

Short Communication

Reference Region Normalization in Amyloid and Tau Imaging with Positron Emission Tomography (PET)

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Tau and amyloid imaging with positron emission tomography (PET) will play an increasingly important role in the design of Alzheimer's disease trials, both for the subject stratification and to evaluate the biological effects of drugs. Clinical PET acquisition protocols utilize short static scans with no arterial blood sampling. These images are normalized by a reference region's activity before semi-quantitative regional or voxel-based analyses. The selection of an appropriate reference region for PET normalization is a critical factor for data interpretation. This study provides an overview on some of the challenges associated with PET normalization process in Alzheimer's disease.

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Pathological biomarkers of Alzheimer's disease (AD), such as amyloid beta plaques ($A\beta$) and neurofibrillary tau tangles, will play an increasingly important role in the design of AD trials to identify appropriate subjects and to track the effects of drugs. For instance, $A\beta$ biomarkers have helped to identify $A\beta+$ individuals, thus presumably on the AD trajectory, and improved the ability of more recent trials to detect anti- $A\beta$ treatment effects [1]. Abnormal levels of $A\beta$ and tau can be detected with the analysis of the uptake of layer-specific PET tracers, such as [^{18}F] Florbetapir [2], [^{11}C] PIB [3], [^{18}F] Flutemetamol [4], [^{18}F] Florbetaben [5] for $A\beta$ and [^{18}F] T807 (AV-1451) [6], [^{11}C] PBB3 [7], [^{18}F] THK5317 [8] for tau. $A\beta$ /tau-PET images are normalized by a reference region's activity. The PET intensity normalization process is a simple way of determining activity in PET images and allows semi-quantitative comparisons between different scans and subjects. These normalized images serve as starting points for subsequent analyses. PET data analysis techniques can be as simple as the regional/global mean of standardized uptake value ratio (SUVR) [9] or sophisticated data-driven, voxel-based analyses [10-12] or machine learning diagnostic techniques [13], which have found wider applications in research settings than in routine clinical diagnosis. Most clinical $A\beta$ -PET images are visually assessed by trained experts [14]. However, quantitative methods will continue to play an increasingly important role in AD trials.

In the past, $A\beta$ -PET studies in AD used cerebellum as the reference region. Price and colleagues [15] showed that in amyloid-negative subjects, the pharmacokinetics of [^{11}C] PIB in the cerebellar gray matter are similar to the cerebral gray matter target regions. This pharmacokinetic similarity was one of the main factors for the selection of cerebellum as the reference region in previous $A\beta$ -PET studies. However, recent research with Florbetapir [16-19] has found variabilities associated with the cerebellar normalization of PET images, presumably due to the location of the cerebellum, which falls on the edge of the scanner's axial field of view (FOV). Pathological effects could also contribute to the cerebellar amyloid uptake. Based on the Braak staging [20], the cerebellar amyloid deposit is present in stage three. Knight and colleagues [21] reported increased cerebellar

retention of [^{11}C] PIB in presenilin-1 (PS1) mutation carriers. Catafau and colleagues [22] used post-mortem immunohistochemistry on 64 AD subjects and nine non-demented volunteers to demonstrate parenchymal and vascular $A\beta$ deposits in cerebellum and determine their impact on [^{18}F] Florbetaben SUVR values. While they found diffuse plaques in 6% and vascular $A\beta$ in 25.3% of their samples, the effect of cerebellar plaques on cortical SUVRs appeared to be negligible in this study. In comparison to cerebellar gray matter, the clearance of [^{11}C] PIB in the pons and subcortical white matter (WM) is different than in cerebral gray matter target regions. Nevertheless, The utilization of white matter (and pons) as an alternative reference region has reduced variabilities in the longitudinal progression of $A\beta$ -PET retention [16-19], improved discrimination power between subject groups [16] as well as increased the association between $A\beta$ -PET and clinical decline [17] and cerebrospinal $A\beta$ 1-42 [19].

Several cross-sectional tau-PET studies [6] have used cerebellar gray matter as the reference region. Given that longitudinal studies are of particular interest in tau imaging, the selection of cerebellum as the reference region for tau can obscure the subtle longitudinal changes in tracer uptake due to the additional noise created by its axial position in the scanner. While WM can be used as an alternative reference region in tau-PET to reduce the scan-to-scan variability, there are potentially other problems related to WM normalization. These include the presence of physiological and structural damages that can influence the non-specific uptake of amyloid [23,24] and possibly also tau radiotracers. For instance, Veronese and colleagues [24] showed that [^{11}C] PIB uptake is sensitive to myelin changes in both preclinical models and humans. WM damages are prevalent in the aging population and are related to vascular risk factors, cognitive impairment and dementia [25]. Therefore, the utility of WM as a PET reference region may depend on its structural and functional integrity, which can vary among individual subjects. WM damages appear as hyperintensities on T2-weighted, proton density and fluid-attenuated inversion recovery sequences (FLAIR) of magnetic resonance imaging (MRI). Other advanced MRI imaging techniques, such as Diffusion Tensor Imaging (DTI) [26,27] can provide additional information on the micro structural integrity of the white matter by measuring fractional anisotropy (FA), which reflects the diffusion directionality

and mean diffusivity (MD), which shows the magnitude of water diffusion. DTI-MRI techniques can detect decreased structural integrity in normal-appearing white matter (NAWM) surrounding white matter hyperintensities [28]. Partial volume effects pose additional challenges to WM normalization, particularly in tau-PET imaging. Unlike amyloid tracers, tau retention in gray matter spreads into the nearby white matter, and the PET spatial resolution is too poor to separate the gray matter contribution from the white matter signal [29]. The presence of other pathophysiological conditions could also impact the non-specific radiotracer uptake in both cerebellum and WM. For instance, a history of traumatic brain injury (TBI) may pose an additional challenge to cerebellar normalization due to the increased likelihood of A β accumulation in the cerebellum of TBI subjects [30]. After age, family history, and APOE- ϵ 4, TBI is the strongest risk factor for AD [31,32]. TBI affected subjects, such as Veterans and retired athletes will be increasingly recruited into AD drug trials. Therefore, cerebellum may not be a suitable reference region for this population. The binding characteristics of amyloid/tau radiotracers in the study of WM diseases are not entirely understood. There is a need for further studies related to this topic. Additional MRI scans to evaluate WM damages in individual subjects would be a good idea in clinical trials. Recent work by Fleischer and colleagues [33]. Found that the longitudinal percent change between placebo and solanezumab groups was not significant when cerebellum was used as reference region but became significant when the subcortical white matter was used as the reference region. This study is a good example of where the detection of damaged white matter regions (e.g. due to ARIA-E [34]) and their exclusion from the reference region ROI could further increase the effect size between treatment and placebo groups.

In summary, we have identified that the PET normalization process is a critical challenge for A β /tau-PET imaging in the aging population and Alzheimer's disease. One alternative solution would be to eliminate the reference region normalization from semi-quantitative PET studies. For amyloid imaging, this can be done by using topographic techniques, such as weighted two-point correlation functions [19], Haralick features [35], and peak cortical laminar deposition [36] that can be performed on non-normalized PET images. The idea behind these techniques is based on previous pathological observations on the spatiotemporal progression of amyloid in AD. For instance, from studies by Bruce and colleagues [37], we know that over the time the progressive amyloid deposition encompasses a greater extent of cerebral cortical laminae. The same pattern of progression is observed in PET radiotracer uptake, which starts from the interface between gray matter and white matter and gradually expands toward the brain surface.

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References

- Sevigny J, Chiao P, Bussi re T, Weinreb PH, Williams L, Maier M, et al. The antibody aducanumab reduces A β plaques in Alzheimer's disease. *Nature*. 2016; 537: 50-56.
- Wong DF, Rosenberg PB, Zhou Y, Kumar A, Raymond V, Ravert HT, et al. In vivo imaging of amyloid deposition in Alzheimer disease using the radioligand 18F-AV-45 (florbetapir [corrected] F 18). *J Nucl Med*. 2010; 51: 913-920.
- Klunk WE, Engler H, Nordberg A, Wang Y, Blomqvist G, Holt DP, et al. Imaging brain amyloid in Alzheimer's disease with Pittsburgh Compound-B. *Ann Neurol*. 2004; 55: 306-319.
- Koole M, Lewis DM, Buckley C, Nelissen N, Vandenbulcke M, Brooks DJ, et al. Whole-body biodistribution and radiation dosimetry of 18F-GE067: a radioligand for in vivo brain amyloid imaging. *J Nucl Med*. 2009; 50: 818-822.
- Sabri O, Seibyl J, Rowe C, Barthel H. Beta-amyloid imaging with florbetaben. *Clin Transl Imaging*. 2015; 3: 13-26.
- Schwarz AJ, Yu P, Miller BB, Shcherbinin S, Dickson J, Navitsky M, et al. Regional profiles of the candidate tau PET ligand 18F-AV-1451 recapitulate key features of Braak histopathological stages. *Brain*. 2016; 139: 1539-1550.
- Wang M, Gao M, Xu Z, Zheng QH. Synthesis of a PET tau tracer [(11)C] PBB3 for imaging of Alzheimer's disease. *Bioorg Med Chem Lett*. 2015; 25: 4587-4592.
- Chiotis K, Saint-Aubert L, Savitcheva I2, Jelic V3, Andersen P3, Jonasson M4, et al. Imaging in-vivo tau pathology in Alzheimer's disease with THK5317 PET in a multimodal paradigm. *Eur J Nucl Med Mol Imaging*. 2016; 43: 1686-1699.
- Lopresti BJ, Klunk WE, Mathis CA, Hoge JA, Ziolkowski SK, Lu X, et al. Simplified quantification of Pittsburgh Compound B amyloid imaging PET studies: a comparative analysis. *J Nucl Med*. 2005; 46: 1959-1972.
- Minoshima S, Frey KA, Koeppe RA, Foster NL, Kuil DE. A diagnostic approach in Alzheimer's disease using three-dimensional stereotactic surface projections of fluorine-18-FDG PET. *J Nucl Med*. 1995; 36: 1238-1248.
- Fleisher AS, Chen K, Quiroz YT, Jakimovich LJ, Gomez MG, Langois CM, et al. Florbetapir PET analysis of amyloid- β deposition in the presenilin 1 E280A autosomal dominant Alzheimer's disease kindred: a cross-sectional study. *Lancet Neurology*. 2012; 11: 1057-1065.
- Villain N, Chetelat G, Grassiot B, Bourgeat P, Jones G, Ellis KA, et al. AIBL Research Group. Regional dynamics of amyloid-beta deposition in healthy elderly, mild cognitive impairment and Alzheimer's disease: a voxelwise PiB-PET longitudinal study. *Brain*. 2012; 135: 2126-2139.
- Cattell L, Platsch G, Pfeiffer R, Declerck J, Schnabel JA, Hutton C; Alzheimer's Disease Neuroimaging Initiative. Classification of amyloid status using machine learning with histograms of oriented 3D gradients. *Neuroimage Clin*. 2016; 12: 990-1003.
- Daerr S, Brendel M, Zach C, Mille E, Schilling D, Zacherl MJ, et al. Evaluation of early-phase [18F]-florbetaben PET acquisition in clinical routine cases. *Neuroimage Clin*. 2016; 14: 77-86.
- Price JC, Klunk WE, Lopresti BJ, Lu X, Hoge JA, Ziolkowski SK, et al. Kinetic modeling of amyloid binding in humans using PET imaging and Pittsburgh Compound-B. *J Cereb Blood Flow Metab*. 2005; 25: 1528-1547.
- Brendel M, H genauer M, Delker A, Sauerbeck J, Bartenstein P, Seibyl J, et al. Alzheimer's Disease Neuroimaging Initiative. Improved longitudinal [(18)F]-AV45 amyloid PET by white matter reference and VOI-based partial volume effect correction. *Neuroimage*. 2015; 108: 450-459.
- Chen K, Roontiva A, Thiyyagura P, Lee W, Liu X, Ayutyanont N, et al. Improved power for characterizing longitudinal amyloid- β PET changes and evaluating amyloid-modifying treatments with a cerebral white matter reference region. *J Nucl Med*. 2015; 56: 560-566.
- Landau SM, Fero A, Baker SL, Koeppe R, Mintun M, Chen K, et al. Measurement of longitudinal 125 I-amyloid change with 18F-florbetapir PET and standardized uptake value ratios. *J Nucl Med*. 2015; 56: 567-574.
- Shokouhi S, McKay JW, Baker SL, Kang H, Brill AB, Gwirtsman HE, et al. Reference Tissue Normalization in Longitudinal 18F-Florbetapir Positron Emission Tomography of Late Mild Cognitive Impairment. *Alzheimers Res Ther*. 2016; 8: 2.
- Braak H, Braak E. Neuropathological staging of Alzheimer-related changes. *Acta Neuropathol*. 1991; 82: 239-259.
- Knight WD, Okello AA, Ryan NS, Turkheimer FE, Rodr guez Martinez de Llano S, Edison P, et al. Carbon-11-Pittsburgh compound B positron emission

- tomography imaging of amyloid deposition in presenilin 1 mutation carriers. *Brain*. 2011; 134: 293-300.
22. Catafau AM, Bullich S, Seibyl JP, Barthel H, Ghetti B, Leverenz J, et al. Cerebellar Amyloid- β Plaques: How Frequent Are They, and Do They Influence 18F-Florbetaben SUV Ratios? *J Nucl Med*. 2016; 57: 1740-1745.
 23. Matías-Guiu JA, Cabrera-Martín MN, Matías-Guiu J, Oreja-Guevara C, Riola-Parada C, Moreno-Ramos T, et al. Amyloid PET imaging in multiple sclerosis: an (18)F-florbetaben study. *BMC Neurol*. 2015; 15: 243.
 24. Veronese M, Bodini B, García-Lorenzo D, Battaglini M, Bongarzone S, Comtat C, Bottlaender M, et al. Quantification of [11C]PIB for imaging myelin in the human brain: a test-retest reproducibility study in high-resolution research tomography. *J Cereb Blood Flow Metab*. 2015; 35: 1771-1782.
 25. Xiong YY, Mok V. Age-related white matter changes. *J Aging Res*. 2011; 2011: 617927.
 26. Jones DK, Leemans A. Diffusion tensor imaging. *Methods Mol Biol*. 2011; 711: 127-144.
 27. Basser PJ, Mattiello J, LeBihan D. MR diffusion tensor spectroscopy and imaging. *Biophys J*. 1994; 66: 259-267.
 28. Promjunyakul N, Lahna DL, Kaye JA, Dodge HH, Erten-Lyons D, Rooney WD, et al. Comparison of cerebral blood flow and structural penumbras in relation to white matter hyperintensities: A multi-modal magnetic resonance imaging study. *Journal of Cerebral Blood Flow & Metabolism*. 2016; 36: 1528-1536.
 29. Alzheimer's Association International Conference News 23 Aug 2016.
 30. Scott G, Ramlackhansingh AF, Edison P, Hellyer P, Cole J, Veronese M, et al. Amyloid pathology and axonal injury after brain trauma. *Neurology*. 2016; 86: 821-828.
 31. Van Den Heuvel C, Thornton E, Vink R. Traumatic brain injury and Alzheimer's disease: a review. *Prog Brain Res*. 2007; 161: 303-316.
 32. Daviglus ML, Plassman BL, Pirzada A, Bell CC, Bowen PE, Burke JR, et al. Risk Factors and Preventive Interventions for Alzheimer Disease: State of the Science. *Arch Neurol*, 2011; 1185-1190.
 33. Fleisher AS, Joshi AD, Sundell KL, Chen YF, Kollack-Walker S, Lu M, et al. Use of white matter reference regions for detection of change in florbetapir positron emission tomography from completed phase 3 solanezumab trials. *Alzheimers Dement*. 2017: S1552-5260(17)30094-8.
 34. Sperling RA, Jack CR, Black SE, Frosch MP, Greenberg SM, Hyman BT, et al. Amyloid Related Imaging Abnormalities (ARIA) in Amyloid Modifying Therapeutic Trials: Recommendations from the Alzheimer's Association Research Roundtable Workgroup. *Alzheimer's Dement*. 2011; 7: 367-385.
 35. Campbell DL, Shokouhi S. Application of Haralick texture features in brain 18F-Florbetapir positron emission tomography. *Society of Nuclear Medicine and Molecular Imaging Conference 2017*.
 36. Minoshima S, Cross D, Wang A, Foster N, Drzezga A. Peak Location of Amyloid PET Tracer Uptake within Cortical Gray Matter: Topographic Patterns and Diagnostic Application in Alzheimer's Disease. *Society of Nuclear Medicine and Molecular Imaging (SNMMI) Annual meeting*. 2016.
 37. Bruce CV, Clinton J, Gentleman SM, Roberts GW, Royston MC. Quantifying the pattern of beta/A4 amyloid protein distribution in Alzheimer's disease by image analysis. *Neuropathol Appl Neurobiol*. 1992; 18: 125-136.