Association of Insulin Resistance With Cerebral Glucose Uptake in Late Middle-Aged Adults at Risk for Alzheimer Disease

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IMPORTANCE Converging evidence suggests that Alzheimer disease (AD) involves insulin signaling impairment. Patients with AD and individuals at risk for AD show reduced glucose metabolism, as indexed by fludeoxyglucose F 18-labeled positron emission tomography (FDG-PET).

OBJECTIVES To determine whether insulin resistance predicts AD-like global and regional glucose metabolism deficits in late middle-aged participants at risk for AD and to examine whether insulin resistance–predicted variation in regional glucose metabolism is associated with worse cognitive performance.

DESIGN, SETTING, AND PARTICIPANTS This population-based, cross-sectional study included 150 cognitively normal, late middle-aged (mean [SD] age, 60.7 [5.8] years) adults from the Wisconsin Registry for Alzheimer’s Prevention (WRAP) study, a general community sample enriched for AD parental history. Participants underwent cognitive testing, fasting blood draw, and FDG-PET at baseline. We used the homeostatic model assessment of peripheral insulin resistance (HOMA-IR). Regression analysis tested the statistical effect of HOMA-IR on global glucose metabolism. We used a voxelwise analysis to determine whether HOMA-IR predicted regional glucose metabolism. Finally, predicted variation in regional glucose metabolism was regressed against cognitive factors. Covariates included age, sex, body mass index, apolipoprotein E ε4 genotype, AD parental history status, and a reference region used to normalize regional uptake.

MAIN OUTCOMES AND MEASURES Regional glucose uptake determined using FDG-PET and neuropsychological factors.

RESULTS Higher HOMA-IR was associated with lower global glucose metabolism ($\beta = -0.29; P < .01$) and lower regional glucose metabolism across large portions of the frontal, lateral parietal, lateral temporal, and medial temporal lobes ($P < .001$). The association was especially robust in the left medial temporal lobe ($R^2 = 0.178$). Lower glucose metabolism in the left medial temporal lobe predicted by HOMA-IR was significantly related to worse performance on the immediate memory ($\beta = 0.317; t_{148} = 4.08; P < .001$) and delayed memory ($\beta = 0.305; t_{148} = 3.895; P < .001$) factor scores.

CONCLUSIONS AND RELEVANCE Our results show that insulin resistance, a prevalent and increasingly common condition in developed countries, is associated with significantly lower regional cerebral glucose metabolism, which in turn may predict worse memory performance. Midlife may be a critical period for initiating treatments to lower peripheral insulin resistance to maintain neural metabolism and cognitive function.
Glucoregulatory impairment has reached epidemic proportions in the United States. According to the American Diabetes Association, 1,29.1 million individuals in the United States have diabetes mellitus, and more than half of adults older than 64 years have prediabetes. Type 2 diabetes mellitus increases the risk for Alzheimer disease (AD), and clinical and preclinical hyperglycemia are characterized by insulin resistance. Insulin resistance is broadly defined as reduced tissue responsiveness to the action of insulin. Insulin, a key hormone involved in carbohydrate metabolism, facilitates microvascular blood flow, glucose uptake, and glucose oxidation for adenosine-5’-triphosphate generation. In addition to its function in the periphery of the body, insulin has increasingly been recognized as playing an important role in the brain. Insulin resistance is related to a higher risk for AD, and the results of several animal studies as reviewed by de La Monte1 link central insulin resistance with the pathologic features of AD, including atrophy, mitochondrial dysfunction, neuroinflammation, and progressive memory deficits. In humans, brain insulin resistance has been found in postmortem hippocampal tissue in patients with AD, and the degree of insulin signaling inhibition corresponds to the severity of antemortem cognitive dysfunction. Several studies have shown a deleterious effect of insulin resistance on regional brain volume in cross-sectional19-20 and longitudinal15-19 analyses. Birdsell et al21 have shown recently that participants with metabolic syndrome, a condition linked with insulin resistance, have markedly lower cerebral blood flow, a presumed index of neuronal function. Finally, Willette et al22 have found that higher insulin resistance predicts temporal and frontal amyloid deposition in late-middle-aged individuals at risk for AD.

Peripheral insulin resistance has also been linked with an impaired cerebral metabolic rate of glucose in the brain. Glucose metabolism is commonly assessed using fludeoxyglucose F 18 uptake on positron emission tomography (FDG-PET). Baker and colleagues13 showed that higher insulin resistance predicts reduced glucose uptake in older, cognitively intact adults (mean age, 74.4 years) with dysglycemia. Willette et al22 showed similar associations in patients with AD. Patterns of lower glucose metabolism included hypometabolism in the posterior cingulate cortex and precuneus and in the frontal and temporal cortices. Peripheral insulin resistance strongly corresponds to brain insulin resistance owing to reduced insulin transport into the brain or potentially similar changes in receptor sensitivity and activation.5,15 These findings are intriguing given that lower glucose metabolism in these brain regions is also a feature characteristic of AD.16-19 Lower glucose metabolism has also been observed in mild cognitive impairment20 and in cognitively healthy carriers of the apolipoprotein E ε4 (APOE ε4) allele, a genetic risk factor for AD.21,22 However, the relationship between insulin resistance and brain glucose metabolism in middle-aged adults is unknown. Understanding the neural effects of midlife insulin resistance is important given that the onset of type 2 diabetes mellitus is most common in this population and increases the risk for AD.23

In this study, we assessed the effect of insulin resistance on glucose metabolism as indexed by FDG-PET scanning in a cognitively healthy, late middle-aged cohort of adults enriched for a parental history of AD. We hypothesized that participants with higher insulin resistance would show lower glucose metabolism in brain regions that exhibit hypometabolism in early AD. Given that glucose metabolism is tied to functional status, we also tested the extent to which the variation in medial temporal lobe (MTL) glucose metabolism predicted by insulin resistance was associated with cognitive performance. Finally, based on prior work in this area,21,24 we tested the main effects of the APOE ε4 genotype and parental history of AD on glucose metabolism.

### Methods

**Participants**

Demographic characteristics are listed in the Table. One hundred fifty cognitively normal, older middle-aged adults (mean [SD] age, 60.7 [5.8] years) were recruited from the Wisconsin Registry for Alzheimer’s Prevention (WRAP) study.29 This ongoing study examines genetic, biological, and lifestyle factors that contribute to the development of dementia-related cognitive decline and neural dysfunction. The University of Wisconsin-Madison Health Sciences Institutional Review Board approved our study, and all participants provided written informed consent. Participants were originally recruited from 40 to 65 years of age and were classified as having a positive or negative parental history of AD.29 A positive parental history of AD was defined as having one or both parents with AD as determined by autopsy results (13 cases) or by validated interview,30 reviewed by a multidisciplinary diagnostic consensus panel, and outlined by research criteria.31,32 Detailed medical history and telephone interviews were
conducted to confirm AD-negative parental history. The inclusion criteria for this study consisted of no clinical diagnosis of a memory disorder, no contraindication for brain imaging, subsequent normal findings on magnetic resonance imaging, no current diagnosis of major psychiatric disorder or other major medical conditions (eg, myocardial infarction or recent history of cancer), and no history of head trauma. All participants underwent magnetic resonance imaging, FDG-PET, and neuropsychological testing. Participants were categorized as APOE ε4 carriers (1 or 2 ε4 alleles) or noncarriers (no ε4 alleles). Extraction and isoform classification of APOE alleles have been described previously.23

**Neuropsychological Testing**

Participants in the WRAP study undergo an extensive battery of neuropsychological tests. A previous factor-analytic study of the WRAP cognitive battery within the larger cohort found that the tests map onto 6 cognitive factors.34,35 The scores used in the present study were derived from tests administered at the participants’ most recent WRAP visit and represent cognitive domains known to change with age and AD, including immediate memory,25 verbal learning and memory,26 working memory,26 and speed and flexibility.27,28

**Homeostatic Model Assessment of Insulin Resistance, Diabetes Mellitus Status, and Body Mass Index**

Glucose and insulin levels were measured after a 12-hour fast during the clinical visit nearest in time to the FDG-PET scan. Insulin resistance was indexed by the homeostatic model assessment (HOMA-IR) and calculated by taking the product of basal glucose (in milligrams per deciliter) and basal insulin (in microunits per milliliter) levels and dividing by 405.36 Matthews et al originally derived HOMA-IR using glucose levels in millimoles per liter. Although HOMA-IR was considered a continuous variable, we also determined how many participants in our sample had type 2 diabetes mellitus by using criteria from the American Diabetes Association, that is, fasting blood glucose levels of greater than 125 mg/dL (to convert to millimoles per liter, multiply by 0.0555). No participants were currently or previously taking medication for glycemic control; however, 5 participants self-reported a history of diabetes mellitus. Participants without type 2 diabetes mellitus display normal beta cell function.37 Body mass index was calculated as weight in kilograms divided by height in meters squared.

**FDG-PET Imaging**

Images were acquired in the supine position and head first on a PET scanner (HR+ scanner; Siemens) in a 3-dimensional mode after a 4-hour fast (with water allowed). Blood glucose levels were closely monitored before the injection of FDG. After injection of a mean (SD) of 5.0 (0.5) mCi of FDG, participants remained awake but relaxed in a quiet room. Imaging began 45 minutes after the injection. The scan was acquired as six 5-minute frames. A 5-minute transmission scan was acquired after the emission scan. The dynamic PET data were reconstructed using software from the manufacturer (ECAT, version 7.2.2; Siemens). A filtered back-projection algorithm (DIFT) was used with brain mode sinogram trimming, with a 2.8 zoom and a 4-mm gaussian filter to a reconstructed image of 128 × 128 × 63-voxel matrix (voxel size, 1.84 × 1.84 × 2.43 mm). The PET data were corrected for the attenuation of annihilation radiation (using segmented attenuation maps), scanner normalization, and scatter radiation.

To account for between- and within-subject noise, a reference cluster was used as a covariate in statistical analyses.38,39 The reference cluster consisted of sixty-five 2 × 2 × 2-mm contiguous voxels centered in the right cuneus, a region where no significant relationship between the FDG signal and HOMA-IR was found (controlling for age, sex, APOE ε4 genotype, and parental history status) (β = −0.09; t148 = −1.1; P = .27). The region was derived via a data-driven method that identifies brain regions unaffected by the variable of interest and has been shown to improve detection of disease-related hypometabolism.39-41 The raw values from the reference region were extracted with a region-of-interest toolbox (MarsBaR) for Statistical Parametric Mapping (SPM).42

**Statistical Analysis**

To test for the effects of HOMA-IR, APOE ε4 genotype, and parental history status on global glucose metabolism (adjusted by the reference region), multiple regression analysis was implemented in SPSS (SPSS Statistics for Windows, version 21.0; IBM Corp). In addition to testing for main effects, we assessed for interactions between HOMA-IR and APOE ε4 genotype and between HOMA-IR and parental history status. Both analyses of main effects and interactions were conducted using a single-design matrix model and controlled for age at the time of scanning, sex, body mass index, and reference region. Voxelwise statistical analysis used SPM8 software (http://www.fil.ion.ucl.ac.uk/spm) to test for the regional effect of HOMA-IR on glucose metabolism. We controlled for covariates identical to those in the global analysis. A voxelwise analysis was also conducted to test for the regional main effects of parental history status and APOE ε4 genotype on glucose metabolism, again controlling for age, sex, body mass index, and reference region. Models were also run that included an interaction term for HOMA-IR and APOE ε4 genotype or for HOMA-IR and parental history status. Type I error was minimized by using a voxel-level familywise error correction of P < .05. Results were also interrogated at a threshold of P < .001. Multiple regression analysis was also used to regress HOMA-IR-predicted variation in glucose metabolism from an a priori-defined left MTL region against cognitive factors. These analyses covaried age at the time of the scan, sex, parental history status, APOE ε4 genotype, body mass index, and reference region. The sample size was insufficient to conduct robust mediation analysis.43 Logarithmic transformation of HOMA-IR was used in all analyses to optimize normality and reduce heteroscedasticity.

**Results**

**Demographic Characteristics and Cognition**

Demographic characteristics, the 4 cognitive factors, and other summary data across participants are shown in the
Table. Of the 150 participants, 108 (72.0%) were women; 103 (68.7%) had a parental history of AD, 61 (40.7%) had an APOE ε4 allele, and 7 (4.7%) had type 2 diabetes mellitus. Additional metabolic risk factor characterization is provided in the eTable in the Supplement.

Associations of Insulin Resistance, APOE ε4 Genotype, and Family History Status

Global FDG-PET
A single multiple regression model tested the statistical effects of HOMA-IR, APOE ε4 status, and parental history of AD on global glucose metabolism. Higher HOMA-IR was associated with lower global glucose metabolism (β = −0.29; t143 = −3.10; P < .01). Figure 1 shows the degree of this insulin resistance association. The same regression model showed a significant effect of the APOE ε4 genotype, with carriers with 1 or 2 ε4 alleles had lower global glucose metabolism (β = −0.16; t143 = −2.01; P < .05) compared with noncarriers. We found no significant effect of parental history status on global glucose metabolism, nor did insulin resistance interact with APOE ε4 genotype or parental history status to affect uptake of FDG-PET.

Regional FDG-PET
In a single voxelwise regression model (P < .001), HOMA-IR, APOE ε4 genotype, and parental history status were used to determine whether regional associations with glucose metabolism existed. Higher HOMA-IR was robustly associated with lower glucose metabolism in portions of the ventral prefrontal, cingulate, temporal, insula, and posteromedial cortices (Figure 2A). In addition, associations were seen in the bilateral cerebellum. Coverage was sparse or absent in motor and premotor cortices and the dorsal prefrontal cortex. Thresholding revealed that associations were strongest in the hippocampus and MTL, rostral and posterior cingulate, and precuneus and cuneus. Left MTL survived at P < .05, familywise error corrected. Figure 2B shows the degree of this association between insulin resistance and the mean signal in the left MTL. We observed no significant effect of APOE ε4 genotype or parental history status on regional glucose metabolism. In addition, we found no interaction between APOE ε4 genotype and HOMA-IR or between parental history status and HOMA-IR.

Insulin Resistance, MTL FDG-PET, and Cognitive Function
Mean predicted variation in glucose metabolism specific to HOMA-IR was extracted using the Eigenvariate tool in SPM8.
The region of interest was the left MTL, which was of a priori interest before we conducted voxelwise analyses. Left medial temporal glucose metabolism was regressed against each of the 4 cognitive factors. Covariates were identical to the voxelwise analysis. Adjusted glucose metabolism was associated with the immediate memory ($\beta = 0.317; t_{148} = 4.08; P < .001$) and the verbal learning and memory ($\beta = 0.305; t_{148} = 3.895; P < .001$) factor scores and weakly associated with the speed and flexibility factor ($\beta = 0.118; t_{148} = 1.448; P = .08$) (Figure 3). We found no significant relationship with the working memory factor score ($\beta = 0.204; t_{148} = 2.537; P = .01$) (Figure 3).

Discussion

Several studies\textsuperscript{44-46} suggest that insulin resistance is associated with brain changes that may contribute to the pathologic features of AD in the preclinical phase. This study assessed the extent to which insulin resistance may affect glucose metabolism as measured by FDG-PET. We found that late middle-aged adults with higher HOMA-IR showed lower glucose metabolism and that glucose metabolism was related to memory function.

Our results concur with findings in older adults indicating that insulin resistance,\textsuperscript{13} hyperglycemia,\textsuperscript{43} and diabetes mellitus\textsuperscript{48} are associated with hypometabolism on FDG-PET. Insulin resistance and hyperglycemia are related conditions, and hyperglycemia, even in the prediabetic range, is associated with a significantly increased risk for later development of dementia.\textsuperscript{49} However, fasting insulin and glucose levels are not always correlated,\textsuperscript{50} and insulin resistance may confer an increased risk for AD independently of glycemic status within 3 years of assessment.\textsuperscript{4} Our sample was a mean of 15 years younger than the sample studied by Baker et al,\textsuperscript{13} although the affected regions are similar. In particular, both studies show an association between higher HOMA-IR and less glucose metabolism in the bilateral prefrontal cortex, temporal cortex, and posteromedial parietal cortex. Willette et al\textsuperscript{14} found similar associations in the same regions among patients with AD. We also found bilateral hypometabolism in the cerebellar cortex, a region with appreciable insulin receptor density.\textsuperscript{51,52} Although not typically considered a region affected by AD, a few reports have found mild cerebellar hypometabolism in AD.\textsuperscript{53,54} Brains with AD undergoing postmortem examination also show deficient insulin signaling in the cerebellum,\textsuperscript{6} whereas intranasal insulin levels may maintain glucose metabolism.\textsuperscript{55}

An association between higher HOMA-IR and lower glucose metabolism was found in the left MTL. In rodents, the piriform cortex and adjoining cornu ammonis fields 1 and 2 are well established as having a high density of insulin receptors relative to the moderate density in the cerebral cortex.\textsuperscript{52,55} When we examined mean glucose metabolism in the left MTL, we found that lower glucose metabolism was associated with worse immediate and delayed memory performance factors. This result is in line with that of Wolk and Dickerson,\textsuperscript{56} who found that entorhinal and perirhinal volumes in patients with mild AD predicted later trials of the Rey Auditory Verbal Learning Test.\textsuperscript{25}
on which the verbal learning and memory factors used in the present study are based. This finding provides a potential link between insulin resistance and cognitive decline.

Several possible mechanisms may underlie the association between higher insulin resistance and lower glucose metabolism. For example, our group has found that higher peripheral insulin resistance in asymptomatic late middle-aged participants is linked with amyloid deposition measured in vivo. In participants with stable mild cognitive impairment enriched for amyloid status, Willette et al observed that higher HOMA-IR predicted less prefrontal glucose metabolism only in the amyloid-positive group. Loss of neuronal function owing to mitochondrial damage is another possible mechanism. Alzheimer disease and type 2 diabetes mellitus are characterized by mitochondrial dysfunction, providing a potential common link between the 2 diseases. Neurons rely heavily on mitochondria for the synthesis of adenosine triphosphate and are therefore vulnerable to mitochondrial dysfunction. Indeed, lower expression of nuclear genes influencing mitochondrial energy metabolism colocalize with brain regions that show deficits in glucose metabolism in patients with AD. Insulin resistance may also facilitate several additional mechanisms that result in neurodegeneration, including increased oxidative stress, neuroinflammation, and dysregulated lipid metabolism.

Akin to the report by Willette et al examining HOMA-IR and brain atrophy in asymptomatic adults, HOMA-IR associations with regional glucose metabolism in this study were robust, even with familywise error correction, with relationships ranging from moderate to moderately strong in most regions. Baker and colleagues found stronger relationships among cognitively normal older adults with prediabetes or type 2 diabetes mellitus. Our finding that insulin resistance is associated with increased amyloid burden and decreased glucose uptake in AD-centric brain regions indicates that insulin resistance confers a nontrivial risk for AD in midlife. In addition, HOMA-IR in this cohort appeared to predict glucose metabolism more strongly than APOE \( \epsilon 4 \) genotype, which is an important predictor of glucose uptake deficits and AD in general. However, this study was cross-sectional, and no causal inferences about insulin resistance may be inferred. Although we found an effect of APOE \( \epsilon 4 \) genotype on global glucose metabolism, we did not observe any regional effects. Furthermore, HOMA-IR did not interact with APOE \( \epsilon 4 \) status to affect global or regional glucose metabolism. Interactions between APOE \( \epsilon 4 \) genotype and metabolic dysfunction have been observed in previous studies, including effects on cerebrospinal fluid biomarkers in preclinical AD, postmortem plaque and tangle burden in patients with AD dementia, development of amnestic mild cognitive impairment, and response to intranasal insulin therapy. However, found no interactions between hyperglycemia and APOE \( \epsilon 4 \) genotype on FDG-PET. Although several studies point toward a moderating effect of APOE \( \epsilon 4 \) genotype when considering the effects of insulin resistance on neural pathologic features, findings across the field are still mixed. In an existing report that shares similarities with our study, Roberts et al examined individuals with diabetes mellitus and did not find an interaction between diabetes and APOE \( \epsilon 4 \) genotype on FDG-PET. Although our sample was enriched for parental history of AD, HOMA-IR did not interact with parental history status, and parental history alone did not have an effect on glucose metabolism. Previous studies have found an effect of parental history of AD on cerebral glucose metabolism; however, the findings are most robust when both parents are affected or maternal family history is considered.

Additional studies are needed to clarify interactions between insulin resistance, parental history, and APOE \( \epsilon 4 \) across the spectrum of disease development, from preclinical AD to diagnosed dementia. This information will be central to developing prevention and treatment therapies centered on insulin dysregulation.

### Conclusions

This study provides evidence that insulin resistance is associated with brain glucose metabolism in a late middle-aged cohort enriched for AD risk factors. Several studies indicate that peripheral insulin resistance and related conditions, such as metabolic syndrome and diabetes mellitus, are risk factors for cognitive decline and AD and are linked with an increased risk for death from dementia. The prevalence of AD continues to grow, and midlife may be a critical period for initiating treatments aimed at preventing or delaying the onset of AD. Accumulating evidence suggests that treatments targeting mechanisms involved in insulin signaling may affect central glucose metabolism and should be investigated in the context of presymptomatic AD.
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