General and Abdominal Obesity and Incident Distal Sensorimotor Polyneuropathy: Insights Into Inflammatory Biomarkers as Potential Mediators in the KORA F4/FF4 Cohort

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OBJECTIVE

To investigate the associations between different anthropometric measurements and development of distal sensorimotor polyneuropathy (DSPN) considering interaction effects with prediabetes/diabetes and to evaluate subclinical inflammation as a potential mediator.

RESEARCH DESIGN AND METHODS

This study was conducted among 513 participants from the Cooperative Health Research in the Region of Augsburg (KORA) F4/FF4 cohort (aged 62–81 years). Anthropometry was measured at baseline. Incident DSPN was defined by neuropathic impairments using the Michigan Neuropathy Screening Instrument at baseline and follow-up. Associations between anthropometric measurements and DSPN were estimated by multivariable logistic regression. Potential differences by diabetes status were assessed using interaction terms. Mediation analysis was conducted to determine the mediation effect of subclinical inflammation in these associations.

RESULTS

After a mean follow-up of 6.5 years, 127 cases with incident DSPN were detected. Both general and abdominal obesity were associated with development of DSPN. The odds ratios (95% CI) of DSPN were 3.06 (1.57; 5.97) for overweight, 3.47 (1.72; 7.00) for obesity (reference: normal BMI), and 1.22 (1.07; 1.38) for 5-cm differences in waist circumference, respectively. Interaction analyses did not indicate any differences by diabetes status. Two chemokines (C-C motif chemokine ligand 7 [CCL7] and C-X-C motif chemokine ligand 10 [CXCL10]) and one neuron-specific marker (Delta/Notch-like epidermal growth factor–related receptor [DNER]) were identified as potential mediators, which explained a proportion of the total effect up to 11% per biomarker.

CONCLUSIONS

General and abdominal obesity were associated with incident DSPN among individuals with and without diabetes, and this association was partly mediated by inflammatory markers. However, further mechanisms and biomarkers should be investigated as additional mediators to explain the remainder of this association.

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Distal sensorimotor polyneuropathy (DSPN) is associated with several adverse health outcomes, including increased morbidity such as foot ulcers and lower-extremity amputation, occurrence of chronic pain, depression, impaired quality of life, and risk of premature death (1,2). The prevalence and incidence vary according to assessment method of DSPN, prediabetes/diabetes status, and diabetes duration. The prevalence of DSPN has been estimated to be ~30% for hospital-based populations, 20% for the general population, and 10–15% for newly diagnosed patients with type 2 diabetes, whereas with longer duration of diabetes (>10 years), the prevalence increases up to 50% (3,4). Evidence has emerged suggesting that the prevalence of DSPN is increased even in individuals with prediabetes compared to those with normal glucose tolerance (1). Major risk factors of DSPN include advanced age, smoking, and metabolic factors, such as diabetes, hyperglycemia, and obesity (5).

Previous studies that reported associations between obesity, measured by BMI or waist circumference, and DSPN were mostly cross-sectional (2–6). Only one recent prospective study reported a positive association between obesity and increased risk of diabetic polyneuropathy (7,8). Further data on this relationship are lacking, and the previous study focused only on DSPN in patients with type 2 diabetes. It remains unclear whether diabetes or prediabetes, defined as impaired fasting glucose (IFG) and/or impaired glucose tolerance (IGT), are effect modifiers of the relationship between obesity and DSPN. Moreover, the underlying mechanisms explaining this association are not well understood. A possible mediator for this relationship might be a low-grade inflammatory state. In two recent reports, two proinflammatory cytokines (interleukin-6 [IL-6] and tumor necrosis factor-α [TNF-α]), three chemokines (C-C motif chemokine ligand 7 [CCL7], C-X-C motif chemokine ligand 9 [CXCL9], and CXCL10), as well as three soluble forms of inflammation-related membrane proteins (Delta/Notch-like epidermal growth factor–related receptor [DNER], CD40L receptor [CD40], and TNF receptor superfamily member 9 [TNFRSF9]) have been identified as independent risk markers for incident DSPN in the Cooperative Health Research in the Region of Augsburg (KORA) F4/FF4 cohort (9,10). It remains to be answered whether and to what extent these biomarkers may explain the link between obesity and DSPN.

The aims of this study were threefold. First, we investigated the association between general and abdominal obesity and incident DSPN. Second, potential differences in these associations between individuals with and without prediabetes/diabetes were assessed. Third, inflammatory biomarkers were considered as potential mediators of the observed associations.

RESEARCH DESIGN AND METHODS

Study Design and Participants

The study design has been described previously in detail (9,11). Briefly, this prospective cohort study is based on the KORA F4 (2006–2008) and KORA FF4 studies (2013 to 2014), which are both follow-up examinations of the KORA S4 study (1999–2001), a population-based cohort study conducted in the region of Augsburg, Germany.

The study sample is identical to the sample used for a comprehensive analysis of associations between biomarkers of inflammation and incident DSPN (10). Briefly, out of the 1,161 KORA F4 study participants aged 62–81 years, we excluded participants with missing information of exposure variables and covariates (n = 113), missing follow-up information (n = 452), and prevalent DSPN (n = 83), resulting in an analysis sample of 513 participants.

Characteristics of the cohort, including age, sex, socioeconomic status, anthropometry, metabolic variables, lifestyle factors, and glucose tolerance status using standard 75-g oral glucose tolerance tests were assessed as previously reported. Prediabetes was defined as the presence of IFG and/or IGT (12,13). DSPN was assessed at baseline and follow-up examinations. All examinations were carried out in accordance with the Declaration of Helsinki, including written informed consent from all participants. The study was approved by the ethics board of the Bavarian Chamber of Physicians (Munich, Germany).

Assessment of DSPN

DSPN was assessed using the Michigan Neuropathy Screening Instrument (MNSI) as described (9). Briefly, the clinical examination part of the MNSI comprised the following items: appearance of feet, foot ulceration, ankle reflexes, and vibration perception threshold using the C 64 Hz Rydel-Seiffer tuning fork at the great toes. Age-dependent lower limits of normal for vibration perception threshold at the fifth percentile were computed according to Martina et al. (14) using the equation $y = 5.75 - 0.026 \times \text{age}$. The neuropathy assessment also included a bilateral examination of sensory perception with a 10-g monofilament (Neuropen) (9). Thus, the total MNSI score ranged from 0 (all aspects normal) to a maximum of 10 points. Prevalent or incident DSPN were defined based on a cutoff value of >3 points as described in our previous study (9).

Assessment of Anthropometry

Body weight, height, and waist and hip circumference were measured by trained personnel based on standard protocols (12). Body weight was assessed in light clothing to the nearest 0.1 kg. BMI (kg/m^2) was calculated as a measure of general body fatness, and individuals with a BMI of 18.5 to <25 kg/m^2 were defined as normal weight, 25 to <30 kg/m^2 as overweight, and ≥30 kg/m^2 as obese (15). Waist circumference was measured at the minimum abdominal girth, and hip circumference was assessed at the maximum protrusion of the hips at the level of the symphysis pubis to the nearest 0.1 cm (16). Abdominal obesity was defined by the waist circumference according to the classification of the International Diabetes Federation (IDF) with sex-specific cutoff points (<94 vs. ≥94 cm for men and <80 vs. ≥80 cm for women) (17) and according to the classification of the World Health Organization (WHO) (<102 vs. ≥102 cm for men and <88 vs. ≥88 cm for women) (15). Further measures for abdominal obesity were defined by the waist-to-hip ratio (WHR), calculated as waist circumference divided by hip circumference (cutoffs <0.95 vs. ≥0.95 for men and <0.80 vs. ≥0.80 for women (15), and the waist-to-height ratio (WHtR), calculated as waist circumference divided by height.

Measurement of Biomarkers of Subclinical Inflammation

Fasting serum levels of IL-6 and TNF-α were measured by ELISA as described previously (9,18). Fasting serum levels of CCL7, CXCL9, CXCL10, DNER, CD40, and

TNFRSF9) were measured using the OLINK Inflammation multiplex immunoassay (OLINK Proteomics, Uppsala, Sweden) with intra-assay coefficients of variation ranging from 2.3% to 6.5% and interassay coefficients of variation ranging from 5.4% to 10.3% as described (10). The assay uses proximity extension assay technology, which combines a detection step using oligonucleotide-labeled antibodies, a proximity-dependent DNA polymerization event, and a real-time quantitative PCR amplification. Relative protein values are given as normalized protein expression (NPX) values (19,20), which have similar distributions as $\log_2$-transformed protein concentrations. Using this technology, novel associations between biomarkers and cardiometabolic risk were identified in multiple cohorts (19–23).

Statistical Analysis

Descriptive statistics are reported for the participants stratified by incident DSPN shown as mean ± SD, median (interquartile range), or percentages.

The associations between anthropometry (BMI, waist circumference, WHR, and WHtR) and incident DSPN were analyzed by calculating odds ratios (ORs) with corresponding 95% CIs in logistic regression models. The exact date of the manifestation of DSPN was not known, and thus, we conducted logistic regression analysis rather than survival analysis. Anthropometric measures were investigated as predefined categories as described above and as continuous measures (per 5 kg/m^2 for BMI, per 5 cm for waist circumference, and per 0.1 units for WHR and WHtR).

We fitted three different regression models. The first model was adjusted for age and sex and the second (our main model) for age, sex, socioeconomic status (including information on education, income, and occupation, combined as a score [range: 1–27 points] and classified as low [1–9 points], moderate low [10–12 points], moderate [13–15 points], moderate high [16–19 points], or high [>19 points]), smoking status (never, former, or current), alcohol intake (abstainer [0 g/day], moderate [>0 to <40 g/day for men and >0 to <20 g/day for women], and high [≥40 g/day for men and ≥20 g/day for women]), physical activity (inactive [<1 h of sports/week] or active [≥1 h sports/week]), and height (not in models for BMI and WHtR). In addition, we investigated in a third model whether cardiometabolic factors, which might lie on the pathway from obesity to incident DSPN, influence the association under investigation. Model 3 was based on model 2 with additional adjustment for cardiometabolic factors, including HbA_{1c}, hypertension (defined as blood pressure ≥140/90 mmHg or use of antihypertensive medication), HDL cholesterol, LDL cholesterol, and triglycerides.

The main analysis (model 2) was performed in the total cohort and stratified by prediabetes/diabetes status (prediabetes and diabetes vs. normal glucose tolerance). Prediabetes and diabetes were combined to increase statistical power in these analyses. Potential differences between persons with and without prediabetes/diabetes were assessed by including an interaction term between anthropometric measurement and prediabetes/diabetes status in the logistic regression models. We investigated potential nonlinear associations between anthropometric measurements and incident DSPN by using restricted cubic spline regression models, with three equally spaced knots between the 0.05 and 0.95 quantiles.

In addition, we investigated whether biomarkers of subclinical inflammation were mediators of the identified association between general and abdominal obesity and DSPN. In total, eight biomarkers of subclinical inflammation (IL-6, TNF-α, CCL7, CXCL9, CXCL10, DNER, CD40, and TNFRSF9) showing an association with DSPN (which is one criterion that needs to be fulfilled for the mediation analysis [24]) were selected based on previous reports (9,10). Another criterion that needs to be satisfied is the relation between the exposure (anthropometric measures) and the mediator (biomarkers of subclinical inflammation). BMI was used to assess general obesity and waist circumference for abdominal obesity. Thus, we checked if the potential mediators were associated with BMI or waist circumference, respectively. Finally, each potential mediator was added to the main model (model 2) to estimate the proportion mediated as indirect natural effect divided by the sum of the direct and indirect natural effect by applying causal mediation analysis (25). If the 95% CI of the percent mediated effect did not include the null value, the mediation effect was interpreted as statistically significant.

The statistical analyses were conducted with R version 3.3.3 (www.R-project.org/). Restricted cubic spline regression models were analyzed using the "rms" package in R. We used the R package "mediation" performing a quasi-Bayesian approximation with 1,000 Monte Carlo draws for CI estimation. To test for exposure–mediator interaction, we used the test.TMint function (26). The *P* values presented were two tailed, and $P < 0.05$ was considered statistically significant.

RESULTS

Characteristics

Out of 513 participants, 127 developed DSPN during a mean follow-up time of 6.5 (SD ± 0.2) years. Table 1 shows the baseline characteristics of study participants with and without DSPN at follow-up as partly reported before (10). Participants with incident DSPN were older, had higher BMI values, were more often overweight and obese, had a higher waist circumference, WHR, and HbA_{1c}, were more often smokers, and were physically less active compared with participants without DSPN. Differences were also observed for biomarkers of subclinical inflammation with higher levels of IL-6, CCL7, CXCL9, CXCL10, CD40, and TNFRSF9 in individuals with incident DSPN compared with individuals without DSPN.

Association Between Anthropometric Measures and DSPN

General obesity defined by the BMI was positively associated with incident DSPN (Table 2). The multivariable adjusted OR (95% CI) for DSPN was 3.06 (1.57; 5.97) for overweight and 3.47 (1.72; 7.00) for obesity (Table 2, model 2) compared with normal weight. The multivariable adjusted OR (95% CI) of DSPN was 1.68 (1.28; 2.22) for an increment of 5 kg/m^2 of BMI. When additionally adjusted for metabolic factors (HbA_{1c}, hypertension, HDL cholesterol, LDL cholesterol, and triglycerides), there was no substantial change in the OR (Table 2, model 3). There was no indication for a nonlinear association between BMI and DSPN (Supplementary Fig. 1*A*) (*P* for nonlinearity = 0.163).

For abdominal obesity defined as waist circumference, the multivariable

Table 1—Baseline characteristics of the study population in KORA F4 stratified by incidence of DSPN*

Variable	No incident DSPN	Incident DSPN	*P*
N	386	127	
Age (years)	67.9 ± 4.6	70.2 ± 5.0	<0.001
Men (%)	49.2	55.9	0.169
Socioeconomic status (%)†			0.139
Low	23.6	27.0	
Moderate low	21.2	19.8	
Moderate	21.8	27.8	
Moderate high	19.2	11.1	
High	14.2	14.3	
Height (cm)	166 ± 9	167 ± 9	0.016
BMI (kg/m^2)	27.6 ± 3.9	29.2 ± 4	<0.001
BMI classes (%)			0.001
Normal weight (18.5 to <25 kg/m^2)	27.5	11.0	
Overweight (25 to <30 kg/m^2)	46.1	52.8	
Obese (≥30 kg/m^2)	26.4	36.2	
Waist circumference (cm)	94.7 ± 11.3	100.1 ± 11.4	<0.001
Waist circumference classes (%)			<0.001
WHO definition			
Low (M: <102, W: <88 cm)	53.1	34.6	
High (M: ≥102, W: ≥88 cm)	46.9	65.4	
IDF definition			0.001
Low (M: <94, W: <80 cm)	24.1	11.0	
High (M: ≥94, W: ≥80 cm)	75.9	89.0	
WHR	0.90 ± 0.08	0.92 ± 0.08	0.004
WHR classes (%)			0.071
Low (M: <0.95, W: <0.8)	35.8	27.6	
High (M: ≥0.95, W: ≥0.8)	64.2	72.4	
HbA_{1c} (%)	5.66 ± 0.48	5.83 ± 0.69	0.014
HbA_{1c} (mmol/mol)	39 ± 6	40 ± 7	0.014
Diabetes status (%)			0.299
No diabetes	45.9	38.6	
Prediabetes§	40.2	40.9	
Diabetes	14.0	20.5	
Hypertension (%)‡	56.5	65.4	0.452
Total cholesterol mmol/L (mmol/L)¶	6.06 ± 1.01	5.82 ± 1.12	0.094
Fasting triglycerides (mmol/L)¶	1.27 (0.97; 1.77)	1.29 (1.05; 1.67)	0.745
Use of lipid-lowering drugs (%)	23.1	26.8	0.643
eGFR (mL/min/1.73 m^2)	80.2 ± 13.1	76.9 ± 14.3	0.498
Smoking status (%)			0.002
Never	51.6	54.3	
Former	42.7	33.9	
Current	5.7	11.8	
Alcohol intake (%)#			0.584
Abstainer	31.1	30.7	
Moderate intake	52.3	51.2	
High intake	16.6	18.1	
Physically active (%)‖	62.2	41.7	<0.001
Myocardial infarction (%)	5.2	7.1	0.850
Neurological conditions that might cause nerve damage (%)	15.1	20.5	0.084
Use of nonsteroidal anti-inflammatory drugs (%)††	1.0	2.4	0.375
IL-6 (pg/mL)	1.33 (0.94; 1.96)	1.71 (1.22; 2.52)	0.005
TNF-α (pg/mL)	1.90 (1.39; 2.73)	2.09 (1.60; 3.43)	0.062
CCL7 (NPX)	1.83 ± 0.50	2.09 ± 0.64	<0.001
CXCL9 (NPX)	7.27 ± 0.75	7.68 ± 0.89	<0.001
CXCL10 (NPX)	9.32 ± 0.84	9.74 ± 1.09	<0.001

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OR (95% CI) for DSPN was 2.06 (1.30; 3.26) when using WHO criteria and 2.72 (1.42; 5.19) when using the stricter IDF classification (Table 2, model 2). A 5-cm increment of waist circumference was also associated with incident DSPN (multivariable OR [95% CI] 1.24 [1.11; 1.39]). The associations between waist circumference and DSPN were robust after additional adjustment for metabolic factors (model 3). The association between waist circumference and DSPN was linear (Supplementary Fig. 1*B*) (*P* for nonlinearity = 0.941).

For further measurements of abdominal obesity (WHR and WHtR), similar associations were observed (Table 2). Higher compared with lower WHR indicated a positive, but not statistically significant, association with DSPN (multivariable adjusted OR [95% CI] 1.51 [0.91; 2.50]). When we investigated WHR as a continuous trait, a positive statistically significant association was observed for an increment of 0.1 units of WHR and incident DSPN (multivariable adjusted OR [95% CI] 1.79 [1.20; 2.67]). After additional adjustment for cardiometabolic factors, the associations were slightly attenuated (model 3). The OR (95% CI) of DSPN was 1.68 (1.19; 2.35) for an increment of 0.1 units of WHtR, with slightly weaker associations after adjustment for the cardiometabolic markers (model 3).

Interaction analyses did not point toward differences by prediabetes/diabetes status regarding associations between anthropometric measures and DSPN (Supplementary Table 1).

Mediation Analysis

Table 3 shows the associations between the potential mediators (IL-6, TNF-α, CCL7, CXCL9, CXCL10, DNER, CD40, and TNFRSF9) and BMI or waist circumference. CCL7, CXCL10, and DNER were associated with both anthropometric measurements. None of the other biomarkers showed an association with the anthropometric measures. In line with this observation, statistically significant mediation effects for the associations of BMI and waist circumference with DSPN were observed for these three markers (CCL7, CXCL10, and DNER) (Table 4). The proportion of the total effect for the association between BMI and DSPN explained by the mediator was 6.8% (0%; 22.2%) for CCL7, 10.5% (1.3%; 32.8%) for

Table 1—Continued

Variable	No incident DSPN	Incident DSPN	*P*
DNER (NPX)	8.32 ± 0.26	8.34 ± 0.26	0.084
CD40 (NPX)	10.19 ± 0.32	10.31 ± 0.34	0.001
TNFRSF9 (NPX)	6.19 ± 0.44	6.41 ± 0.47	<0.001

Data are shown as mean ± SD, median (interquartile range), or percentages. The *P* values are derived from logistic regression analysis (likelihood ratio tests comparing models with the respective variable and age and sex as independent variables to models with age and sex only). All analyses were adjusted for age and sex except associations with age (sex-adjusted only) or sex (age-adjusted only). Data from this table have been published in a previous report from the KORA study (10). CD, cluster of differentiation; eGFR, estimated glomerular filtration rate; HbA_{1c}, glycated hemoglobin; M, men; W, women. *DSPN defined by MNSI >3. †Socioeconomic status based on a score including education, income, and occupation and classified as low (1–9 points), moderate low (10–12 points), moderate (13–15 points), moderate high (16–19 points), and high (>19 points). §Prediabetes was defined as IFG and/or IGT. ‡Blood pressure of ≥140/90 mmHg or antihypertensive medication given that the subjects were aware of as hypertensive. ¶Individuals using lipid-lowering drugs excluded (*n* = 123). #Classified as none (0 g/day), moderate (≥0 to <20 g/day for women and ≥0 to <40 g/day for men), or high (≥20 g/day for women and ≥40 g/day for men). ‖Defined as >1 h sports/week in summer and winter. ††Nonsteroidal anti-inflammatory drugs except acetylsalicylic acid used as platelet aggregation inhibitor.

CXCL10, and 7.5% (0.4%; 25.6%) for DNER. Similar mediation effects were observed for waist circumference, with proportions explaining the associations between waist circumference and DSPN of 6.9% (0.7%; 21.3%) for CCL7, 11.2% (2.8%; 30.2%) for CXCL10, and 9.2% (1.3%; 32.4%) for DNER.

Table 2—ORs and 95% CIs of incident DSPN according to anthropometric measures in KORA F4 (*n* = 513)

	N	Cases	Model 1*	Model 2†	Model 3‡
BMI (kg/m^2)					
Normal weight (<25)	120	14	1	1	1
Overweight (25 to <30)	245	67	2.75 (1.45; 5.22)	3.06 (1.57; 5.97)	3.20 (1.57; 6.53)
Obese (≥30)	148	46	3.31 (1.70; 6.47)	3.47 (1.72; 7.00)	3.21 (1.50; 6.87)
P trend			0.001	0.002	0.017
BMI per 5 kg/m^2	513	127	1.66 (1.27; 2.16)	1.68 (1.28; 2.22)	1.58 (1.17; 2.14)
Waist circumference (cm)					
WHO definition					
Low (M: <102, W: <88)	249	44	1	1	1
High (M: ≥102, W: ≥88)	264	83	2.17 (1.40; 3.36)	2.06 (1.30; 3.26)	1.87 (1.13; 3.08)
IDF definition					
Low (M: <94, W: <80)	107	14	1	1	1
High (M: ≥94, W: ≥80)	406	113	2.60 (1.40; 4.84)	2.72 (1.42; 5.19)	2.55 (1.28; 5.05)
Waist circumference per 5 cm	513	127	1.25 (1.12; 1.39)	1.24 (1.11; 1.39)	1.21 (1.07; 1.36)
WHR					
Low (M: <0.95, W: <0.8)	173	35	1	1	1
High (M: ≥0.95, W: ≥0.8)	340	92	1.62 (1.01; 2.61)	1.51 (0.91; 2.50)	1.30 (0.76; 2.23)
WHR per 0.1 units	513	127	1.83 (1.24; 2.68)	1.79 (1.20; 2.67)	1.60 (1.03; 2.49)
WHtR					
WHtR per 0.1 units	513	127	1.73 (1.25; 2.38)	1.68 (1.19; 2.35)	1.51 (1.04; 2.18)

M, men; W, women. *Model 1: adjusted for age and sex. †Model 2: model 1 plus adjustment for socioeconomic status, smoking status, alcohol intake, physical activity, and height (not in models for BMI and WHtR). ‡Model 3: model 2 plus adjustment for HbA_{1c}, hypertension, HDL cholesterol, LDL cholesterol, and triglycerides.

CONCLUSIONS

This prospective study shows a positive association of general and abdominal obesity with the development of DSPN in a population-based cohort of older individuals. The associations were linear and no interaction by prediabetes/diabetes status was observed. Three biomarkers of subclinical inflammation (CCL7, CXCL10, and DNER) partially mediated the association between anthropometric measures and DSPN.

This study is the first to investigate the association of general and abdominal obesity with incident DSPN using a prospective population-based design. One of our main findings is that both general and abdominal obesity were associated with the development of DSPN.

Another finding of our study is that we did not observe an interaction effect by diabetes or prediabetes in the association between obesity and DSPN. There is one prospective cohort study that investigated the association between BMI and waist circumference and risk of diabetic polyneuropathy among individuals with type 2 diabetes (7,8). In line with our findings, the ADDITION Denmark study also reported a positive association between both anthropometric measures and risk of diabetic polyneuropathy. In that cohort including solely subjects with type 2 diabetes, the relative risks (RRs) of diabetic polyneuropathy for anthropometric measures were slightly lower (RR [95% CI] 1.14 [1.05; 1.25] for BMI per 2 kg/m^2 and 1.14 [1.06; 1.23] for 5-cm differences in waist circumference, respectively) compared with our findings (OR [95% CI] 1.68 [1.28; 2.22] for BMI per 5 kg/m^2 and 1.24 [1.11; 1.39] for waist circumference per 5 cm, respectively) (7). However, the ADDITION Denmark study used the MNSI questionnaire to assess neuropathic symptoms rather than the examination portion of the MNSI to assess neuropathic signs as used in the current study. It is well known that neuropathic signs (deficits and impairments) rather than symptoms are associated with neuropathy confirmed by nerve conduction studies (27). Thus, the sensitivity to detect cases with incident DSPN was higher in the KORA F4/FF4 study than in the ADDITION Denmark study. Investigating patients with type 1 diabetes, the Diabetes Control and Complications Trial/Epidemiology of Diabetes Interventions and Complications

Table 3—Association between potential mediators and anthropometric measures in KORA F4 (n = 513)*

	BMI (kg/m^2)		Waist circumference (cm)	
Potential mediator	β	95% CI	β	95% CI
IL-6 (pg/mL)	0.086	−0.158; 0.329	0.030	−0.062; 0.122
TNF-α (pg/mL)	0.161	−0.174; 0.496	0.069	−0.059; 0.196
CCL7 (NPX)	**0.063**	**0.001; 0.125**	**0.025**	**0.001; 0.048**
CXCL9 (NPX)	0.008	−0.080; 0.958	0.012	−0.021; 0.046
CXCL10 (NPX)	**0.135**	**0.032; 0.239**	**0.059**	**0.020; 0.098**
DNER (NPX)	**−0.043**	**−0.072; −0.014**	**−0.018**	**−0.029; −0.007**
CD40 (NPX)	0.008	−0.029; 0.045	0.003	−0.011; 0.017
TNFRSF9 (NPX)	0.046	−0.005; 0.098	0.014	−0.006; 0.034

Boldface type indicates statistically significant findings. The association was interpreted as statistically significant if the 95% CI of the β-estimate did not include the null value.
*Associations are adjusted for age, sex, socioeconomic status, smoking status, alcohol intake, physical activity, and height (not in models for BMI).

(DCCT/EDIC) study reported that combining the MNSI questionnaire and examination scores did not substantially improve the prediction of clinical polyneuropathy compared with the separate use of both tools (28). Moreover, in a more recent report from the ADDITION Denmark study, neuropathy was confirmed by a point-of-care nerve conduction study device (DPNCheck). This information was only available at follow-up after 13 years, but not at baseline (8). Thus, that study is limited in its ability to assess the predictive value of risk factors for the development of diabetic polyneuropathy. Furthermore, that study only adjusted for few confounders, namely sex, age, diabetes duration, and trial randomization, and it remains unclear whether other factors might influence this association (8). In our report, we investigated several potential confounders for this association, including socioeconomic status, smoking status, alcohol intake, physical activity, and height (if appropriate) and also cardiometabolic factors, such as HbA_{1c}, hypertension, HDL cholesterol, LDL cholesterol, and triglycerides. Our findings show that the association between general and abdominal obesity still persists even after controlling for important risk factors. In addition, several cross-sectional studies reported associations between obesity and polyneuropathy in individuals with and without diabetes (2–6); however, findings of these studies are more difficult to interpret because cross-sectional studies are more prone to recall bias and reverse causality.

Importantly, general and abdominal obesity are modifiable risk factors, and thus, the question arises whether weight reduction in individuals with overweight or obesity by interventions such as lifestyle modification or bariatric surgery have an influence on the prevention of DSPN. So far, there is hardly any evidence available in this context. In the Diabetes Prevention Program (DPP), a post hoc analysis was performed after a mean follow-up of 15 years. Among the subgroup of participants whose most recent HbA_{1c} was ≥6.5%, representing ~26% of the cohort, the lifestyle intervention group showed reductions in the onset of neuropathy assessed by the 10-g monofilament compared with the placebo (RR [95% CI] 0.38 [0.19; 0.75]) and metformin (RR [95% CI] 0.39 [0.19; 0.79]) groups, suggesting that people who convert to diabetes lifestyle intervention may reduce the incidence of neuropathy (29). A recent retrospective cohort study among patients with type 2 diabetes demonstrated that bariatric surgery was associated with a 63% lower risk

Table 4—Effect estimates (95% CI) for incident DSPN for 5-unit increase in anthropometric measure and estimated percent mediated by inflammatory biomarker*

	Direct effect	Indirect effect	Total effect	Percent mediated†
BMI (kg/m^2)				
TNF-α	0.013 (0.005; 0.021)	0.000 (0.000; 0.002)	0.014 (0.005; 0.021)	1.6 (−2.0; 9.7)
IL-6	0.014 (0.005; 0.024)	0.000 (−0.004; 0.003)	0.014 (0.005; 0.022)	1.3 (−18.6; 20.3)
CCL7	0.013 (0.005; 0.020)	0.001 (0.000; 0.004)	0.014 (0.005; 0.022)	**6.8 (0.0; 22.2)**
CXCL9	0.014 (0.005; 0.023)	0.000 (−0.003; 0.002)	0.014 (0.006; 0.021)	7.0 (−16.4; 14.2)
CXCL10	0.012 (0.005; 0.019)	0.002 (0.000; 0.006)	0.014 (0.005; 0.021)	**10.5 (1.3; 32.8)**
DNER	0.015 (0.006; 0.026)	−0.001 (−0.005; 0.000)	0.014 (0.005; 0.021)	**−7.5 (−0.4; −25.6)**
CD40	0.014 (0.005; 0.022)	0.000 (−0.002; 0.002)	0.014 (0.005; 0.021)	1.4 (−9.6; 11.3)
TNFRSF9	0.012 (0.005; 0.020)	0.001 (0.000; 0.004)	0.014 (0.006; 0.021)	7.8 (−1.7; 21.6)
Waist circumference (cm)				
TNF-α	0.002 (0.000; 0.005)	0.000 (0.000; 0.000)	0.002 (0.000; 0.005)	1.9 (−1.1; 8.8)
IL-6	0.002 (0.000; 0.006)	0.000 (−0.001; 0.000)	0.002 (0.000; 0.005)	9.0 (−1.7; 15.2)
CCL7	0.002 (0.000; 0.004)	0.000 (0.000; 0.001)	0.002 (0.000; 0.005)	**6.9 (0.7; 21.3)**
CXCL9	0.002 (0.000; 0.005)	0.000 (0.000; 0.001)	0.002 (0.000; 0.005)	2.8 (−10.5; 15.6)
CXCL10	0.002 (0.000; 0.004)	0.000 (0.000; 0.001)	0.002 (0.000; 0.005)	**11.2 (2.8; 30.2)**
DNER	0.002 (0.000; 0.007)	0.000 (−0.001; 0.000)	0.002 (0.000; 0.005)	**−9.2 (−1.3; −32.4)**
CD40	0.002 (0.000; 0.005)	0.000 (0.000; 0.000)	0.002 (0.000; 0.005)	1.5 (−9.7; 13.0)
TNFRSF9	0.002 (0.000; 0.005)	0.000 (0.000; 0.001)	0.002 (0.000; 0.005)	5.4 (−5.4; 18.3)

Data are β (95% CI) unless otherwise indicated. *Models adjusted for age, sex, socioeconomic status, smoking status, alcohol intake, physical activity, and height (not in models for BMI). †Boldface type indicates statistically significant findings. The mediation effect was interpreted as statistically significant if the 95% CI of the percent mediated effect did not include the null value.

of DSPN (hazard ratio [95% CI] 0.37 [0.30; 0.47]) compared with patients who did not have surgery (30). However, more well-designed clinical and observational studies are needed to investigate the potential of weight loss regarding risk reduction of DSPN.

Furthermore, our study extends the current literature, because we investigated potential mediators in the relationship between obesity and incident DSPN and found three biomarkers of inflammation, which partly explained the association. Obesity is a proinflammatory state (31,32), and we previously identified multiple proinflammatory cytokines, chemokines, and soluble forms of membrane-bound inflammation-related proteins as novel risk factors for DSPN (9,10). In the current study, we identified two chemokines (CCL7 and CXCL10) and the soluble form of the transmembrane receptor DNER as proteins, which may explain the mechanism linking obesity and incident DSPN. CCL7 and CXCL10 are both expressed in subcutaneous and visceral adipose tissue (33). CCL7 expression has primarily been found in adipocytes compared with stromal vascular cells, and its expression is upregulated in obesity, potentially due to activation of the inhibitor of κB kinase-β and c-Jun N-terminal kinase pathways (34). CXCL10 expression has been reported for mature human adipocytes (35), and this chemokine can be induced by factors derived from macrophages for which numbers in adipose tissue are positively correlated with the degree of obesity (36). Interestingly, CCR2 and CXCR3, the receptors for CCL7 and CXCL10, respectively, are expressed on nerves (33), suggesting that a direct effect on neuronal cells and DSPN risk is biologically plausible. In line with this, we recently found that both chemokines had neurotoxic effects on the SH-SY5Y neuroblastoma cell line, which is commonly used to assess neurotoxicity (10). DNER is a transmembrane protein that is mainly expressed in brain, adrenal gland, and pituitary tissue. However, it is also found in human adipose tissue–derived mesenchymal stem cells, and its knockdown has been shown to promote human adipose tissue–derived mesenchymal stem cell adipogenesis (37), which may link the quality of adipose tissue expansion with DSPN.

Our mediation analysis for these three biomarkers revealed a potential mediation effect ranging from 7% to 11%. The effect size of the mediation indicates that factors other than subclinical inflammation explain part of the relationship between obesity and DSPN. First, it is conceivable that a larger cohort would have allowed us to identify additional inflammation-related biomarkers associated with anthropometric measures (and thus more potential mediators), so that the current analysis based on three biomarkers most likely underestimates the proportion of the association between obesity and incident DSPN that is explained by subclinical inflammation. Second, biomarkers of oxidative stress appear as promising candidates because oxidative stress and inflammation have been described as pathomechanisms that may integrate metabolic derangements such as hyperglycemia and dyslipidemia and link them to the development of cardiometabolic diseases (38,39). In line with this, higher levels of methylglyoxal, myeloperoxidase, and extracellular superoxide dismutase 3 have recently been found associated with prevalent and/or incident DSPN (7,40).

The current study has a number of strengths, including the prospective study design and the population-based sample, which reduced recall bias, selection bias, and risk of reverse causality. The use of different statistical approaches enabled us to investigate multivariable adjusted associations between general and abdominal obesity with the development of DSPN by exploring linear and nonlinear relations, interactions with prediabetes/diabetes, and mediation effects of biomarkers of inflammation for these associations. This study has also some limitations. First, the MNSI allowed the diagnosis of possible DSPN, but further examinations were not possible in this epidemiological setting. Second, we had to combine participants with prediabetes and diabetes for a meaningful interaction analysis. To confirm our results, more studies are needed that investigate the interaction effect between prediabetes or diabetes and obesity related to risk of DSPN. Generally, findings from observational studies do not necessarily serve as a proof of mechanistic or causal interactions. Third, we had to rely on anthropometric measurements based on height, weight, and waist and hip circumference, whereas data on fat mass (absolute or as percentages) were only available for KORA FF4 but not for KORA F4 (baseline). Fourth, follow-up information was missing for a large number of study participants, and it has been shown previously that participants with missing follow-up information were older, had a higher BMI and waist circumference, and were in general less healthy (higher HbA_{1c}, lower estimated glomerular filtration rate, and higher levels of inflammatory biomarkers) compared with participants with follow-up information (9). Thus, it is likely that a relevant proportion of those lost to follow-up had developed DSPN. Finally, our study sample was at an older age and from one area in South Germany, and thus, the generalizability of the data to populations at a younger age or from different geographic locations or ethnic backgrounds is unknown. In conclusion, this study demonstrates that general obesity and abdominal obesity are prospectively associated with the development of DSPN in a population-based cohort. We identified a linear relationship between anthropometric measures and DSPN, and no differences by prediabetes/diabetes status were observed. Biomarkers of subclinical inflammation partly mediated the association between obesity and DSPN. Importantly, general and abdominal obesity are modifiable risk factors for DSPN, and thus, recommendations regarding the prevention of DSPN should include the adherence to a healthy body weight with a normal BMI and waist circumference.

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