THERAPEUTIC ADVANCES in
Chronic Disease

Meta-analysis

Metformin and the risk of dementia based on an analysis of 396,332 participants

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Abstract

Background: AMPK has attracted widespread interest as a potential therapeutic target for age-related diseases, given its key role in controlling energy homeostasis. Metformin (Met) has historically been used to treat Type 2 diabetes and has been shown to counteract age-related diseases. However, studies regarding the relationship between Met and a variety of age-related classifications of cognitive decline have reported mixed findings.

Objective: To assess the potential effect of Met on the onset of dementia and discuss the possible biological mechanisms involved.

Methods: This study was registered in the PROSPERO database (CRD420201251468). PubMed, Embase, and Cochrane Library were searched from inception to 25 May 2021, for population-based cohort studies. Effect estimates with 95% confidence intervals (CIs) were pooled using the random-effects model. Meta-regression and subgroup analyses were performed to explore sources of heterogeneity and the stability of the results.

Results: Fourteen population-based cohort studies (17 individual comparisons) involving 396,332 participants were identified. Meta-analysis showed that Met exposure was significantly associated with reduced risk of all subtypes of dementias [relative risk (RR) = 0.79, 95% CI = 0.68–0.91; $p < 0.001$]. Conversely, no significant reduction in risk was observed for those who received Met monotherapy at the onset of vascular dementia (VD), Parkinson's disease (PD), and Alzheimer's disease (AD). The effect was more prominent in patients who had long-term Met exposure (⩾4 years) (RR = 0.38, 95% CI = 0.32–0.46; $p < 0.001$), while no such significant effect was found with short-term Met exposure (1–2 years) (RR = 1.20, 95% CI = 0.87–1.66; $p < 0.001$). Moreover, no association was observed for Met exposure in participants of European descent (RR = 1.01, 95% CI = 0.66–1.54; $p = 0.003$) compared with those from other countries.

Conclusion: Based on the evidence from population-based cohort studies, our findings suggest that the AMPK activator, Met, is a potential geroprotective agent for dementias, particularly among long-term Met users. Due to the significant heterogeneity among the included studies, we should interpret the results with caution.

Keywords: cohort study, dementia, diabetes mellitus, neurodegenerative disease, metformin

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Introduction

AMPK is well known for regulating whole-body energy metabolism. AMPK has sparked widespread interest as a potential therapeutic target for age-related diseases because of its critical role in energy homeostasis control.[1] Metformin (Met) is an AMPK activator, and there is growing evidence that suggests it can help prevent age-related diseases. Diabetes and pre-diabetes have been linked to accelerated cognitive decline. Patients with diabetes display an approximately twofold increased risk of dementia.[2] Studies also revealed that patients with Type 2 diabetes mellitus (T2DM) have a 2.2-fold increased risk of developing Parkinson's disease (PD).[3] Insulin resistance and consequent glucose metabolism in patients with

diabetes could play critical roles in the progression of Alzheimer's disease (AD) and dementia.[4,5] A number of researchers have focused on the effect of diabetes mellitus (DM) on the cognitive decline within the elderly population. Studies have shown that elderly patients with diabetes and impaired fasting glucose have different risk factors for cognitive impairment compared with elderly patients with normal glucose levels.[6] Evidence strongly supports the concept that insulin resistance plays a crucial role in both cognitive decline and dementia, which further suggests that when brain insulin signals are stimulated, the protective effect against cognitive deficits may be activated.[7] The link between diabetes and dementia is probably multifactorial, and the mechanisms may involve chronic low-grade inflammation, oxidative stress, atherosclerosis, amyloid-β deposition, brain insulin resistance with hyperinsulinemia, advanced glycation end products, and dysregulation of lipid metabolism.[8,9] Furthermore, some evidence suggests that insulin acts on the central nervous system to modulate behavior and systemic metabolism. Insulin sensitivity involving central and peripheral regions may be mediated by dopamine, suggesting a potential association between cognitive health and glucose metabolism.[10,11]

Met is a widely used, cost-effective, and safe drug for the treatment of T2DM.[12] The mechanism of action of Met is similar to caloric restriction, which depends on the activation of AMPK.[13] Animal studies have shown that both caloric restriction and Met can slow down the aging process.[14,15] Several *in vitro* experiments and animal studies have shown that Met affects brain function, including its inhibitory effect on mammalian target of rapamycin (mTOR) *via* activation of AMPK and suppression of tau hyperphosphorylation and inflammation.[16–18] Similarly, another study has shown that Met, and its derivatives, can improve the activity of human acetylcholinesterase (AChE) and inhibit beta-amyloid aggregation.[19] However, epidemiological researchers have reported inconsistencies concerning the studies that Met is related to an increased risk of neurodegenerative diseases (NDs).[3,20,21] Some studies suggested a reduced risk of NDs with Met treatment,[22–28] while others reported no association.[29–32] At present, there are few randomized controlled trials (RCTs) of Met in NDs. It is difficult to directly compare previous studies due to variations in study design, data quality, and so on. To better understand this issue, we conducted data analysis to comprehensively evaluate the link between Met exposure and the risk of dementias. Moreover, we investigated potential moderators, including study design, geographic regions, age and gender, age at T2DM diagnosis, sample size, length of Met exposure, dementia type, and methodologic quality.

Methods

This systematic review was carried according to a predefined protocol and in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA).

Search strategy and selection criteria

The cohort studies published in PubMed, Cochrane Library, and Embase were systematically searched from inception to 25 May 2021, by two independent investigators (S.J. and X.Z.) without time restrictions. We employ the following search strategies (including synonyms and near-synonyms words) that were associated with Met and dementias. The detailed search strategy and specific terms (metformin/Met) AND (neurodegenerative diseases OR vascular dementia/VD OR Parkinson Disease/PD OR Alzheimer Disease/AD OR Dementia OR Cognitive disorder/CD) AND (cohort/longitudinal/follow-up/prospective/retrospective studies) were used, which were searched as free text words and as MeSH/Emtree terms. Moreover, we manually scrutinized the reference lists of meta-analyses, reviews, reports, and other possibly relevant articles. When ⩾2 articles used the same cohort data, we preferred the most up-to-date one with full-text information available.

Eligibility criteria

Studies were considered appropriate when meeting the following criteria: (1) participants: patients previously exposed to Met, who had no history of dementias; (2) design: prospective or retrospective, population-based cohort studies and simultaneously, the primary outcome of the study was the incidence of various dementias reported in English; and (3) the calculation of association: relative risk (RR), hazard ratio (HR), and odds ratio (OR) or provided data. We excluded hospital-based or community-based

observational studies and those studies that did not provide adequate data to generate risk ratios for the association between Met exposure and risk of dementia.

Study selection, data collection, and data extraction

Two investigators (S.J. and X.Z.) independently extracted data by using standardized, predesigned extraction forms. Discrepancies were discussed between the researchers, and a consensus was reached. The following data were extracted: author, period/year of publication, design, geographic region, country of the population studied, matched for age and gender, patient age at diabetes diagnosis, sample size, length of Met exposure, dementia type, primary outcome reported, and estimates of the association of Met exposure with dementias.

Quality assessment

Two authors (S.J. and Y.D.) evaluated the methodological quality separately in accordance with the 9-star Newcastle–Ottawa scale (NOS) tool.[33] These are based on the main terms, which included representativeness and election of the participants, detection of exposure, assessment of denouements, and evaluation of follow-up. Any disagreement was resolved by a joint re-evaluation and consensus was reached. The cumulative NOS score of $\geqslant 7$ was considered a high-quality study.

Statistical analysis

All analyses were conducted using STATA (version 14.0; Stata Corp, College Station, TX). The main outcome was the pooled RR of dementias for Met use compared with the RR in non-Met users. Due to the anticipated heterogeneity of enrolled patients, we also used the Der Simonian and Laird random-effects model to calculate RR along with the 95% CIs.[34] In studies that did not report the RR of dementias, other risk measurements (HR or OR) were used and were considered as approximations of RR to compare the risks between Met exposure and dementias. When the incidence of outcome was relatively low, we proposed RR, HR, and OR to be comparable. In order to explain the confounding variables, adjusted RR was used for analysis. I^2 test was calculated to assess heterogeneity with an $I^2 \geqslant 50\%$ representing substantial heterogeneity.[35]

We first assessed whether Met use might reduce the risk of dementias. To test the potential sources of heterogeneity, we carried out several stratified analyses based on study design (prospective or retrospective cohort), geographic regions (the USA, Europe, and Asia), sample size ($<$10,000 or $\geqslant$10,000), patient age at diabetes diagnosis ($<$70 or $\geqslant$70 years), matched for age and sex (yes or no), length of Met exposure (1–2, 2–4, or $\geqslant$4 years), dementias type (dementia, PD, AD, VD, or CD), and methodologic quality (low or high). We also carried out meta-regression to examine the causes of inter-subgroup heterogeneity. We test publication bias by observing funnel plot symmetry, combined with Egger's or Begg's test.[36] Sensitivity analysis was conducted by the leave-one-out method. Furthermore, the trim-and-fill technique was used to further adjust the risk estimates.[37]

Results

Characteristics of included studies

The selection process is based on PRISMA and Meta-Analysis of Observational Studies in Epidemiology (MOOSE) guidelines (Figure 1). The 1022 potentially relevant citations were retrieved in the initial search and were reduced to 732 after removing duplicates. Subsequently, we excluded 699 irrelevant studies, and the remaining articles were screened by reading the full text. There were two studies that were published throughout multiple publications, but in the quantitative analysis, we treated them as one cohort.[32,38] We excluded non-population-based cohorts, reviews, meta-analyses, or other unqualified studies; 14 studies in total involving 10,479,530 participants (8,493,998 metformin exposure *versus* 1,985,532 controls) satisfied the inclusion criteria and ultimately were entered into the analysis.[3,20–32]

The baseline characteristics of the included studies are presented in Tables 1 and 2. Among the studies published between 2011 and 2020, four were performed in the United States,[22,26,28,32] three were from Europe,[21,27,30] and seven were from Asia.[3,20,23–25,29,31] Most of the studies (8 out of 14) were retrospective cohort studies, and 86% of the included studies (12 out of 14) had a NOS score $\geqslant$8. The sample size of the studies included ranged from 365 to 112,845 participants, with a median sample size of 28,309. The median length of Met exposure ranged from 1 to 6 years.

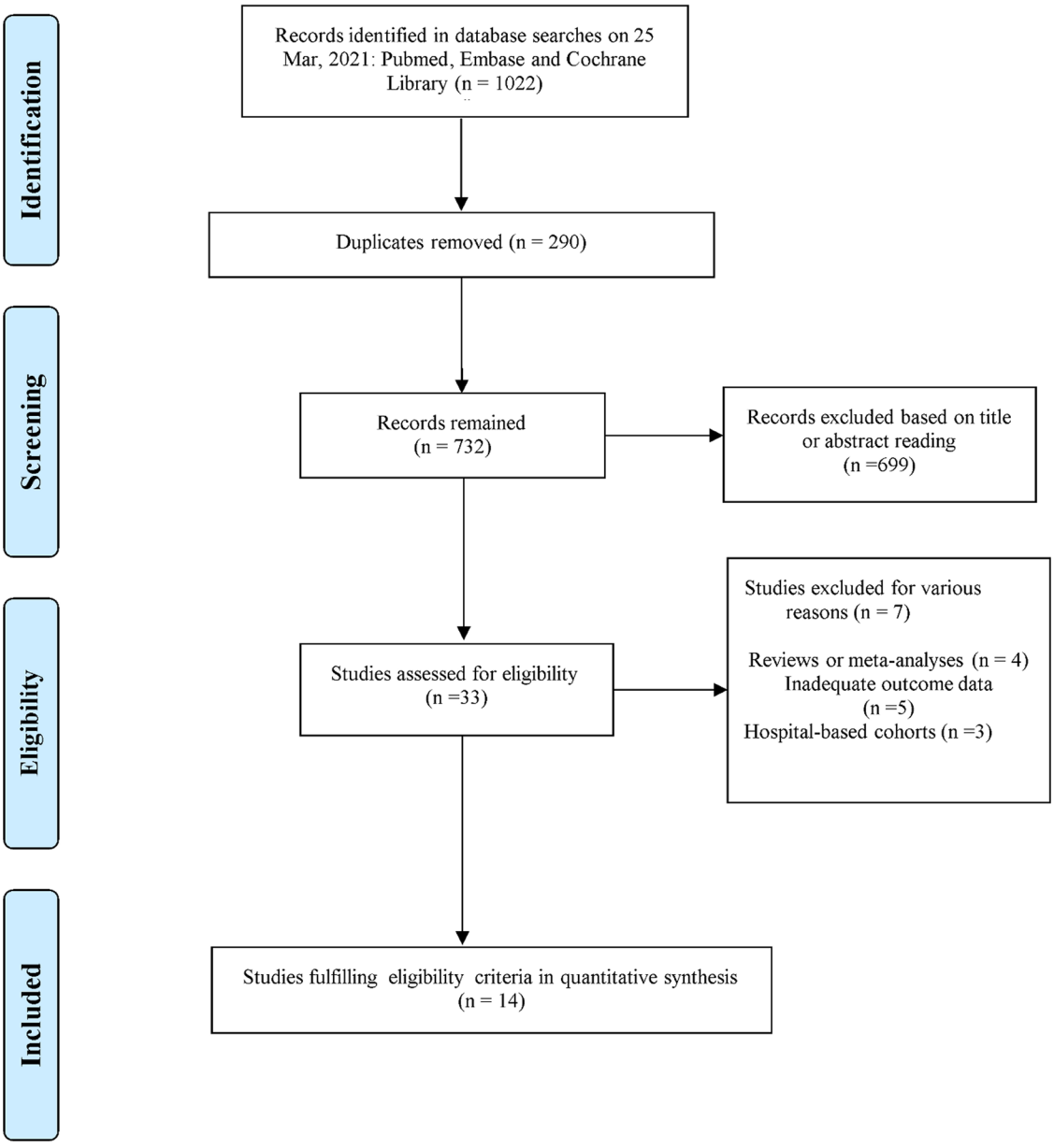

Figure 1. Flowchart of study selection based on PRISMA and MOOSE guidelines.

Six studies enrolled patients with Met and non-Met controls matched for age and gender.[20,24,25,27,29,31] Most studies identified dementia and NDs through medical records, according to the International Classification of Diseases (9th Revision, Clinical Modification; ICD-9-CM) or ICD-10.

Quality assessment

The quality evaluation is summarized in Table 3. Using the NOS tool for cohort studies, we found that a total of two studies had a moderate risk of bias and each study had two to three possible sources of bias.[21,26] Bias was most common when the adequacy of exposure time and treatment compliance was self-reported. In addition, all studies provided detailed information about participant drop-out and therefore were considered to have a low risk of bias in the reporting of results.

Effects of Met use on incidence of dementias

When we meta-analyzed the 14 studies, as shown in Figure 2, the results showed that the pooled

Table 1. Summary findings of the included studies.

Study	Year	Study design	Location	Observation period	Population, *n* (exposure *versus* control)	Age, years (mean/SD)	Control population	Reported outcomes
Samaras *et al.*[27]	2020	Prospective cohort study	Australia	NA	123 (67 *versus* 56)	78.25 (4.6) *versus* 80.0 (4.7)	The Sydney Memory and Ageing Study	Incident dementia and cognitive decline
Salas *et al.*[32]	2020	Retrospective cohort study	USA	1996–2015	112,845 (18,904 *versus* 93,941); 14,333 (1793 *versus* 12,540)	59.5 (7.8) *versus* 63.2(9.1); 59.9 (7.8) *versus* 64.3 (9.9)	Veterans Health Affairs (VHA); Kaiser Permanente Washington (KPW)	Risk of incident dementia
Chin-Hsiao *et al.*[23]	2019	Retrospective cohort	China, Taiwan	1999–2011	31,352 (15,676 *versus* 15,676)	63.5 ± 9.9 *versus* 63.4 ± 10.4	Taiwan's National Health Insurance	Risk of dementia
Shi *et al.*[28]	2019	Retrospective longitudinal cohort	USA	2004–2010	396 (219 *versus* 177)	63.2 ± 10.9	Veterans Affairs electronic medical record database	The risk of neurodegenerative disease (ND)
Kuan *et al.*[20]	2017	Prospective cohort study	China, Taiwan	2000–2010	9302 (4651 *versus* 4651)	64.7 (9.46) *versus* 64.7 (10.0)	Taiwan's National Health Insurance Research Database	Risk of dementia and PD
Brakedal *et al.*[21]	2017	Retrospective cohort study	Norway	2005–2014	10,2745 (94,349 *versus* 8396)	64.3 (11.6) *versus* 62.6 (10,7)	Norwegian Prescription Database	Risk of incident PD
Orkaby *et al.*[22]	2017	Retrospective cohort study	USA	2001–2012	14,562 (10,437 *versus* 4125); 14,078 (6763 *versus* 7315)	$65 \geqslant$ Age $<$ 75 years; Age $\geqslant$ 75 years	US veterans	Risk of dementia
Wang *et al.*[26]	2017	Retrospective cohort study	USA	2004–2012	41,204 (8393 *versus* 32,811)	74.6 ± 5.8	Male veterans in the United States	Age-related comorbidity (ARC) diagnoses
Heneka *et al.*[30]	2015	Prospective cohort study	Germany	2004–2010	5332 (1478 *versus* 3854)	$\geqslant$60; Age 60–69; Age 70–79; Age 80+	General population	Incidence of dementia
Cheng *et al.*[29]	2014	Prospective cohort study	China, Taiwan	2004–2009	1829 (1033 *versus* 796)	73.2 (6.0) *versus* 74.1 (6.5)	General population	Risk of dementia
Ng *et al.*[25]	2014	Prospective cohort study	Singapore	2003–2005	365 (204 *versus* 161)	67.0 (65.5) *versus* 67.6 (7.16)	Population-based Singapore Longitudinal Aging Study	Risk of cognitive impairment
Huang *et al.*[31]	2014	Retrospective cohort study	China, Taiwan	1997–2007	30,170 (4978 *versus* 25,192)	58.76 ± 14.0	Taiwanese population	The risk of AD
Wahlqvist et a1.[3]	2012	Retrospective cohort study	China, Taiwan	1996–2007	5313 (1879 *versus* 3431)	64.8 ± 9.55 *versus* 65.3 ± 9.44	General population	Risk of Parkinson's disease
Hsu *et al.*[24]	2011	Prospective cohort study	China, Taiwan	2000–2007	12,383 (1864 *versus* 10,519)	$>$50	Taiwan's National Health Insurance database	Incidence of dementia

AD, Alzheimer's disease; NA, not applicable; PD, Parkinson's disease; SD, standard deviation.

Table 2. Characteristics of included studies of metformin use in relation to risk of NDs: exposure and outcome assessment, results, and measure of associations.

Study	Ascertainment of metformin exposure	Ascertainment of outcome	Results	Type of dementia	Measure of associations	Adjusted variables
Samaras *et al.*[27]	DM with Met	Dementia/*DSM*-IV	Incident dementia was significantly higher in DM-noMF compared with DM1MF (odds ratio, 5.29; 95% CI, 1.17–23.88)	Dementia	OR	Adjusted for sex and mean-centered values of age and years of education
Salas *et al.*[32]	Early diabetic with Met	Dementia/ICD-10-CM	There was no association between initiation of metformin (*versus* no initial treatment) and incident dementia in VHA (HR = 1.04; 95% CI: 0.95–1.13) or KPW (HR = 0.81; 95% CI: 0.51–1.28)	Dementia	HR	Inverse probability of treatment weighted data with robust
Chin-Hsiao *et al.*[23]	DM with Met	Dementia/ICD-9	Analyses in the matched cohort showed an overall HR of 0.707 (0.632–0.791)	Dementia	HR	NA
Shi *et al.*[28]	DM with Met	ND/ICD-9-CM	Compared with no metformin use, 2–4 years and >4 years of metformin exposure were significantly associated with lower risk of ND (aHR = 0.62, 95% CI = 0.45–0.85; aHR = 0.19, 95% CI = 0.12–0.31, respectively)	AD, PD, HD, dementia, and mild cognitive impairment	HR	PSW
Kuan *et al.*[20]	T2DM with Met	PD/ICD-9-CM Dementia/ICD-9-CM	The metformin cohort exhibited a higher risk of PD than the noMF cohort (HR: 2.27, 95% CI: 1.68–3.07). The metformin cohort had an increased risk of all-cause dementia (HR: 1.66, 95% CI: 1.35–2.04)	Dementia; PD; AD; vascular dementia	HR	Adjustment for age; sex; Charlson Comorbidity Index
Brakedal *et al.*[21]	DM with Met	PD/ICD-10	Glitazone use was associated with a significantly lower incidence of PD compared with metformin only use (HR: 0.72; 95% CI, 0.55–0.94; $p < 0.01$)	PD	HR	NA
Orkaby *et al.*[22]	DM with Met	Dementia/ICD-9	After PS IPTW adjustment, results remained significant in veterans, 75 years of age (HR = 0.89; 95% CI = 0.79–0.99), but not for those <75 years of age (HR = 0.96; 95% CI = 0.87–1.05)	Dementia; AD: vascular dementia	HR	Inverse probability of treatment weighting
Wang *et al.*[26]	DM with Met	Dementia/ICD-10	Metformin reduced likelihoods of CVD (18.8%), cancer (3.9%), dementia (3.8%), depression (15.6%), and FRD (23.8%)	Dementia	OR	NA
Heneka *et al.*[30]	DM with Met	Dementia/ICD-10	NA	Dementia	RR	NA
Cheng *et al.*[29]	Early diabetic with Met	Dementia/ICD-9-CM	The relative rate of dementia was 1.22 (95% CI: 0.78–1.91) for those taking sulfonylureas ($n = 796$) compared with those taking metformin ($n = 1033$)	Dementia	HR	Adjusted for age, sex, hypertension, hyperlipidemia, and cerebrovascular disease
Ng *et al.* [25]	DM with Met	Cognitive dysfunction/MMSE	Metformin use showed a significant inverse association with cognitive impairment in longitudinal analysis (OR: 0.49, 95% CI = 0.25–0.95)	Cognitive impairment	OR	Adjusted for age, gender, education, and so on
Huang *et al.*[31]	DM with Met	AD/ICD-9	Metformin as monotherapy 0.69 (0.28–1.71)	AD	HR	Adjusted for age, sex, comorbidities
Wahlqvist *et al.*[3]	Early diabetic with Met only	PD/ICD-9-CM or A code	NR	PD	HR	Adjusted for monthly income; delete those diabetes patients who have used insulin
Hsu *et al.*[24]	Early diabetic with Met	Dementia/ICD-9-CM or A-code	For T2DM, compared with no medication, metformin alone to 0.76 (0.58–0.98)	Dementia	HR	Adjusted for age group, gender, type of stroke, and CCI score

AD, Alzheimer's disease; aHR, adjusted hazard ratio; CCI, Charlson Comorbidity Index; CI, confidence interval; CVD, cardiovascular disease; DM, diabetes mellitus; *DSM*-IV, *Diagnostic and Statistical Manual of Mental Disorders* (4th ed.); FRD, frailty related diseases; HD, Huntington's Disease; HR, hazard ratio; ICD, International Classification of Diseases; ICD-10-CM, International Classification of Diseases (10th Revision, Clinical Modification); IPTW, inverse-probability-of-treatment-weight; MMSE, Mini-Mental State Examination; NA, not applicable; ND, neurodegenerative disease; noMF, nonmetformin; NR, not reported; OR, odds ratio; PD, Parkinson's disease; PS, propensity score; PSW, propensity score weight; RR, relative risk; T2DM, Type 2 diabetes mellitus.

Table 3. Methodological quality score of the included studies based on the Newcastle–Ottawa scale (NOS) tool.

Study	Year	Study design	Selection				Comparability	Exposure/outcome			Total score	Risk of bias
			Representativeness of cohort *	Selection of control cohort *	Ascertainment of exposure *	Outcome not present at start*	Comparability of cohorts**	Assessment of outcome*	Length of follow-up*	Adequacy of follow-up*	Total score 9*	
Cheng *et al.*[29]	2014	Prospective cohort study	*	*	*	*	**	*	*		8	Low
Wahlqvist et a1.[3]	2012	Retrospective cohort study	*	*	*	*	**	*	*		8	Low
Hsu *et al.*[24]	2011	Prospective cohort study	*	*	*	*	**	*	*		8	Low
Kuan *et al.*[20]	2017	Prospective cohort study	*	*	*	*	**	*	*	*	9	Low
Ng *et al.*[25]	2014	Prospective cohort study	*	*	*	*	**	*	*	*	9	Low
Heneka *et al.*[30]	2015	Prospective cohort study	*	*	*	*	**	*	*		8	Low
Brakedal *et al.*[21]	2017	Retrospective cohort study	*	*	*	*	**	*			7	High
Orkaby *et al.*[22]	2017	Retrospective cohort study	*	*	*	*	**	*	*		8	Low
Wang *et al.*[26]	2017	Retrospective cohort study	*	*	*	*	*		*		6	High
Huang *et al.*[31]	2014	Retrospective cohort study	*	*	*	*	**		*	*	8	Low
Chin-Hsiao *et al.*[23]	2019	Retrospective cohort study	*	*	*	*	**	*	*	*	9	Low
Samaras *et al.*[27]	2020	Prospective cohort study	*	*	*	*	**	*	*		8	Low
Shi *et al.*[28]	2019	Retrospective cohort study	*	*	*	*	**	*	*	*	9	Low
Salas *et al.*[32]	2020	Retrospective cohort study	*	*	*	*	**	*	*		8	Low

Each asterisk (*) represents one Newcastle–Ottawa Scale score.

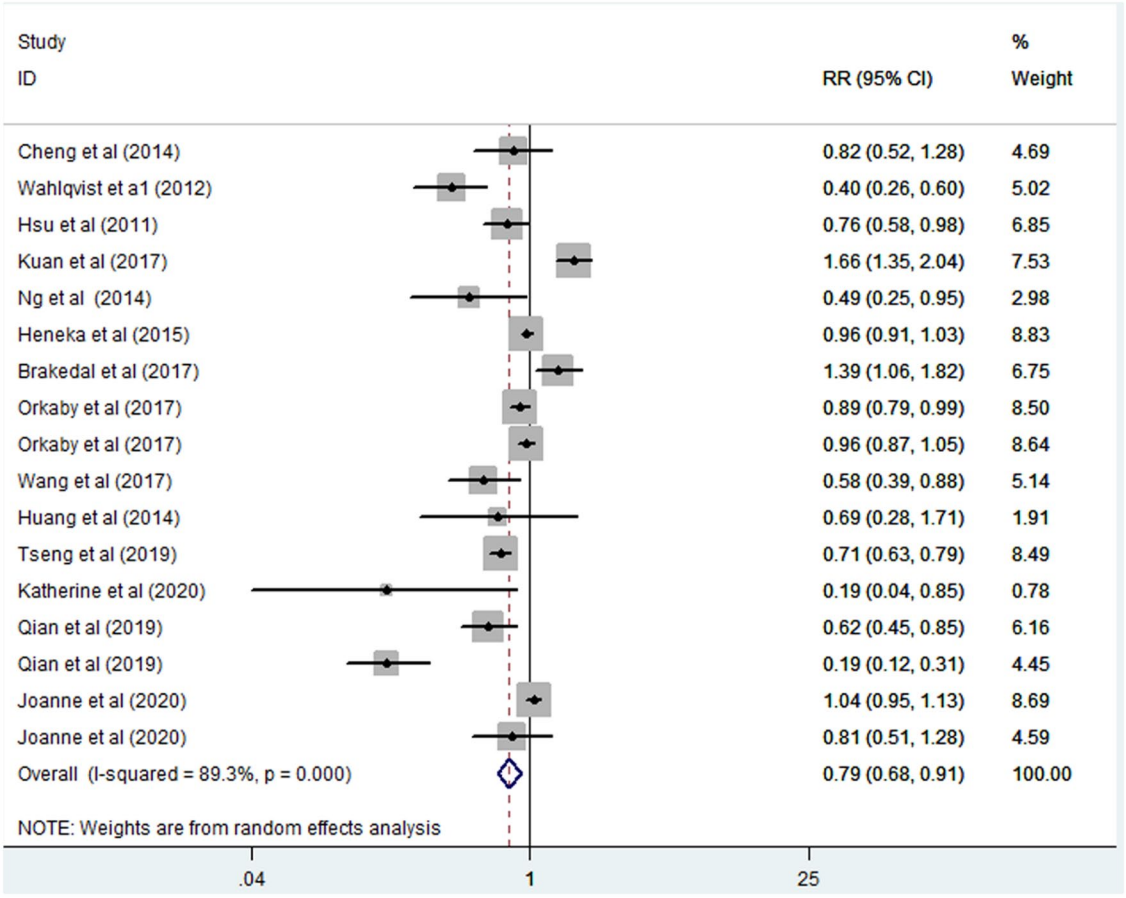

Figure 2. Relative risk (RR) for association of dementias with metformin exposure.

RR of dementias reached 0.79 (95% CI = 0.68–0.91) in Met users compared with non-Met controls. Heterogeneity among studies was high ($I^2 = 89.3\%$; $p < 0.001$).

Subgroup analysis and meta-regression

Subgroup analysis was conducted to determine whether the baseline age of the study population affected the incidence of dementias (Table 4). A significant reduction in the incidence of dementias was observed in patients <70 years (0.77; 95% CI = 0.62–0.96). Conversely, there was no significant difference for those ⩾70 years (0.94; 95% CI = 0.86–1.02), particularly for those >75 years (0.96; 95% CI = 0.91–1.02). For specific types of dementia, we noted that the Met was closely related to a lower risk of subsequent dementia than non-Met use (RR = 0.81, 95% CI = 0.70–0.93), while there was no significant difference in PD (RR = 0.70, 95% CI = 0.32–1.52), AD (RR = 0.94, 95% CI = 0.70–1.26), CD (RR = 0.67, 95% CI = 0.36–1.24), and VD (RR = 1.22, 95% CI = 0.79–1.88) (Supplementary Figure S1). Meanwhile, we also explored the effect of glycemic statuses on the incidence of dementia after Met treatment. In the studies collected for analysis, 25% included patients with hyperglycemia, while the remaining studies included patients with T2DM. These results indicated that Met treatment did not reduce the incidence of dementia in patients with early onset of diabetes (0.75; 95% CI = 0.55–1.03), while it significantly lessened the incidence of dementia in patients with late-onset diabetes (0.79; 95% CI = 0.66–0.94). We further investigated whether the length of exposure post-Met treatment impacts the incidence of dementia. We found that the risk of dementia was significantly decreased with the increased length of Met exposure ⩾2 to 4 years (0.68; 95% CI = 0.59–0.79), especially in long-term Met exposure (⩾4 years) (RR = 0.38, 95% CI = 0.32–0.46; $p < 0.001$). However, short-term Met exposure (1–2 years) had no significant effect (RR = 1.20,

Table 4. Subgroup analyses for the effect of metformin exposure on risk of NDs.

Variables	RR	95% CI	I^2, %	No. of studies	*p* for Interaction
Overall	0.79	0.68–0.91	89.3	14	NA
Study design					0.009
Prospective cohort	0.88	0.64–1.20	86.9	6	
Retrospective cohort	0.73	0.60–0.87	90.5	8	
Geographic regions					
USA	0.72	0.58–0.89	90.4	4	0.003
Europe	1.01	0.66–1.54	82.2	3	
Asia	0.74	0.51–1.08	90.8	7	
Population sample size					0.472
<10,000	0.59	0.40–0.89	93.3	7	
⩾10,000	0.88	0.77–1.01	82.4	7	
Age, years					0.382
<70	0.77	0.62–0.96	92.3	9	
⩾70	0.94	0.86–1.02	48.9	6	
>75	0.96	0.91–1.02	0	3	
Type of NDs					0.424
Dementia	0.81	0.70–0.93	90.2	10	
PD	0.70	0.32–1.52	95.8	4	
AD	0.94	0.70–1.26	65.5	4	
VD	1.22	0.79–1.88	74.1	2	
Cognitive disorder	0.67	0.36–1.24	12.7	2	
Glycemic status					
Early diabetic with met	0.75	0.55–1.03	83.4	4	0.244
T2D with Met	0.79	0.66–0.94	91.2	10	
Length of Met exposure (years)					<0.001
1–2	1.20	0.87–1.66	78.3	3	
2–4	0.68	0.59–0.79	0	3	
⩾4	0.38	0.32–0.46	0	2	
Matched for age and sex					0.021
Yes	0.76	0.47–1.25	86.2	6	
No	0.77	0.66–0.89	90.8	8	
Methodologic quality					0.218
Moderate (6–7)	0.91	0.39–2.14	91.9	2	
High (⩾8)	0.77	0.66–0.89	89.7	12	

AD, Alzheimer's disease; CD, cognitive disorder; CI, confidence interval; Met, metformin; NA, not applicable; ND, neurodegenerative disease; PD, Parkinson's disease; RR, relative risk; T2D, type 2 diabetes; VD, vascular dementia.

95% CI = 0.87–1.66; $p < 0.001$). Despite the fact that subgroup analysis was performed previously, significant heterogeneity remained between some studies. Therefore, a univariate meta-regression analysis was conducted, which indicated that the heterogeneity could be due to various factors, including study design, geographic regions, matched for age and gender, patient age at T2DM diagnosis, sample size, length of Met exposure, and dementia type (all $p < 0.05$). However, the remaining heterogeneity may result from other potential baseline changes between individuals enrolled in each study.

Sensitivity analyses and publication bias

Sensitivity analyses were performed by using the leave-one-out method to examine the stability of the results. We found that no individual study significantly changed the pooled RRs (lowest RR = 0.19, 95% CI = 0.12–0.31; highest RR = 1.66, 95% CI = 1.35–2.04). Sensitivity analyses were carried out by summarizing and estimating studies that include only the use of time-to-event risk assessment and HRs. HRs of 10 studies were pooled, yielding a summary estimate of 0.80 (95% CI = 0.67–0.95; $p = 0.002$), which was similar to the prior result. The findings of the contour enhancement funnel chart indicate no potential evidence of publication bias. The Begg's test for small-study effects was non-significant ($p = 0.091$), and the Egger test was also non-significant ($p = 0.078$). Furthermore, the trim-and-fill method adjusted for publication bias showed no potential for missing studies (Supplementary Figure S2).

Discussion

Principal findings

According to the present comprehensive meta-analysis with 396,332 participants, we demonstrated Met plays a beneficial role in reducing the risk of dementia. After adjusting for potential publication bias, the results remained consistent. Moreover, our results indicate that Met treatment reduces the future development of dementia for patients with T2DM, whose age at diagnosis is <70 years, and with Met exposure ⩾2 years. With a population sample size of <10,000, this finding is stable only in the United States.

Comparisons with previous studies

Our results are consistent with one systematic review with meta-analyses,[39] which validated our findings of a decreased dementia risk in patients with Met exposure. However, the review article summarized a range of evidence, including one case as control, two RCTs, four cross-sectional, and seven cohorts. Some studies found that the use of Met had a negative or neutral effect on patients with diabetes. The meta-analysis conducted by Ye *et al.*[40] failed to demonstrate a protective effect (RR = 0.79, 95% CI = 0.82–1.01), in which only six observational studies assessed the effect of Met on dementia. The latest meta-analysis performed by Ping *et al.*[41] concluded that Met had no beneficial effect on dementias (OR = 1.04, 95% CI = 0.92–1.17). Furthermore, it may increase the risk of PD development (OR = 1.66, 95% CI = 1.14–2.42). After that, several observational studies with large sample sizes were reported.[27,28,32] Among them, Samaras *et al.*[27] and Shi *et al.*[28] demonstrated the protective effect of Met on incidental dementia. The above studies made it possible to include much more comparisons for the evaluation of the relationship between Met and risk of dementias in the present meta-analysis. Furthermore, the previously published meta-analyses came mostly from non-population-based cohorts or small sample RCT studies with a high risk of bias. This study is the first involving representative populations with all dementia types to meta-analyze the relationship between Met use and subsequent dementia risk from high-quality population-based cohort studies rather than previously separated or narrative ones.

The previous meta-analysis found that Met had no beneficial effect on PD and might increase the risk of PD development, which may be related to the previous error combination and the latest research results. As for the two cohort studies included, the results should be interpreted cautiously. Wahlqvist *et al.*[3] was included and concluded that patients with T2DM who used sulfonylureas alone but not on oral anti-hyperglycemic agents had an increased risk of PD (HR = 1.57, 95% CI, 1.15–2.13). However, Met alone did not increase the risk (HR = 0.95, 95% CI, 0.53–1.71). Another study by Shi *et al.*[28] reported that the significantly reduced risk of PD was only associated with more than 4 years of Met treatment as compared with the non-Met

exposure group (aHR = 0.04, 95% CI = 0–0.37), and there was no association with ⩾2–4 years of Met exposure (aHR = 0.59, 95% CI = 0.29–1.17).

Potential mechanisms

Our meta-analysis suggests that Met treatment can decrease the risk of developing dementia, which raises a question worth addressing: How does the cheap drug Met prevent dementia? There are several underlying factors that clarify the potential associations between Met use and the subsequent decreased dementia risk. First, Met can penetrate the blood–brain barrier and thus act centrally to exert its neuroprotective function; its concentration in cerebrospinal fluid is nearly 1/10 of that in plasma.[42] Second, Met is an extensively used pharmacological agent that improves whole-body insulin sensitivity. Here, insulin resistance affects Adenosine triphosphate (ATP) production and Reactive oxygen species (ROS) release in neurons and astrocytes and in mixed glial cell cultures. Many studies have shown that High-Fat Diet (HFD)-induced insulin resistance leads to significant impairment of mitochondrial function in the brain, which can be mitigated by exercise and Met, both of which improve insulin sensitivity in the brain.[43] Third, AMPK, insulin, and glucose transporters serve as mediators of the Met effect in AD. Met enhances neuronal bioenergetics by activating AMPK and autophagy, promotes nerve repair, and reduces toxic protein aggregation in nervous system diseases.[44] Met protects against Aβ-induced mitochondrial dysfunction by activating the AMPK pathway in human neural stem cells. It may also act directly on insulin signaling in the brain, which makes Met treatment even more important because it can improve changes in glucose metabolism in the brain.[45] Finally, Met functions as an acetylcholinesterase (AChE) inhibitor or an antioxidant. AChE is a cholinesterase responsible for the hydrolysis of acetylcholine (ACh), which is important pathogenesis of AD. Several *in vivo* studies have evaluated the effect of Met on AChE activity. They hypothesized that Met's inhibition of this key enzyme is associated with neurodegeneration and may be responsible for preventing cholinergic dysfunction in T2DM.[46] Similarly, the results of numerous studies have demonstrated that elevated levels of oxidative stress of its markers, such as oxidized lipids and proteins, play an important role in the pathogenesis of AD. Studies assessing the effects of Met treatment on oxidative stress, as well as its anti-inflammatory response, have been recently reported. These results all imply the anti-inflammatory properties of Met.[44]

Energy metabolism has long been considered to have an effect on the etiology of dementias, and herein, some of the relevant signaling pathways and biological mechanisms that are related to Met's therapeutic potential in neurodegeneration are briefly discussed (Supplementary Figure S3). It mainly includes the following points: (1) In AMPK signaling, Met is an AMPK activator that suppresses hepatic glucose production and increases insulin-mediated glucose uptake. Dysregulation of AMPK is connected with insulin resistance, T2DM, and neuroinflammation.[47] Met restrains complex I of the electron transport chain, which is necessary for mitochondrial respiration, resulting in an energy deficit, which indirectly activates the AMPK pathway.[48] (2) In glucose metabolism, glucose is a fundamental energy substrate necessary to sustain neuronal activity and is absorbed *via* glucose transporters expressed in the brain endothelium, neurons, and astrocytes.[49] Met reduces advanced glycation end products,[9] which promote tissue degeneration and the microvascular complications of hyperglycemia in neural, renal, and vascular tissues. Preclinical and clinical studies have shown that Met has neuroprotective effects on brain structure and function. (3) In insulin signaling, insulin plays an important part in the brain. It serves as a hormonal signal to control ingestion of food, body weight, and metabolic homeostasis.[50] Met prevented neuronal insulin resistance, which has shown AD characteristics in cellular models.[51] Met decreases blood glucose levels by suppressing gluconeogenesis in the liver *via* AMPK.[52] Met is reported to down-regulate the expression of insulin and insulin-like growth factors (IGF-1) receptors and reduces phosphorylation of insulin receptors, including insulin receptor substrate 1 (IRS-1).[53,54] (4) Inflammation, particularly neuroinflammation, is thought to be a primary driving force in the progression of dementias and Met suppresses nuclear factor kappa B (NF-κB) signaling and pro-inflammatory cytokines in various cell types,[55] suggesting that Met could protect against neuroinflammation. In clinics, several mechanisms often exist at the same time.

Whether Met reduces the incidence of dementia in diabetic patients may be related to the duration

of Met exposure. In the only RCT that evaluated cognitive responses after Met exposure in diabetic patients, the short 36-week duration of treatment could possibly account for an apparent lack of protective effect. It is thus possible that a protective effect of Met on cognitive function might be more evident after long-term use (⩾6 years), as suggested by the data (0.27; 95% CI = 0.12–0.60) in the study.[25] Similar findings were found in two other studies.[23,28] These studies indicate that Met therapy may be most effective if started early but still beneficial if started after a cognitive decline (Table 4). It is most likely that Met's main effect is decreasing damage over time rather than directly acting on the brain as a nootropic. This could be confirmed in future studies by comparing the cognitive function of elder people taking Met with that of short-term abstainers.

Strengths and limitations

The current study has several significant advantages. First, this will be the first and largest systematic review and meta-analysis providing the latest evidence for the relationship between Met treatment and subsequent dementia risk. Second, database search strategies have been developed with no search date restriction so that we can retrieve as many relevant articles as possible, avoiding the influence of publication bias on the pooled findings and improving the reproducibility of the results. Third, almost all of the studies included were divided into two cohorts: (1) regional and population-based cohorts and (2) a countrywide and population-based inpatient registry (we excluded general, hospital-based quality samples, aiming at minimizing many other potential sources of bias). Furthermore, a transparent methodologic quality assessment of the included studies was checked using NOS listings recommended for cohort studies. Fourth, subgroup analyses, sensitivity tests, and meta-regression analyses were performed to explore the potential heterogeneity based on the abstracted study-level baseline characteristics. All our reported results remained constant under these sensitivity analyses, and Egger's test or 'Trim and Fill' analysis showed no evidence of publication bias.

However, these above results should be interpreted carefully, due to the small number of studies, which may not be sufficient to draw a reliable conclusion.[25,27,28] We restricted the study language to English, and the articles which have been published in other languages may have been missed in the three databases that we searched. Moreover, significant heterogeneity was found among the included studies, which is predictable and may be due to the differences in population baseline characteristics (age at T2DM diagnosis, gender, ethnicity, dementia type, etc.), exposed treatment (Met use and Met-free), study design (both prospective and retrospective), and statistical methods (adjustment for confounders), which was confirmed by univariate meta-regression analysis. In addition, because our study is a research-level meta-analysis rather than a single-patient-level meta-analysis, we are unable to perform a more detailed subgroup analysis (e.g. time-to-event risk analysis based on length of Met exposure and follow-up duration).

Future directions

Ultimately, notwithstanding its limitations, the current study includes all dementia types and both prospective and retrospective population-based studies, which provides a large enough sample size for a meaningful and robust statistical analysis. A future clinical investigation should focus on establishing risk assessment and individualized treatment strategies for diabetes-related dementia based on both molecular and macroscopic characteristics.

Conclusion

This systematic review and meta-analysis showed that long-term use of Met in T2DM might result in a decreased risk of dementia. This association remains stratified by most baseline variables and is biologically plausible. However, we should interpret the results cautiously until high-level evidence from prospective cohort studies proves this relationship.

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Author contributions

Shiliang Ji: Data curation; Formal analysis; Methodology; Writing – review & editing.

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Lifeng Huang: Conceptualization; Writing – original draft; Writing – review & editing.
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Conflict of interest statement

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Availability of data and materials

All data relevant to the study can be found in the article or supplementary material.

Supplemental material

Supplemental material for this article is available online.

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