

RESEARCH ARTICLE

Association of transketolase polymorphisms with diabetic polyneuropathy in the general population: The KORA F4 study

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Abstract

Aims: We recently reported that genetic variability in the *TKT* gene encoding transketolase, a key enzyme in the pentose phosphate pathway, is associated with measures of diabetic sensorimotor polyneuropathy (DSPN) in recent-onset diabetes. Here, we aimed to substantiate these findings in a population-based KORA F4 study.

Materials and Methods: In this cross-sectional study, we assessed seven single nucleotide polymorphisms (SNPs) in the transketolase gene in 952 participants from the KORA F4 study with normal glucose tolerance (NGT; $n = 394$), prediabetes ($n = 411$), and type 2 diabetes ($n = 147$). DSPN was defined by the examination part of the Michigan Neuropathy Screening Instrument (MNSI) using the original

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MNSI > 2 cut-off and two alternative versions extended by touch/pressure perception (TPP) (MNSI > 3) and by TPP plus cold perception (MNSI > 4).

Results: After adjustment for sex, age, BMI, and HbA1c, in type 2 diabetes participants, four out of seven transketolase SNPs were associated with DSPN for all three MNSI versions (all $p \leq 0.004$). The odds ratios of these associations increased with extending the MNSI score, for example, OR (95% CI) for SNP rs62255988 with MNSI > 2: 1.99 (1.16–3.41), MNSI > 3: 2.27 (1.26–4.09), and MNSI > 4: 4.78 (2.22–10.26); SNP rs9284890 with MNSI > 2: 2.43 (1.42–4.16), MNSI > 3: 3.46 (1.82–6.59), and MNSI > 4: 4.75 (2.15–10.51). In contrast, no associations were found between transketolase SNPs and the three MNSI versions in the NGT and prediabetes groups.

Conclusions: The link of genetic variation in transketolase enzyme to diabetic polyneuropathy corroborated at the population level strengthens the concept suggesting an important role of pathways metabolising glycolytic intermediates in the evolution of diabetic polyneuropathy.

KEYWORDS

diabetic polyneuropathy, genetic variability, pentose phosphate pathway, single nucleotide polymorphisms, transketolase

1 | INTRODUCTION

Diabetic sensorimotor polyneuropathy (DSPN) affects around one third of people with diabetes,¹ predicts major clinical sequels such as foot ulcers and amputations,² and contributes to significant morbidity due to neuropathic pain³, an increased risk of mortality, and reduced quality of life.^{4,5} Polyneuropathy is not a 'late' complication of diabetes as frequently quoted, but rather an early one showing an increased prevalence already in recent-onset diabetes and prediabetes.^{6,7} Since the risk of the development and progression of DSPN cannot be sufficiently predicted by hyperglycemia or other traditional risk factors on an individual basis, the search for genetically determined factors remains of major interest. As a consequence, it has been emphasised that genetic studies should select genes that encode proteins involved in the known mechanisms of nerve protection or damage in diabetes, identify single-nucleotide polymorphisms [single nucleotide polymorphisms (SNPs)], document that these gene polymorphisms affect the activity of the encoded proteins, and search for associations between gene polymorphisms and diabetic neuropathy.⁸ A number of studies have explored candidate genes potentially causal for DSPN,⁹ but the majority of these were not population-based and had a small sample size, thus weakening the ability to identify the contributory role of common alleles.

A single hyperglycemia-induced process of superoxide overproduction by the mitochondrial electron-transport chain has been identified to divert upstream metabolites from glycolysis into four major molecular pathways of diabetic microvascular complications.¹⁰ These metabolites, which are also end products of the

non-oxidative branch of the pentose phosphate pathway (PPP), are generated by the thiamine diphosphate-dependent enzyme transketolase that catalyses several key reactions of the non-oxidative branch of the PPP. Shunting of glycolytic intermediates into the PPP has been suggested to protect from hyperglycemia-induced microvascular damage.¹¹ Given that thiamine (vitamin B1) is an essential cofactor in glucose metabolism, we recently performed a meta-analysis showing that diabetes is associated with lower levels of various systemic thiamine derivatives, while erythrocyte transketolase activity tended to be lower in diabetes participants without reaching statistical significance.¹²

Studies that investigated SNPs related to thiamine and its metabolism in the context of diabetic microvascular complications are scarce.^{13–16} Only three previous studies assessed associations between genetic variability in transketolase (*TKT*) and diabetic microvascular complications.^{14–16} In the first study, among 10 tagging SNPs in the transketolase gene, no differences were found in allelic, genotype, or haplotype distributions between groups of diabetes patients with and without nephropathy.¹⁴ A prospective study from the same institution reported that genetic variability in the transketolase enzyme in individuals with diabetes was associated with progression of diabetic nephropathy and incidence of major cardiovascular events.¹⁵ We recently genotyped nine SNPs of the transketolase gene in participants with recent-onset diabetes from the German Diabetes Study (GDS) baseline cohort. Associations with neuropathic symptoms and reduced thermal detection thresholds were found for seven SNPs.¹⁶ The present study was conducted to substantiate these findings in the general population.

2 | MATERIALS AND METHODS

The studies were carried out in accordance with the Declaration of Helsinki, including written informed consent from all participants, and were approved by the ethics committee of the Bavarian Chamber of Physicians (Munich). The total study sample included 1160 participants aged 61–82 years from the KORA (Cooperative Health Research in the Region of Augsburg) F4 study (2006–2008), a follow-up examination of the population-based KORA S4 study (1999–2001) conducted in Augsburg (Germany) and two adjacent counties. Anthropometric and metabolic parameters, lifestyle factors, and glucose tolerance status using a standard 75 g oral glucose tolerance test (OGTT) were assessed as reported previously.¹⁷ Normal glucose tolerance (NGT) and prediabetes (impaired fasting glucose [IFG] and/or impaired glucose tolerance [IGT]) were classified according to the American Diabetes Association criteria using fasting glucose and 2-h glucose values.¹⁸ Among 1160 individuals, 112 were excluded due to type 1 diabetes, and unclear glucose tolerance status, 93 due to a missing Michigan Neuropathy Screening Instrument (MNSI), NeuroQuick, or monofilament data, and 3 were excluded due to missing demographic variables, resulting in a final sample size of 952 participants. (Supporting Information S1), 394 of whom had NGT, 411 had prediabetes, and 147 had type 2 diabetes (known type 2 diabetes: $n = 78$), type 2 diabetes newly detected by OGTT: $n = 69$).

2.1 | Single nucleotide polymorphisms (SNPs)

Genomic DNA was extracted from whole blood using the blood extraction kit (Qiagen) as previously described.¹⁶ In brief, selection of tagging SNPs was based on publicly available data of the International 1000 genomes Project derived from Utah residents with Central European ancestry <http://1000genomes.org>. The complete transketolase gene spanning 53258723–53290130 (chromosome assembly GRCh37.p13) on chromosome 3 was screened in silico. We found 76 SNPs with a minor allele frequency (MAF) ≥ 0.2 . Among these, 13 tagging SNPs were selected for genotyping using tagger software from haploview freeware (<https://www.broadinstitute.org/scientific-community/science/programs/medical-and-population-genetics/haploview/haploview>). The nine tagging SNPs for which genotyping was successful covered 73% of all common genetic variation in the locus (MAF ≥ 0.2) and were genotyped using the Sequenom massARRAY system with iPLEX software (Sequenom, Hamburg, Germany) as previously described¹⁹ with genotyping success rates of 99.7%. The Sequenom results were validated by bidirectional sequencing in 50 randomly selected persons with 100% identical results for both methods ($r = 1.00$). We previously reported associations of seven transketolase SNPs (rs7648309, rs62255988, rs7633966, rs9820979, rs11130362, rs9284890, rs12629312) with neuropathic symptoms and reduced thermal sensation (before Bonferroni correction) in participants with recent-onset diabetes from the GDS baseline cohort.¹⁶

The KORA Survey S4 was genotyped using the Affymetrix Axiom Chip covering genome wide ~700000 SNPs. The genome-wide association studies data have been imputed using the latest HRC reference panel granting estimated genotypes for all known SNPs to date. As all seven transketolase SNPs relevant for this project are imputed and thus are based on estimates, we additionally directly genotyped them with another technology to guarantee accurate data. Genotyping was done using the iPLEX assay from Agena in combination with Matrix-Assisted Laser Desorption/Ionization Time-Of-Flight Mass Spectrometry (MALDI-TOF MS), which is a solution for fast and accurate genotyping of SNPs in a high-throughput manner.²⁰ Alleles were distinguished through mass-modified nucleotides in a single base primer extension. Results of new genotyping and imputation were compared and robust genotypes were taken forward for statistical analysis.

2.2 | Neurological examination

Neurological examination to diagnose clinical DSPN was performed using the examination part of the MNSI as previously described, including items for the bilateral appearance of feet, presence of foot ulceration, ankle reflexes, and vibration perception threshold (VPT) using the Rydel-Seiffer C64 tuning fork at the great toes.²¹ Age-dependent limits of control persons were considered to define abnormal VPT.²² To define clinical DSPN using the original MNSI score ranging from 0 to 8 points, a cut-off >2 (MNSI > 2) was applied as previously suggested.²¹ Since the original MNSI does not include two important measures of sensory function predicting the development of foot ulcers, that is, testing of touch/pressure perception (TPP) and thermal perception,²³ these items were added to extend the original MNSI to obtain two alternative scores. Accordingly, the MNSI was extended by TPP using the 10 g monofilament (Neuropen) as previously reported,^{17,24} thus ranging from 0 to 10 points, and a cut-off > 3 (MNSI > 3) was chosen to define clinical DSPN as previously suggested by us.¹⁷ The third extended version included the TPP and cold perception using the NeuroQuick,²⁵ ranging from 0 to 12 points (MNSI > 4). These three definitions of DSPN satisfy the minimal diagnostic criteria for possible DSPN according to the Toronto Diabetic Neuropathy Expert Group.²⁶ Other potential causes of peripheral neuropathy, such as HIV infection and heavy alcohol consumption, were excluded, whereas data on hypothyroidism, vitamin deficiencies, or chronic inflammatory demyelinating polyneuropathy were not available.

Measurement of cold perception was added not only because of its predictive value for foot ulcers,²³ but also because in our previous study we found associations between transketolase SNPs and abnormal thermal detection thresholds in individuals recently diagnosed with diabetes.¹⁶ Cold perception threshold was assessed on the dorsum of the feet using the NeuroQuick (Schweers) as previously described.²⁵ In brief, this instrument for quantitative bedside testing of cold thermal perception is based on the wind chill factor, that is, the effect that wind has on the perception of cold. The

handheld microprocessor-operated electronic device comprises a fan adjustable to rotate at 10 different velocities (levels), while a constant distance to the skin (23 cm) is ensured by laser diodes. The flash ROM embedded application controls a fan (Papst, Landshut, Germany) at revolutions from 1300 to 4600 rpm blowing the accelerated air to the spot at 10 different levels. The maximum perceivable level is 10. If the stimulus was not perceived at this level, the threshold was set at level 11. The NeuroQuick threshold was defined as the mean of three readings.

2.3 | Statistical analysis

Categorical data were expressed as percentages of participants, while continuous data were expressed as medians (1st, 3rd quartile). Categorical variables were compared using the χ^2 test or Fisher's exact test where appropriate. Continuous data were assessed using the non-parametric Mann-Whitney *U* test. Associations between variables were assessed using multiple linear and logistic regression analyses with adjustments for sex, age, BMI, and HbA1c. All statistical tests were performed two sided. The level of significance was set at $\alpha = 0.05$. All analyses were performed using SPSS v.22.0 software.

3 | RESULTS

The demographic and clinical characteristics of the study participants by glucose status are listed in Table 1. The groups with prediabetes and type 2 diabetes showed higher proportions of men than the NGT group (both $p < 0.05$). Heart rate was higher in the group with type 2 diabetes compared with participants with NGT ($p < 0.05$). Compared to participants with NGT and prediabetes, the group with type 2 diabetes was older and had higher BMI, systolic blood pressure, triglycerides, HbA1c, fasting glucose, 2 h glucose and insulin in OGTT, CRP, cystatin C, and a higher proportion of antihypertensive agents as well as lower total cholesterol, high-density lipoprotein, HDL cholesterol, and low-density lipoprotein cholesterol (all $p < 0.05$). Compared to NGT participants, those with prediabetes were older and had higher BMI, heart rate, systolic and diastolic blood pressure, triglycerides, HbA1c, fasting glucose, 2 h glucose and insulin in OGTT, CRP, and a higher proportion of antihypertensive agents as well as lower HDL cholesterol levels (all $p < 0.05$). Compared to prediabetes participants, those with type 2 diabetes had higher serum creatinine levels ($p < 0.05$). No differences between the groups were found for height, current smoking status, fasting insulin, and the use of lipid lowering drugs.

Table 2 shows the prevalence of polyneuropathy defined by three different MNSI scores and cut-offs in the three groups studied. In general, the prevalence of polyneuropathy decreased with increasing cut-points in all three groups. Participants with type 2 diabetes showed a higher prevalence of polyneuropathy than those with NGT and prediabetes for all three MNSI cut-offs (all $p < 0.05$).

No differences in the prevalence of polyneuropathy were found between participants with prediabetes and those with NGT.

The associations of transketolase SNPs with the three MNSI scores as continuous variables computed by multiple linear regression analyses with adjustments for sex, age, BMI, and HbA1c in the group with type 2 diabetes are given in Table 3. All three MNSI scores were associated with four transketolase SNPs (rs62255988, rs9284890, rs9820979, rs7648309) (all $p \leq 0.004$), whereas no associations were found for the remaining three SNPs (rs7633966, rs11130362, rs12629312). No correlations between transketolase SNPs and the three MNSI versions were found in the entire study population as well as in the NGT and prediabetes subgroups (Supporting Information S2).

The results of the logistic regression analysis including adjustments for sex, age, BMI, and HbA1c between the transketolase SNPs and the three MNSI scores as categorical variables in participants with type 2 diabetes are presented in Figure 1. DSPN defined by all three MNSI cut-offs was associated with four transketolase SNPs (rs62255988, rs9284890, rs9820979, rs7648309), whereas no associations with DSPN were found for the remaining three SNPs (rs7633966, rs11130362, rs12629312).

4 | DISCUSSION

We recently genotyped nine SNPs of the transketolase gene in 165 type 1 and 373 type 2 diabetes individuals with a diabetes duration up to 1 year from the GDS baseline cohort. Seven of these SNPs were associated with measures of DSPN prior to Bonferroni correction; albeit thereafter, only the associations of transketolase SNP rs7648309 with neuropathic symptoms and rs62255988 with warm perception threshold remained statistically significant.¹⁶ In the present study, these findings were corroborated in the general population by demonstrating that four transketolase SNPs (including rs7648309 and rs62255988) were associated with DSPN defined by the original MNSI and two extended versions of the score with different cut-offs. In contrast, no associations were found between transketolase SNPs and the three MNSI versions in the NGT and prediabetes groups. Collectively, these data suggest that the risk of DSPN could be linked to activation of pathways metabolising glycolytic intermediates and their consecutive shunting into the PPP, which is selectively triggered by diabetes.

Apart from our recent study,¹⁶ genetic variability in transketolase enzyme has not been previously explored in individuals with DSPN, while two reports from a single institution focused on diabetic nephropathy. In the first report, among 10 tagging SNPs in the transketolase gene, neither allelic nor genotype or haplotype distributions were associated with prevalent nephropathy in participants with diabetes. Furthermore, there were no associations between genotypes of these 10 transketolase SNPs and erythrocyte transketolase activity.¹⁴ The second study found that progression of nephropathy in participants with type 2 diabetes was predicted by a combination of transketolase SNP rs11130362 and fructosamine 3-

TABLE 1 Demographic and clinical characteristics of the KORA F4 study participants by glucose metabolism status.

Variable	NGT	Prediabetes	Type 2 diabetes
n (% male)	394 (43)	411 (55) ^a	147 (62) ^a
Age (years)	69 (65, 74)	70 (66, 74) ^a	71 (67, 76) ^{a,b}
Height (cm)	165 (158, 172)	166 (159, 173)	166 (160, 172)
BMI (kg/m ²)	26.9 (24.7, 29.4)	28.5 (26.1, 31.4) ^a	30.3 (27.5, 33.7) ^{a,b}
Current smoking status (% yes)	9.9	5.6	6.8
Heart rate (bpm) ^e	70.0 (63.0, 77.0)	71.0 (64.0, 79.0) ^c	72.0 (64.0, 81.0) ^c
Systolic blood pressure (mmHg) ^e	125 (112, 135)	130 (118, 141) ^c	136 (121, 146) ^{c,d}
Diastolic blood pressure (mmHg) ^e	73.0 (67.0, 80.0)	75.0 (68.5, 81.5) ^c	74.0 (69.0, 80.0)
Triglycerides (mmol/L) ^e	1.14 (0.85, 1.55)	1.36 (1.02, 1.91) ^c	1.74 (1.15, 2.47) ^{c,d}
Cholesterol (mmol/L)	5.78 (5.14, 6.49)	5.74 (5.04, 6.43)	5.50 (4.78, 6.15) ^{c,d}
HDL cholesterol (mmol/L) ^e	1.50 (1.29, 1.76)	1.37 (1.14, 1.60) ^c	1.24 (1.06, 1.42) ^{c,d}
LDL cholesterol (mmol/L)	3.63 (3.07, 4.35)	3.64 (2.95, 4.24)	3.31 (2.82, 3.98) ^{c,d}
HbA1c (%) ^e	5.5 (5.3, 5.7)	5.6 (5.4, 5.9) ^c	6.6 (6.1, 7.3) ^{c,d}
HbA1c (mol/mmol) ^e	37.0 (34.0, 39.0)	38.0 (36.0, 41.0) ^c	49.0 (43.0, 56.0) ^{c,d}
Fasting glucose (mmol/L) ^e	5.11 (4.87, 5.27)	5.72 (5.55, 6.05) ^c	7.60 (6.66, 8.82) ^{c,d}
2h glucose in OGTT (mmol/L) ^e	5.7 (5.0, 6.6)	7.8 (6.3, 8.8) ^c	11.9 (11.3, 13.1) ^{c,d}
Fasting insulin (pmol/L) ^e	4.20 (2.85, 6.40)	5.55 (3.90, 9.60)	7.70 (4.60, 15.00)
2h insulin in OGTT (pmol/L) ^e	38 (20, 60)	67 (40, 101) ^c	116 (73, 208) ^{c,d}
Creatinine (nmol/L)	79 (69, 91)	82 (72, 95)	87 (77, 103) ^d
C-reactive protein (mg/L)	1.33 (0.68, 2.42)	1.73 (0.87, 3.41) ^c	2.42 (1.15, 4.91) ^{c,d}
Cystatin C (mg/L)	0.80 (0.73, 0.89)	0.81 (0.72, 0.91)	0.87 (0.76, 1.00) ^d
Diabetes duration (years) ^f	-	-	9.0 (4.8, 14.0)
Insulin treatment (%)	0	0	11
Glucose lowering drugs (%)	0	0	43
Antihypertensive drugs (%)	45	59 ^c	77 ^{c,d}
Lipid lowering drugs (%)	23	22	30

Note: Data are % or median (1st, 3rd quartile).

Abbreviations: HDL, high-density lipoprotein; LDL, low-density lipoprotein; OGTT, oral glucose tolerance test.

^a*p* < 0.05 versus NGT group.

^b*p* < 0.05 versus prediabetes group (nonparametric Mann-Whitney *U* test); after adjustment (linear and logistic regression analyses).

^c*p* < 0.05 versus NGT group.

^d*p* < 0.05 versus prediabetes group.

^eVariables adjusted for sex, age, BMI, and current smoking status.

^fDiabetes duration is shown only for participants with known type 2 diabetes (*n* = 78), the newly detected cases (*n* = 69) were not included.

kinase (FN3K) SNP rs1056534. In addition, transketolase SNP rs3736156 alone and also in combination with the aforementioned two SNPs predicted the incidence of major cardiovascular events.¹⁵ The only other genetic study in the context of thiamine metabolism focused on variants in specific thiamine transporters in individuals with type 1 diabetes.¹³ Thiamine is carried into the cells by two high-affinity thiamine transporters, hTHTR1 and hTHTR2, and by a low-affinity transporter. Two transcription factors (SP1, SP2) are known

to affect the expression of SLC19A2 and SLC19A3 genes encoding hTHTR1 and hTHTR2. In the FinnDiane cohort, 134 SNPs in SLC19A2 and SLC19A3 as well as SP1 and SP2 were examined for an association with severe retinopathy or nephropathy. Two SNPs were found in strong linkage disequilibrium in the SLC19A3 locus associated with a lower rate of severe retinopathy and severe retinopathy and end-stage renal disease combined. The results for severe retinopathy could not be replicated in the DCCT/EDIC and WESDR

	NGT (n = 394)	Prediabetes (n = 411)	Type 2 diabetes (n = 147)
MNSI > 2 (%)	23.1	26.3	37.4 ^{a,b}
MNSI > 3 (%)	18.6	17.9	26.7 ^{a,b}
MNSI > 4 (%)	9.9	7.6	17.1 ^{a,b}

TABLE 2 Prevalence of polyneuropathy defined by three different MNSI scores and cut-offs.

Abbreviations: MNSI, Michigan Neuropathy Screening Instrument; MNSI > 2, original MNSI > 2 points; MNSI > 3, MNSI extended by touch/pressure perception (TPP) > 3 points; MNSI > 4, MNSI extended by TPP and cold perception > 4.

^a $p < 0.05$ versus NGT.

^b $p < 0.05$ versus prediabetes (χ^2 test).

TABLE 3 Association of transketolase SNPs with three MNSI scores in the group with type 2 diabetes (n = 147).

SNP	EA	NEA	EAF	MNSI		MNSI + TPP		MNSI + TPP + CP	
				β	p Value	β	p Value	β	p Value
rs62255988	A	G	0.16	0.264	0.001	0.264	0.001	0.259	0.001
rs9284890	C	T	0.27	0.335	0.00002	0.343	0.00001	0.339	0.00002
rs9820979	G	A	0.63	0.230	0.004	0.229	0.004	0.231	0.004
rs7648309	A	G	0.16	0.253	0.001	0.257	0.001	0.257	0.001

Note: Multiple linear regression analysis: adjusted for sex, age, BMI, and HbA1c.

Abbreviations: EA, effect allele; EAF, effect allele frequency; MNSI, Michigan Neuropathy Screening Instrument; MNSI + TPP, MNSI extended by touch/pressure perception; MNSI + TPP + CP, MNSI extended by touch/pressure perception and cold perception; NEA, non-effect allele.

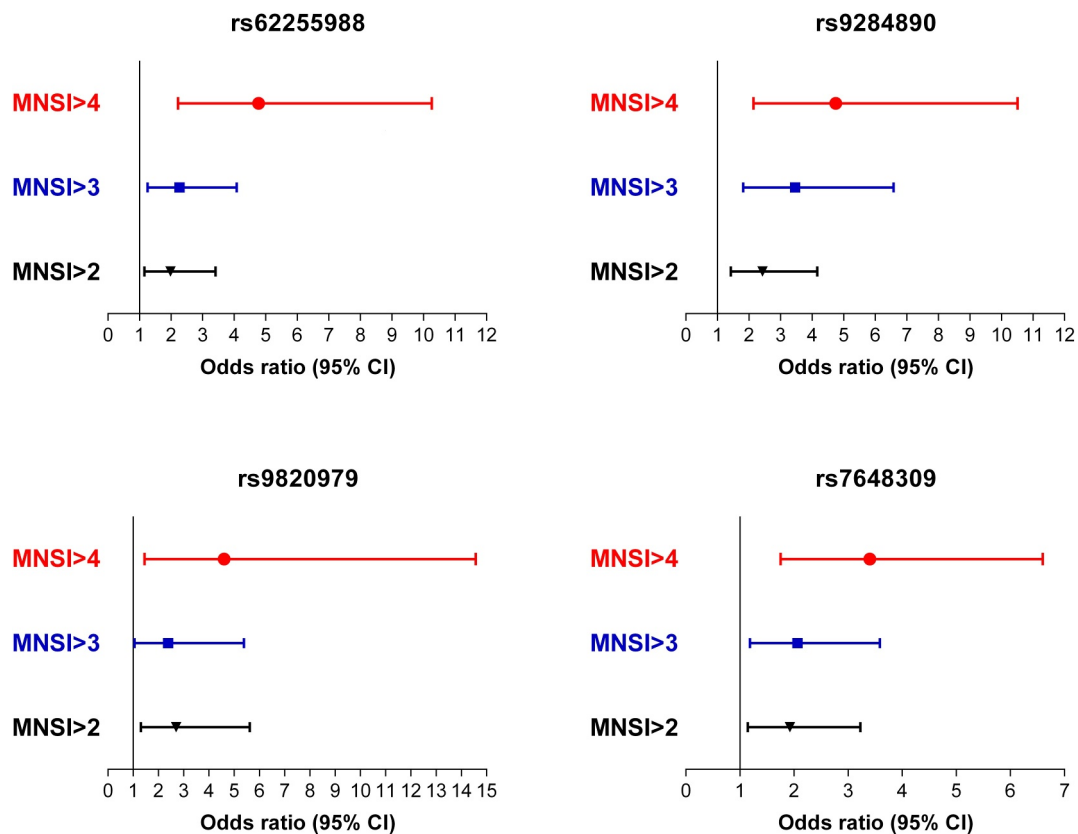


FIGURE 1 Association of transketolase SNPs with polyneuropathy defined by three different MNSI scores in the group with type 2 diabetes (n = 147). The odds ratios were calculated using logistic regression analyses adjusted for sex, age, BMI, and HbA1c. The cut-offs for the different MNSI score definitions were: three points for MNSI (MNSI > 2), four points for MNSI extended by touch/pressure perception (TPP) (MNSI > 3, blue), and five points for MNSI extended by TPP and cold perception (MNSI > 4, red).

cohorts. However, the association for the combined phenotype reached genome-wide significance for one SNP in a meta-analysis including the FinnDiane and WESDR cohorts. These findings suggest that genetic variability in genes encoding thiamine transporters may protect from developing microvascular damage in type 1 diabetes.¹³

Mitochondrial overproduction of superoxide has been suggested as the initial event causing diabetic microvascular complications by activation of four major pathways comprising the polyol pathway, advanced glycation end-product formation, protein kinase C isoforms, and hexosamine pathway.^{10,11} This activation can be limited by transketolase, which itself is activated by its cofactor thiamine, by reducing the availability of the glycolytic intermediates via the PPP, which fulfils three important functions: first, supporting intracellular antioxidant defence by production of reducing equivalent NADPH necessary for reduction of oxidised glutathione; second, maintaining the synthesis of nucleotides by supplying ribose-5-phosphate; and third, metabolising dietary pentoses.^{27–29} Benfotiamine, a lipid-soluble allithiamine homologue, has been shown to prevent experimental diabetic retinopathy by blocking the aforementioned major pathways leading to hyperglycemia-induced microvascular damage by reducing the accumulation of triosephosphates and fructose-6-phosphate via the PPP.^{11,29,30} In the BENDIP study, treatment with benfotiamine has been shown to improve neuropathic symptoms after 6 weeks in patients with DSPN,³¹ but long-term studies are needed to demonstrate benefits on nerve dysfunction. In a 2-year study, treatment with benfotiamine (300 mg/day) did not exert an effect on peripheral nerve function superior to placebo in patients with type 1 diabetes.³² However, since the majority of the participants did not have DSPN, the design and clinical relevance of this trial have been questioned.³³ To fill this gap, the BOND study assessing the effects of 1-year treatment with benfotiamine on morphometric, neurophysiological, and clinical measures in individuals with type 2 diabetes and mild to moderate symptomatic DSPN is currently underway.³⁴

The present study has several limitations. First, we did not explore a possible genotype-phenotype interaction between thiamine metabolism and transketolase SNPs in relation to DSPN. Second, in contrast to our previous study in recent-onset diabetes, which used sophisticated quantitative measures of nerve function such as nerve conduction studies and quantitative sensory testing of warm/cold perception thresholds among others,¹⁶ the present large population-based study allowed for using only measures to clinically diagnose DSPN such as the MNSI examination part. To further harmonise the diagnostic procedures between the two studies, measurement of the cold perception threshold using the NeuroQuick device was added to the original MNSI score. Third, the cross-sectional study setting does not allow the determination of the predictive value of transketolase SNPs on the development and progression of DSPN. The strengths of this work are the relatively large sample, population-based setting and comprehensive metabolic phenotyping.

In conclusion, the results of this population-based study corroborated our recently reported associations between transketolase SNPs and measures of DSPN, albeit the two cohorts differed in their demographic aspects and methods of assessing DSPN. While the prior study included participants with type 1 and type 2 diabetes with a diabetes duration up to 1 year and used quantitative neurophysiological techniques, the present study focused on the elderly population with longer-term diabetes and solely clinical DSPN assessment. Nonetheless, the fact that the main results could be substantiated despite these differences even renders our results more generalisable. In the future, pharmacogenomics considering SNPs in the transketolase gene could be useful to optimise specific treatments targeting the corresponding enzymes in the context of precision medicine and ultimately culminate in an improved drug response.

AUTHOR CONTRIBUTIONS

Dan Ziegler designed the study and wrote the manuscript. Dan Ziegler, Haifa Maalmi, Alexander Strom, Gidon Bönhof, Birgit Knebel, Erwin Schleicher, Wolfgang Rathmann, Christian Herder, Haifa Maalmi, Christian Gieger, and Margit Heier researched data; Barbara Thorand, Alexander Strom, Gidon Bönhof, Birgit Knebel, Erwin Schleicher, Wolfgang Rathmann, Christian Herder, Haifa Maalmi, Christian Gieger, Margit Heier, Christine Meisinger, Michael Roden, Annette Peters and Harald Grallert revised the manuscript. All authors contributed substantially to the conception or design of the work; or the acquisition, analysis, or interpretation of data for the work; and to drafting the work or revising it critically for important intellectual content; gave final approval of the version to be published; and agreement to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. **Dan Ziegler** is the guarantor of this work and, as such, had full access to all of 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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CONFLICT OF INTEREST STATEMENT

DZ received honoraria for speaking and consulting activities from Wörwag Pharma. No dualities of interest were reported by the other authors.

ETHICS STATEMENT

The studies were carried out in accordance with the Declaration of Helsinki, including written informed consent from all participants, and were approved by the ethics committee of the Bavarian Chamber of Physicians (Munich, Germany).

DATA AVAILABILITY STATEMENT

KORA data and biosamples are available upon request by means of a project agreement subject to approval by the KORA Board.

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PEER REVIEW

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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