striatum). There were significant differences between trials in the anatomical structures (and their laterality) demonstrated to have increased inflammation; comparison of results between studies was not helped by the frequent use of different neuroanatomical 'regions of interest' between studies. Thus, whilst the majority of studies investigating neuroinflammation imaging in Alzheimer's disease demonstrated increased neuroinflammation relative to controls, it is not possible to state with certainty the neuroanatomical location where such increased neuroinflammation occurs. This is in contrast with e.g. FDG-PET imaging studies demonstrating reproducible reductions in glucose metabolism in frontal, parietal, temporal and posterior cingulate cortices of Alzheimer's disease patients[51] and suggests either that existing imaging studies do not accurately portray the distribution of neuroinflammation in vivo or that neuroinflammation is globally increased in Alzheimer's disease. Furthermore, if neuroinflammation is globally increased in Alzheimer's disease this might suggest that neuroinflammation does not contribute to the early stages of its disease pathogenesis, since early Alzheimer's disease typically involves preferential atrophy of the temporal cortex and hippocampus[4]. Only three studies were found that investigated non-Alzheimer's types of dementia (FTLD[21], PDD[34] and DLB[35]), but all three, similarly to the findings in Alzheimer's

disease, found increased neuroinflammation in a variety of cortical and subcortical structures. Therefore a plethora of neuroanatomical structures have been implicated as having increased neuroinflammation in all dementias studied, but the published evidence to date does not indicate whether the regional distribution of neuroinflammation differs between dementia types or even whether it is reproducible within a single dementia type between individuals. It also raises the tantalising possibility that neuroinflammation might be a common pathological mechanism in all types of dementia.

Nine included studies compared MCI patients with healthy controls[20, 24, 30, 31, 33, 36, 38, 41, 42]. Taken together, the results of these 9 studies are equivocal; five of these studies found no statistically significant difference in neuroinflammation in MCI relative to control patients[24, 31, 33, 36, 42] whilst 4 of these studies found increased neuroinflammation in MCI relative to controls in frontal cortices[38, 30, 41], parietal cortices[38, 41], temporal cortices[20, 38], anterior cingulate cortices[38], fusiform gyri[20], left parahippocampus[20], striatum[38] and cerebellum[38]. Thus, it is less clear that neuroinflammation is increased relative to controls in MCI than it is for dementia, and therefore it is unclear whether neuroinflammation is part of the pathogenesis in early stages of dementia. However, the regional distribution of any neuroinflammation that might occur appears to be similar to that in dementia i.e. in a variety of cortical and subcortical structures.

Multi-tracer PET studies included in this review found only an inconsistent and weak correlation between neuroinflammation and other, better established markers of Alzheimer's disease load (such as PiB). This suggests that neuroinflammation imaging might highlight novel aspects of dementia not revealed by existing neuroimaging ligands and therefore be more useful in the clinical management of dementia than existing neuroimaging methods have proven to be. Indeed, several of the included studies attempted to investigate neuroinflammation imaging for potential clinical management purposes in dementia. One study[20] found, retrospectively, that increased neuroinflammation in the left inferior temporal lobe distinguished AD patients from controls with no false categorisations of controls and a sensitivity of 75%; further studies are now needed that prospectively trial the use of neuroinflammation imaging as a diagnostic tool in early stages of dementia, including MCI, when a clinical diagnosis is difficult to make. Furthermore, three of the included studies investigated whether neuroinflammation was prognostic in Alzheimer's disease[20, 36, 38]; Cagnin et al (2001)[20] found that neuroanatomical regions with high inflammation demonstrated the highest rate of atrophy over 1-2 years, whilst Yasuno et al (2012)[38] found that 4 out of the 5 MCI patients who were followed up had whole measured region neuroinflammation more than 0.5 standard deviations greater than the control mean and all 4 developed dementia within 5 years. However, Schuitemaker et al (2013)[36] did not find significant differences in neuroinflammation between MCI patients who progressed to dementia and MCI patients who did not; further research is therefore needed before deciding whether neuroinflammation imaging might potentially be used as a prognostic tool in the clinical management of dementia. Similarly, several of the included studies attempted to correlate neuroinflammation with dementia severity, as measured by either standardised cognition questionnaire scores or age of onset. The results were mixed and not convincingly reproducible; 3 studies found no association between cognition and regional neuroinflammation in Alzheimer's disease[36-38], five studies found significant correlations between cognition and regional neuroinflammation in Alzheimer's disease[18, 28, 30, 32, 42], one study found a statistically significant association between cognition and neuroinflammation in Parkinson's Disease Dementia[34], one study found no correlation between age of onset and neuroinflammation in AD[30] whilst one study found that neuroinflammation in the parietal cortex and striatum correlated with earlier disease onset in AD[42]. Therefore the existing literature does not unequivocally demonstrate that neuroinflammation is an objective correlate of dementia severity in vivo.

Despite its great potential, the lack of reproducibility of positive findings in this review therefore demonstrates that neuroinflammation imaging has not thus far been convincingly used for diagnosis, prognosis or severity profiling in any dementia subtype. The observed heterogeneity of results between studies of the same type of dementia (Alzheimer's disease) is likely due to multifactorial causes that include greater variability in the pathogenesis of neuroinflammation than is currently understood; since different inflammatory mechanisms might be responsible for different phases of neuroinflammation in dementia, it is not entirely unsurprising that a single imaging modality detecting one aspect of neuroinflammation (such as microglial activation in the case of the PK11195 ligand) might be unable to detect similar differences between Alzheimer's disease patients at different stages of the disease. Furthermore, existing studies have generally been small and inadequately powered, and whilst PK11195 is a robust marker of neuroinflammation its small signal-to-noise ratio might make it unable to detect subtler patterns of regional neuroinflammation that might exist in dementia. In order to further investigate the role of neuroinflammation imaging in the clinical management of dementia, adequately powered trials are therefore needed which take into account the stage of dementia, patient TSPO genotype and use more sensitive TSPO imaging ligands, ideally in conjunction with alternative markers of neuroinflammation.

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Competing Interests

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