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Increased Risk of Cognitive Impairment in Patients With Diabetes Is Associated With Metformin

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OBJECTIVE—To investigate the associations of metformin, serum vitamin B_{12}, calcium supplements, and cognitive impairment in patients with diabetes.

RESEARCH DESIGN AND METHODS—Participants were recruited from the Primary Research in Memory (PRIME) clinics study, the Australian Imaging, Biomarkers and Lifestyle (AIBL) study of aging, and the Barwon region of southeastern Australia. Patients with Alzheimer disease (AD) (n = 480) or mild cognitive impairment (n = 187) and those who were cognitively intact (n = 687) were included; patients with stroke or with neurodegenerative diseases other than AD were excluded. Subgroup analyses were performed for participants who had either type 2 diabetes (n = 104) or impaired glucose tolerance (n = 22).

RESULTS—Participants with diabetes (n = 126) had worse cognitive performance than participants who did not have diabetes (n = 1,228; adjusted odds ratio 1.51 [95% CI 1.03–2.21]). Among participants with diabetes, worse cognitive performance was associated with metformin use (2.23 [1.05–4.75]). After adjusting for age, sex, level of education, history of depression, serum vitamin B_{12}, and metformin use, participants with diabetes who were taking calcium supplements had better cognitive performance (0.41 [0.19–0.92]).

CONCLUSIONS—Metformin use was associated with impaired cognitive performance. Vitamin B_{12} and calcium supplements may alleviate metformin-induced vitamin B_{12} deficiency and were associated with better cognitive outcomes. Prospective trials are warranted to assess the beneficial effects of vitamin B_{12} and calcium use on cognition in older people with diabetes who are taking metformin.

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In 2010, more than 346 million people had diabetes worldwide. Recent studies from the U.K. (1) and Italy (2) reported that the adult prevalence of diabetes was approximately 4.2%. In the U.S., the prevalence of diabetes in the adult population may be as high as 14% when undiagnosed cases are included (3). The prevalence of diabetes may be higher in some developing nations: in the developing region of southern China it was reported to be 21.7% in 2010 (4). The prevalence of diabetes is more than 20% in some Pacific Island nations, reaching 47% in 22- to 64-year-old American Samoans (5).

In diabetes, hyperglycemia activates the cellular signaling protein kinase C, which induces production of the vasoconstrictor protein endothelin-1. Excess intracellular glucose is converted to sorbitol by the enzyme aldose reductase. When intracellular levels of glucose are high, this process exhausts the energy substrate NADPH, resulting in oxidative stress. High intracellular sorbitol levels cause osmotic stress and cell death. These biochemical changes in hyperglycemia are a proposed mechanism for macrovascular and microvascular complications and neuropathy (6–8). Diabetes is associated with a faster rate of cognitive decline in those with mild cognitive impairment (MCI) (9) and an increased risk for developing Alzheimer disease (AD) (10).

Approximately 90% of patients with diabetes have type 2 diabetes (1). The biguanide metformin is a first-line treatment for type 2 diabetes, increasing glucose uptake in muscle while reducing liver gluconeogenesis (the synthesis of glucose from amino acids). These effects are mediated by activation of the cellular signaling protein AMP–activated protein kinase (11).

Metformin first became available in the U.K. in 1958 and entered the Canadian market in 1971, but it has been available in the U.S. only since 1995. In a survey of 65,000 U.S. war veterans (12), metformin use among those with diabetes increased from 29% in 2000 to 63% in

2005. Among 242 Australian veterans who had diabetes, metformin was used by 75% in 2005 but decreased to 57% in 2009 (13).

The rate of vitamin B_{12} deficiency among patients who are taking metformin is reported to approach 30% (14–16). A drug interaction between metformin and the cubilin receptor inhibits the uptake of vitamin B_{12} from the distal ileum, lowering serum vitamin B_{12} levels. In a long-term, randomized, placebo-controlled trial, metformin therapy in type 2 diabetes was associated with a 19% reduction in serum vitamin B_{12} concentrations compared with placebo (17). In a case-control study, Wile and Toth (18) reported that metformin use was associated with reduced vitamin B_{12} levels and more severe peripheral neuropathy in patients with diabetes.

In a prospective trial, calcium supplements were reported to reverse the drug interaction that causes vitamin B_{12} deficiency induced by metformin (19). The clinical significance of alleviating metformin-induced vitamin B_{12} malabsorption by calcium supplementation has not been previously investigated. By correcting vitamin B_{12} levels in patients with diabetes who use metformin, calcium supplements may help to preserve cognitive function. In addition, neuronal signaling in memory and learning involves a calcium-mediated process, so calcium supplementation may also have a direct effect on the brain. Calcium dysregulation is the subject of one proposed theory for age-related cognitive changes and AD (20). The risks and potential benefits of calcium supplementation on cognition and for alleviating vitamin B_{12} malabsorption merit further investigation.

The amyloid plaques seen in the brains of patients with AD are formed by aggregation of Aβ peptides. In cell cultures, Chen and colleagues (21) reported that activation of AMP–activated protein kinase by metformin increased the expression of β-secretase, an enzyme that increases the formation of Aβ peptides. One recent case-control study that included 14,172 participants 65 years of age or older reported that taking metformin over the long term increased the risk of AD (odds ratio [OR] 1.71 [95% CI 1.12–2.60]) (22).

Recent studies of murine models of diabetes indicate that metformin may attenuate irregularities in phosphorylation of tau proteins (23) or may facilitate neuroneogenesis (24) and so may be of benefit to those with AD. In 25,393 patients older than 50 years with type 2 diabetes, metformin was reported to reduce the risk of dementia by 24% (hazard ratio 0.76 [95% CI 0.58–0.98]) (25). The purpose of our study was to investigate the associations of metformin, serum vitamin B_{12} levels, and cognition in a sample of patients with diabetes.

RESEARCH DESIGN AND METHODS

Participants and settings

Participants were recruited from two prospective studies: the Prospective Research in Memory (PRIME) clinics study and the Australian Imaging, Biomarkers and Lifestyle (AIBL) study of aging. Only data and biochemical measurements pertaining to baseline visits were included in this analysis. The PRIME study recruited 970 participants from 9 sites in Australia, including 3 each in Victoria and New South Wales and 1 each in Queensland, Western Australia, and South Australia. The AIBL study of aging recruited 1,112 participants in Victoria (60%) and Western Australia (40%). The study cohorts and methods of the PRIME and AIBL studies are described elsewhere (26,27).

A further 862 participants who resided in the Barwon region of southeastern Australia between 2001 and 2011 also were recruited through the Cognitive, Dementia and Memory Services clinic at the McKellar Centre, a rehabilitation and aged-care facility. Patients with AD who were seen at a geriatrician's private practice during the same period also were recruited (n = 935).

Study design

Participants in the PRIME study were recruited during routine patient care. AIBL participants volunteered after an advertisement on television in late 2006. The Mini-Mental State Examination (MMSE) was used to assess cognitive ability. Subjects were assessed at scheduled visits during the PRIME and AIBL studies or ad hoc during routine patient care. All participants with serum vitamin B_{12} measurements taken within 6 months of cognitive assessment were included. Participants without serum measurements taken within 6 months of cognitive assessment (n = 1,015) were excluded.

The data from records pertaining to subjects who were recruited from more than one source (n = 566) were merged. Of the remaining participants, there were 121 with stroke and 566 with diagnoses other than AD, such as frontotemporal dementia, Parkinson disease, dementia with Lewy bodies, or mixed dementias. Participants with other neurodegenerative diseases were excluded to limit confounders. A further 291 participants were excluded because they had incomplete medical histories; information that was missing included dates of birth, medication use, and comorbid conditions.

A subgroup analysis was performed with participants who had diabetes (n = 104) or impaired glucose tolerance (n = 22). Patients with type 1 diabetes were not specifically excluded, but there were none with serum vitamin B_{12} measurements taken within 6 months of cognitive assessment.

Ethical approval

Institutional review was performed at each study host site. Reviewing committees included the Barwon Health Human Research Ethics Committee (HREC) (Victoria), Austin Health HREC (Victoria), St. Vincent's Hospital Governance Review Unit (Victoria), Hunter New England HREC (New South Wales), Northern Hospital Network HREC (New South Wales), Northern Sydney Central Coast HREC (New South Wales), Metro North Health Service District HREC (Queensland), South Metropolitan Area Health Service HREC (Western Australia), and The Queen Elizabeth Hospital Ethics of Human Research Committee (South Australia).

Statistical analyses

An ordinal logistic regression model was formed with categories of cognitive performance as the response variable and diabetes as a predictor. Categories were ordered by cognitive performance, including "most impaired" (MMSE <18; n = 137), "mildly impaired" (MMSE 18–23; n = 240), "minimally impaired" (MMSE 24–27; n = 295), and "not impaired" (MMSE 28–30; n = 682). The model was adjusted for age, sex, level of education, and depression because these factors were previously reported to affect MMSE testing (28,29).

A subgroup analysis of only patients with diabetes was then performed. The response variable was cognitive performance. Categories were "most impaired" (MMSE <18; n = 39), "mildly impaired" (MMSE 18–23; n = 40), "minimally impaired" (MMSE 24–27; n = 32), and "not impaired" (MMSE 28–30; n = 15). The

effect of metformin on the cognitive performance of patients with diabetes was investigated in this model, which then was adjusted for serum vitamin B_{12} measurements and use of calcium supplements to investigate any possible interactions. There was insufficient information on use of other antidiabetic drugs, the duration of metformin use, and markers for socioeconomic status, diet, or exercise to investigate these variables.

RESULTS—There were 1,354 participants included in this analysis (Fig. 1). Participants were 51–99 years old (mean age ± SD 73.8 ± 8.3 years); females outnumbered males (59.5 vs. 40.5%). Just more than half of the participants (50.4%) scored 28–30 on the MMSE and so were considered not impaired; 21.8% were minimally impaired (MMSE 24–27), 17.7% were mildly impaired (MMSE 18–23), and 10.1% scored less than 18 on the MMSE (most impaired).

Participants with diabetes were marginally older than participants without diabetes (75.5 vs. 73.6 years; $P = 0.013$). The number of males was proportionally larger among participants with diabetes (46.8 vs. 39.9%), but this difference was not statistically significant. Prevalence of depression was similar between participants with diabetes and those without diabetes (31.7 vs. 27.3%; $P = 1.000$). The proportion of participants with a tertiary level of education was higher among participants without diabetes than participants with diabetes (39.3 vs. 22.2%; $P < 0.001$). The number of participants who scored below 28 on the MMSE was proportionally higher among participants with diabetes than those without diabetes (69.0 vs. 47.6%; $P = 1.000$).

After adjusting for age, sex, education, and depression, participants with type 2 diabetes had worse cognitive performance than participants without diabetes (adjusted OR 1.51 [95% CI 1.03–2.21]). Cognitive performance was better in younger participants, those without depression, and those with a higher level of education. The adjusted ORs for each predictor are shown in Table 1.

Among participants with diabetes, cognitive performance was worse in patients who were taking metformin (adjusted OR 2.23 [95% CI 1.05–4.75]). MMSE scores were lower in participants with diabetes who used metformin (mean score ± SD 22.8 ± 5.5) than in those who did not use metformin (24.7 ± 4.4). Participants with diabetes who had vitamin B_{12} levels <250 ρmol/L also had worse cognitive performance (2.29 [1.12–4.66]). MMSE scores were lower among those with serum vitamin B_{12} <250 ρmol/L (22.9 ± 4.7) than those with higher levels (25.0 ± 4.7).

Each 1-year increase in age was associated with an 8% increased risk of impaired cognitive performance (OR 1.08 [95% CI 1.03–1.13]). A secondary or

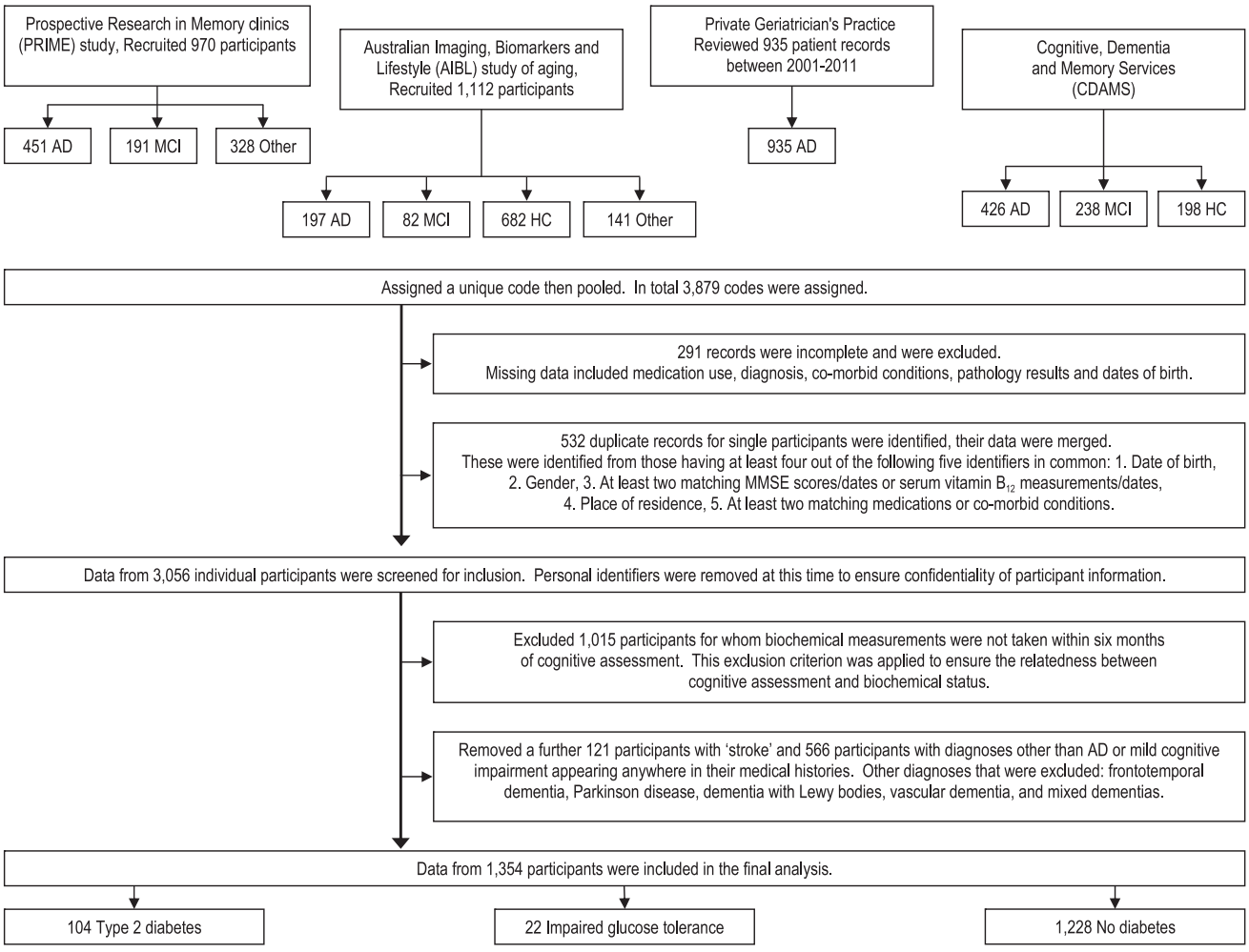

Figure 1—*Recruitment, inclusion, and exclusion.*

Table 1—*Cognitive performance in 1,354 participants who had AD or MCI or were cognitively intact*

	Model not adjusted for serum vitamin B_{12} levels			Model adjusted for serum vitamin B_{12} levels†		
Categories, by predictor*	OR	95% CI	*P* value	OR	95% CI	*P* value
Variable						
Diabetes						
Type 2 diabetes (*n* = 104)	1.51	1.03–2.21	0.033	*1.49*	*1.02–2.19*	*0.039*
Impaired glucose tolerance (*n* = 22)‡	0.79	0.34–1.85	0.584	*0.81*	*0.34–1.90*	*0.624*
No diabetes (*n* = 1,228)	—	—	—	—	—	—
Model adjusters						
Age (*n* = 1,354)	1.11	1.09–1.12	<0.001	*1.10*	*1.09–1.12*	*<0.001*
Sex						
Male (*n* = 549)	0.88	0.70–1.10	0.257	*0.86*	*0.69–1.08*	*0.186*
Female (*n* = 805)	—	—	—	—	—	—
Depression						
Yes (*n* = 375)	1.61	1.27–2.04	<0.001	*1.66*	*1.30–2.10*	*<0.001*
No (*n* = 979)	—	—	—	—	—	—
Level of education§						
Tertiary (*n* = 510)	0.12	0.08–0.17	<0.001	*0.12*	*0.08–0.17*	*<0.001*
Secondary (*n* = 674)	0.36	0.26–0.49	<0.001	*0.36*	*0.26–0.50*	*<0.001*
Primary (*n* = 170)	—	—	—	—	—	—

All models were adjusted for age, sex, reported history of depression, and level of education. Values obtained in the model that also was adjusted for serum vitamin B_{12} levels are italicized. *Reference levels for categories were female sex, no reported history of depression, and having attained a primary level of education only (up to 6 years of schooling). †Serum vitamin B_{12} status was defined as low (serum levels <250 ρmol/L) or normal (serum levels >250 ρmol/L). The reference level for serum vitamin B_{12} status was normal. ‡Impaired glucose tolerance was by self-report or review of patient's medical history. §Categories for level of education were tertiary (>13 years of schooling), secondary (6–13 years of schooling), and primary (<6 years of schooling).

tertiary level of education was the strongest predictor of better cognitive performance in patients with diabetes. The adjusted ORs for each variable are shown in Table 2.

CONCLUSIONS—In our series, patients with diabetes who were taking metformin had worse cognitive performance than participants who were not taking metformin. Our observations agree with those previously reported by Imfeld and colleagues (22), in particular that patients who are taking metformin may be at an increased risk for cognitive impairment. This association was weakened after adjusting for serum vitamin B_{12} levels; thus any effect metformin has on cognitive performance may be at least partially mediated by altering serum vitamin B_{12} levels.

Alternatively, patients who are prescribed metformin may have worse glycemic control or diabetes-related complications than patients with diabetes who are not prescribed metformin, so there remains the potential for confounding, despite restricting the analysis to only patients with diabetes. However, because metformin is a first-line pharmacotherapy for the treatment of type 2 diabetes, this would seem unlikely (30).

There was insufficient information regarding the duration of metformin use, the severity of diabetes (e.g., HbA_{1c} levels), duration of diabetes, or use of other antidiabetic drugs to enable us to investigate these effects in our study, particularly because these findings were based on a small sample. We recommend a larger study to examine the effect of dose and duration of metformin use, and the effects of other antidiabetic agents using a battery of cognitive assessments and following participants over a number of years.

Calcium supplements have previously been reported to reverse vitamin B_{12} deficiency induced by metformin. In this study, patients with diabetes who used calcium supplements were less likely to be cognitively impaired. However, calcium supplements have been reported to be associated with an increased risk for myocardial infarction in postmenopausal women and in patients with chronic kidney disease (31–34). In contrast, a recent meta-analysis indicates that supplementation with both vitamin D and calcium is associated with a reduction in mortality compared with vitamin D supplementation alone (35). Because this population already has increased cardiovascular risk (36), the safety of calcium supplementation in patients with diabetes treated with metformin would need to be established before such interventions could be recommended.

Patients with diabetes are at an increased risk for AD (10). In diabetes, amylin aggregation destroys the β-cells of the pancreas (37). By the same mechanism, a protein misfolding disorder may be related to aggregation of amyloid plaques. Metformin is a widely prescribed first-line monotherapy for type 2 diabetes but is associated with vitamin B_{12} deficiency and peripheral neuropathy. A case-control study of more than 14,000 patients reported that long-term metformin use was associated with an increased risk for AD in those ≥65 years old (22). Metformin at pharmacological doses was reported to increase the expression of β-secretase in cell culture; this may be a possible disease mechanism (21). Alternatively, metformin also impairs absorption of vitamin B_{12} via a drug interaction that occurs at the distal ileum. Low serum vitamin B_{12} levels are associated with AD and other neurodegenerative diseases (38).

Cognitive performance of patients with diabetes was measured using the MMSE. The MMSE may be inadequate for detecting differences between higher functioning adults (39). The MMSE is sensitive to age, depression, and level of education, so all models were adjusted for these factors. Most of our subjects were

Table 2—*Cognitive performance in 126 patients with either type 2 diabetes or impaired glucose tolerance*

	Model not adjusted for serum vitamin B_{12} levels			Model adjusted for serum vitamin B_{12} levels†		
Categories, by predictor*	OR	95% CI	*P* value	OR	95% CI	*P* value
Variables						
Metformin						
Yes (*n* = 35)	2.23	1.05–4.75	0.037	*1.75*	*0.81–3.78*	*0.158*
No (*n* = 91)	—	—	—	—	—	—
Calcium supplements						
Yes (*n* = 44)	0.47	0.22–1.02	0.056	*0.41*	*0.19–0.92*	*0.030*
No (*n* = 82)	—	—	—	—	—	—
Model adjusters						
Age (*n* = 1,354)	1.08	1.03–1.13	0.001	*1.08*	*1.03–1.13*	*0.002*
Sex						
Male (*n* = 59)	0.54	0.27–1.09	0.086	*0.49*	*0.24–1.01*	*0.052*
Female (*n* = 67)	—	—	—	—	—	—
Depression						
Yes (*n* = 40)	0.95	0.45–2.00	0.884	*0.96*	*0.45–2.05*	*0.916*
No (*n* = 86)	—	—	—	—	—	—
Level of education‡						
Tertiary (*n* = 28)	0.02	0.01–0.08	<0.001	*0.03*	*0.01–0.11*	*<0.001*
Secondary (*n* = 73)	0.26	0.11–0.63	0.003	*0.25*	*0.10–0.62*	*0.002*
Primary (*n* = 25)	—	—	—	—	—	—

All models were adjusted for age, sex, reported history of depression, and level of education. Values obtained in the model that also was adjusted for serum vitamin B_{12} levels are italicized. *Reference levels for categories were "not taking metformin," female sex, no reported history of depression, and having attained a primary level of education only (up to 6 years of schooling). †Serum vitamin B_{12} status was defined as low (serum levels <250 ρmol/L) or normal (serum levels >250 ρmol/L). The reference level for serum vitamin B_{12} status was normal. ‡Categories for level of education were tertiary (>13 years of schooling), secondary (6–13 years of schooling), and primary (<6 years of schooling).

assessed during routine clinical care by clinicians who use the MMSE as part of their standard assessment. In Australia, documentation of cognitive impairment using an MMSE score and improvement while receiving therapy is required to obtain subsidized antidementia therapies (40). More comprehensive assessment tools may be preferable for use in future investigations of cognitive impairment in at-risk populations such as those we have studied.

Increased monitoring of cognitive ability in patients with diabetes who use metformin is warranted, particularly among older adults (aged older than 50 years). Vitamin B_{12} supplements are inexpensive and may improve the cognitive outcomes of patients with diabetes. Adequately powered, prospective, controlled trials are warranted to investigate further the association between diabetes, cognitive decline, and the effect of metformin therapy, as well as the possible amelioration using vitamin B_{12} and/or calcium supplementation.

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E.M.M. reviewed the literature, collected biochemical measurements from study host sites, analyzed and interpreted the data, and drafted the manuscript. A.G.M., M.A.K., R.P.C., and D.A.W. interpreted the data and reviewed the manuscript. D.A. interpreted the data and reviewed the manuscript, collected the PRIME data and managed the PRIME study database, and collected the AIBL data and managed the AIBL database. H.B., M.W., and K.B. interpreted the data and reviewed the manuscript and collected the PRIME data and managed the PRIME study database. K.A.E., A.I.B., N.G.F., R.M., C.S., and C.R. interpreted the data and reviewed the manuscript and collected the AIBL data and managed the AIBL database. E.M.M. is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

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References

1. González EL, Johansson S, Wallander MA, Rodríguez LA. Trends in the prevalence and incidence of diabetes in the UK: 1996-2005. J Epidemiol Community Health 2009;63:332–336
2. Monesi L, Baviera M, Marzona I, Avanzini F, Monesi G, Nobili A, et al. Prevalence, incidence and mortality of diagnosed diabetes: evidence from an Italian population-based study. Diabet Med 2012;29:385–392
3. Boyle JP, Thompson TJ, Gregg EW, Barker LE, Williamson DF. Projection of the year 2050 burden of diabetes in the US adult population: dynamic modeling of incidence, mortality, and prediabetes prevalence. Popul Health Metr 2010;8:29–41
4. Zhang YH, Ma WJ, Thomas GN, et al. Diabetes and pre-diabetes as determined by glycated haemoglobin A1c and glucose levels in a developing southern Chinese population. PLoS One 2012;7:e37260
5. Maga A, de Courten M, Dan L, et al. *American Samoa NCD Risk Factors STEPS Report*. Suva, Fiji, Department of Health, World Health Organization, 2007
6. Stratton IM, Adler AI, Neil HA, et al. Association of glycaemia with macrovascular and microvascular complications of type 2 diabetes (UKPDS 35): prospective observational study. BMJ 2000;321:405–412
7. Stratton IM, Kohner EM, Aldington SJ, et al. UKPDS 50: risk factors for incidence and progression of retinopathy in type II diabetes over 6 years from diagnosis. Diabetologia 2001;44:156–163
8. The Diabetes Control and Complications Trial Research Group. The effect of intensive treatment of diabetes on the development and progression of long-term complications in insulin-dependent diabetes mellitus. New Engl J Med 1993;329:977–986
9. Ravona-Springer R, Luo X, Schmeidler J, et al. Diabetes is associated with increased rate of cognitive decline in questionably demented elderly. Dement Geriatr Cogn Disord 2010;29:68–74
10. Profenno LA, Porsteinsson AP, Faraone SV. Meta-analysis of Alzheimer's disease risk with obesity, diabetes, and related disorders. Biol Psychiatry 2009;67:505–512
11. Zhou G, Myers R, Li Y, et al. Role of AMP-activated protein kinase in mechanism of metformin action. J Clin Invest 2001;108:1167–1174
12. Huizinga MM, Roumie CL, Elasy TA, et al. Changing incident diabetes regimens: a Veterans Administration cohort study from 2000 to 2005. Diabetes Care 2007;30:e85
13. Proust-Lima C, Amieva H, Dartigues JF, Jacqmin-Gadda H. Sensitivity of four psychometric tests to measure cognitive changes in brain aging-population-based studies. Am J Epidemiol 2007;165:344–350
14. Tomkin GH, Hadden DR, Weaver JA, Montgomery DA. Vitamin-B12 status of patients on long-term metformin therapy. BMJ 1971;2:685–687
15. Andrès E, Vidal-Alaball J, Federici L, Loukili NH, Zimmer J, Kaltenbach G. Clinical aspects of cobalamin deficiency in elderly patients. Epidemiology, causes, clinical manifestations, and treatment with special focus on oral cobalamin therapy. Eur J Intern Med 2007;18:456–462
16. Stowers JM, Smith OA. Vitamin B 12 and metformin. BMJ 1971;3:246–247
17. de Jager J, Kooy A, Lehert P, et al. Long term treatment with metformin in patients with type 2 diabetes and risk of vitamin B-12 deficiency: randomised placebo controlled trial. BMJ 2010;340:c2181–c2187
18. Wile DJ, Toth C. Association of metformin, elevated homocysteine, and methylmalonic acid levels and clinically worsened diabetic peripheral neuropathy. Diabetes Care 2010;33:156–161
19. Bauman WA, Shaw S, Jayatilleke E, Spungen AM, Herbert V. Increased intake of calcium reverses vitamin B12 malabsorption induced by metformin. Diabetes Care 2000;23:1227–1231
20. Oliveira AM, Bading H. Calcium signaling in cognition and aging-dependent cognitive decline. Biofactors 2011;37:168–174
21. Chen Y, Zhou K, Wang R, et al. Antidiabetic drug metformin (GlucophageR) increases biogenesis of Alzheimer's amyloid peptides via up-regulating BACE1 transcription. Proc Natl Acad Sci U S A 2009;106:3907–3912
22. Imfeld P, Bodmer M, Jick SS, Meier CR. Metformin, other antidiabetic drugs, and risk of Alzheimer's disease: a population-based case-control study. J Am Geriatr Soc 2012;60:916–921
23. Li J, Deng J, Sheng W, Zuo Z. Metformin attenuates Alzheimer's disease-like neuropathology in obese, leptin-resistant mice. Pharmacol Biochem Behav 2012;101:564–574
24. Wang J, Gallagher D, DeVito LM, et al. Metformin activates an atypical PKC-CBP pathway to promote neurogenesis and enhance spatial memory formation. Cell Stem Cell 2012;11:23–35
25. Hsu CC, Wahlqvist ML, Lee MS, Tsai HN. Incidence of dementia is increased in type 2 diabetes and reduced by the use of sulfonylureas and metformin. J Alzheimers Dis 2011;24:485–493
26. Brodaty H, Woodward M, Boundy K, Ames D, Balshaw R; PRIME Study Group Patients in Australian Memory Clinics: baseline characteristics and predictors of decline at six months. Int Psychogeriatr 2011;23:1086–1096
27. Ellis KA, Bush AI, Darby D, et al. The Australian Imaging, Biomarkers and Lifestyle (AIBL) study of aging: methodology and baseline characteristics of 1112 individuals recruited for a longitudinal study of Alzheimer's disease. Int Psychogeriatr 2009;21:672–687
28. Crum RM, Anthony JC, Bassett SS, Folstein MF. Population-based norms for the Mini-Mental State Examination by age and educational level. JAMA 1993;269:2386–2391
29. Iverson GL. Interpretation of Mini-Mental State Examination scores in community-dwelling elderly and geriatric neuropsychiatry patients. Int J Geriatr Psychiatry 1998;13:661–666
30. Colagiuri S, Dickinson S, Girgis S, Colagiuri R. *National Evidence Based Guideline for Blood Glucose Control in Type 2 Diabetes*. Canberra, Diabetes Australia and the NHMRC, 2009
31. Li K, Kaaks R, Linseisen J, Rohrmann S. Associations of dietary calcium intake and calcium supplementation with myocardial infarction and stroke risk and overall cardiovascular mortality in the Heidelberg

cohort of the European Prospective Investigation into Cancer and Nutrition study (EPIC-Heidelberg). Heart 2012;98:920–925
32. Bolland MJ, Avenell A, Baron JA, et al. Effect of calcium supplements on risk of myocardial infarction and cardiovascular events: meta-analysis. BMJ 2010;341:c3691–c3699
33. Bollanc MJ, Grey A, Avenell A, Gamble GD, Reid IR. Calcium supplements with or without vitamin D and risk of cardiovascular events: reanalysis of the Women's Health Initiative limited access dataset and meta-analysis. BMJ 2011;342:d2040–d2048
34. West SL, Swan VJ, Jamal SA. Effects of calcium on cardiovascular events in patients with kidney disease and in a healthy population. Clin J Am Soc Nephrol 2010;5(Suppl. 1):S41–S47
35. Rejnmark L, Avenell A, Masud T, et al. Vitamin D with calcium reduces mortality: patient level pooled analysis of 70,528 patients from eight major vitamin D trials. J Clin Endocrinol Metab 2012;97:2670–2681
36. Morrish NJ, Wang SL, Stevens LK, Fuller JH, Keen H. Mortality and causes of death in the WHO Multinational Study of Vascular Disease in Diabetes. Diabetologia 2001;44(Suppl. 2):S14–S21
37. Hayden MR, Tyagi SC, Kerklo MM, Nicolls MR. Type 2 diabetes mellitus as a conformational disease. JOP 2005;6:287–302
38. Moore E, Mander A, Ames D, Carne R, Sanders K, Watters D. Cognitive impairment and vitamin B12: a review. Int Psychogeriatr 2012;24:1–16
39. Lawrence de Koning AB, Werstuck GH, Zhou J, Austin RC. Hyperhomocysteinemia and its role in the development of atherosclerosis. Clin Biochem 2003;36:431–441
40. Ames D, Flynn E. Dementia services: an Australian view. In *Dementia*. Burns A, Levy R, Eds. London, Chapman and Hall Medical, 1994, p. 611–621