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Urinary metals and incident diabetes in midlife women: Study of Women's Health Across the Nation (SWAN)

Xin Wang ,[1] Carrie A Karvonen-Gutierrez,[1] William H Herman,[2] Bhramar Mukherjee,[3] Siobán D Harlow,[1] Sung Kyun Park [1,4]

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[1]Department of Epidemiology, University of Michigan School of Public Health, Ann Arbor, Michigan, USA
[2]Department of Internal Medicine, University of Michigan School of Medicine, Ann Arbor, Michigan, USA
[3]Department of Biostatistics, University of Michigan School of Public Health, Ann Arbor, Michigan, USA
[4]Department of Environmental Health Sciences, University of Michigan School of Public Health, Ann Arbor, Michigan, USA

Correspondence to
Dr Sung Kyun Park;
sungkyun@umich.edu

ABSTRACT

Introduction Environmental exposure to metals may play a role in the pathogenesis of diabetes; however, evidence from human studies is limited. We prospectively evaluated the associations of 20 urinary metal concentrations and their mixtures with incident diabetes in the Study of Women's Health Across the Nation, a multisite, multiethnic cohort study of midlife women.

Research design and methods The sample included 1237 white, black, Chinese and Japanese-American women, aged 45–56 years, free of diabetes at baseline (1999–2000) who were followed through 2016. Concentrations of 20 metals (arsenic, barium, beryllium, cadmium, cobalt, chromium, cesium, copper, mercury, manganese, molybdenum, nickel, lead, antimony, tin, thallium, uranium, vanadium, tungsten and zinc) were measured in urine specimens at baseline. Incident diabetes was identified annually by fasting glucose ≥126 mg/dL, self-reported doctor-diagnosed diabetes, or self-reported use of antidiabetic medications. A non-parametric clustering method, k-means clustering, was used to identify subgroups with different exposure patterns to metal mixtures.

Results After multivariable adjustment, the HR (95% CI) for diabetes associated with each doubling increase in urinary metal concentrations was 1.19 (1.10 to 1.30) for arsenic and 1.20 (1.05 to 1.37) for lead, in Cox proportional hazards models after controlling for multiple comparisons. A doubling in urinary excretion of zinc was associated with higher risk of diabetes (adjusted HR 1.31, 95% CI 1.11 to 1.53). Two distinct exposure patterns to metal mixtures—'high' versus 'low'—were identified. Participants assigned to the 'high' pattern had higher overall concentrations of all metals compared with those classified into the 'low' pattern. Adjusted HR for diabetes associated with 'high' pattern compared with 'low' was 1.42 (1.08 to 1.87).

Conclusions Higher urinary concentrations of arsenic and lead, increased urinary excretion of zinc, as well as higher overall exposure to metal mixtures were associated with elevated risk of diabetes. Future studies should further investigate the underlying mechanisms.

Significance of this study

What is already known about this subject?

- Exposure to metals, particularly arsenic, may play a role in the induction or exacerbation of type 2 diabetes.
- There is limited evidence on the associations between other metals and type 2 diabetes.
- No epidemiologic studies have examined the association of exposure to metal mixtures with type 2 diabetes.

What are the new findings?

- Higher urinary concentrations of arsenic and lead were associated with a higher risk of developing diabetes over 16 years of follow-up.
- An increase in urinary excretion of zinc was associated with a higher risk of developing diabetes.
- A high overall exposure to metal mixtures was also associated with a higher risk of developing diabetes.

How might these results change the focus of research or clinical practice?

- Widespread exposure to metals and their mixtures may be a key contributor to the epidemics of type 2 diabetes and needs to be considered in diabetes care.

INTRODUCTION

To date, most epidemiologic studies of risk of type 2 diabetes mellitus have focused on the potential impact of genetics, unhealthy diets, and sedentary lifestyles. The potential contributions of environmental toxicants to the epidemic of diabetes have received less attention. The general population is commonly exposed to metals through food, drinking water, and ambient air. Dietary intake of toxic metals has been a significant public health concern, particularly for populations that consume contaminated drinking water and/or rice.[1 2] Exposure to metals may play a role in the induction or exacerbation of diabetes: arsenic has been associated with diabetes in a number of studies.[3] Other metals including cadmium and lead have been examined in relation to risk of diabetes but the studies have been limited, inconsistent and mostly cross-sectional.[4 5] Associations of environmental exposure to most other metals with diabetes have not been investigated.

Toxic metals such as arsenic, cadmium and lead are well-known inducers of oxidative stress in a variety of tissues and cell types.[6] The accumulation of these metals in the pancreatic islets is hypothesized to lead to impaired function and apoptotic death of β cells via the induction of oxidative stress.[7 8] Arsenic and cadmium have also been demonstrated to interfere with gene expression involving signal transduction and gene transcription related to insulin pathways, leading to insulin resistance.[9 10] On the other hand, deficiency in essential metals such as zinc attributed to excessive excretion in urine has been related to dysregulation of insulin secretion and glucose transportation.[11] Metal exposures could also be associated with obesity. A recent cross-sectional study found that exposure to mixtures of metals was associated with body mass index (BMI) and waist circumference in the US general population.[12] These findings suggest a need to investigate the role of metal exposures in risk of diabetes in humans, especially in a well-characterized prospective cohort study.

In this study, we report on the associations of 20 urinary metals with the incidence of diabetes over 16 years of follow-up in the Study of Women's Health Across the Nation (SWAN), a multisite, multiethnic prospective cohort study of midlife women. We have previously identified two distinct exposure patterns to metal mixtures in SWAN.[1] The present study was designed to further assess the role of metal mixtures in risk of diabetes.

METHODS

Study population

SWAN is an ongoing, multisite, multiethnic, community-based longitudinal study of the natural history of menopause designed to address the effect of the menopausal transition on subsequent health endpoints.[13] In 1996–1997, 3302 women were enrolled from seven study sites where white women and women from one specified minority group were recruited (black women from Boston, Massachusetts; Pittsburgh, Pennsylvania; Southeast Michigan, Michigan; and Chicago, Illinois; Hispanic women from Newark, New Jersey; Chinese women from Oakland, California; and Japanese women from Los Angeles, California). Eligibility criteria for enrollment into the SWAN cohort included the following: age 42–52 years, intact uterus and at least one ovary, no use of exogenous hormones affecting ovarian function in the past 3 months, at least one menstrual period in the previous 3 months, and self-identification with a site's designated racial/ethnic groups. These women returned for regular examinations annually, and approximately 75% of still-living participants completed the 15th SWAN follow-up visit (2015–2016). All participants provided signed informed consent at each study visit.

Metal concentrations were measured in 1400 participants from the SWAN Multi-Pollutant Substudy (SWAN-MPS),[1 14] which used urine samples from the SWAN Repository collected during the third SWAN follow-up visit (1999–2000) for environmental exposure assessments. SWAN-MPS included only five study sites: Michigan, Boston, Oakland, Los Angeles, and Pittsburgh. Therefore, only white, black, Chinese and Japanese women were included. Of 1400 participants, we excluded 82 women with prevalent diabetes at SWAN-MPS baseline, 1 participant who provided an insufficient quantity of urine, and 80 participants who had no information on key covariates, leaving a final analytic sample of 1237 women including 11 715 observations followed from 1999 to 2016. An overview of our analytic sample is illustrated in online supplementary figure 1.

Diabetes ascertainment

Fasting serum glucose level was determined by hexokinase method (Boehringer Mannheim Diagnostics, Indianapolis, Indiana, USA). At any follow-up visit, participants with one or more of the following were defined as having incident diabetes: (1) fasting serum glucose level ≥126 mg/dL; (2) self-reported use of insulin or oral medications for diabetes; and (3) self-reported physician diagnosis of diabetes. The vast majority of the diabetes cases in this life stage are considered type 2 diabetes.

Urinary metals

Details regarding urinary metal measurements can be found in online supplementary methods. Baseline concentrations of the following 20 metals—total arsenic, barium, beryllium, cadmium, cobalt, chromium, cesium, copper, mercury, manganese, molybdenum, nickel, lead, antimony, tin, thallium, uranium, vanadium, tungsten, and zinc—were measured in these urine samples using high-resolution inductively coupled plasma-mass spectrometry (Thermo Scientific iCAP RQ, Waltham, Massachusetts) following the Centers for Disease Control and Prevention method 3018.3,[15] with modifications for the expanded metals panel, by the Applied Research Center of NSF International (Ann Arbor, Michigan). The limits of detection (LOD) and detection rates are presented in table 1. Participants with metal concentration below LOD were assigned a value equal to LOD divided by the square root of 2. Pairwise Spearman correlations among urinary metal concentrations were calculated.

Other covariates

Age, self-reported race/ethnicity, and education level were assessed through a self-administered questionnaire at the SWAN baseline examination (1996–1997). At each study visit, annual household income, smoking status, alcohol drinking, menopausal status, and use of exogenous hormones were self-reported. BMI was calculated as weight in kilograms divided by the square of height in meters. Waist circumference was measured to the nearest 0.1 cm with a measuring tape placed horizontally around the participant at the narrowest part of the torso. Physical activity was measured by a total score indicating the activity levels during the previous 12 months. Activity was assessed with a modified version of the Kaiser Physical

Table 1 Detection rates and concentrations of urinary metals by incident diabetes: the Study of Women's Health Across the Nation Multi-Pollutant Substudy (N=1237)

Metals	LOD	Per cent >LOD	Median concentration (IQR), µg/L	
			Non-diabetes (n=1135)	Incident diabetes (n=102)
Arsenic	0.3	100	14.24 (6.51–36.69)	14.28 (6.44–39.09)
Barium	0.1	99.7	1.79 (1.00–3.01)	1.70 (1.05–2.91)
Beryllium	0.04	15.7	<LOD (<LOD, <LOD)	<LOD (<LOD, <LOD)
Cadmium	0.06	94.5	0.47 (0.23–0.82)	0.50 (0.22–0.85)
Cobalt	0.05	99.3	0.62 (0.38–0.97)	0.63 (0.41–1.00)
Chromium	0.4	24.3	<LOD (<LOD, <LOD)	<LOD (<LOD, <LOD)
Cesium	0.01	100	4.71 (3.05–7.27)	4.41 (2.70–7.46)
Copper	2.5	97.1	9.60 (6.15–13.85)	10.87 (6.69–16.77)
Mercury	0.05	99.8	1.23 (0.67–2.45)	1.08 (0.55–2.02)
Manganese	0.08	99.6	0.91 (0.62–1.46)	1.07 (0.65–1.78)
Molybdenum	0.3	100	43.83 (24.60–70.73)	41.30 (20.60–68.86)
Nickel	0.8	96.2	3.75 (2.27–5.81)	3.82 (2.58–5.50)
Lead	0.1	97.6	0.80 (0.49–1.27)	0.83 (0.39–1.46)
Antimony	0.04	78.5	0.08 (0.05–0.13)	0.10 (0.05–0.18)
Tin	0.1	96.7	0.95 (0.49–1.81)	1.19 (0.64–2.44)
Thallium	0.02	92.4	0.15 (0.08–0.23)	0.15 (0.08–0.24)
Uranium	0.01	33.3	<LOD (<LOD, 0.01)	<LOD (<LOD, 0.01)
Vanadium	0.6	37.4	<LOD (<LOD, 1.18)	<LOD (<LOD, 0.81)
Tungsten	0.2	29.0	<LOD (<LOD, 0.21)	<LOD (<LOD, 0.24)
Zinc	2	100	304 (168–506)	434 (257–657)

LOD, limit of detection.

Activity Survey,[16] which consists of 38 questions with primarily Likert-scale responses about physical activity in various domains, including sports/exercise, household/caregiving, and daily routine. Domain-specific indices were derived by averaging the ordinal responses to questions in each domain, resulting in values from 1 to 5. Thus, the total physical activity score ranged from 3 to 15, with 15 indicating the highest level of activity. Dietary seafood and rice intake and zinc intake from diet and supplements were collected using a detailed semiquantitative Food Frequency Questionnaire (FFQ) adopted from the Block FFQ.[1] Total energy intake was obtained from the FFQ based on each food intake. Urinary-specific gravity was determined using a handheld digital refractometer (ATAGO Model PAL-10S, Tokyo, Japan) at the same time as metal measurements as a marker of urine dilution. In SWAN, uniform protocols were used to collect information on these covariates across the study sites.

Statistical analysis

Cox proportional hazards models were used to estimate the HR and 95% CI of incident diabetes in relation to each metal concentration. Participants contributed follow-up time from the date of metal measurements to the date of first diabetes event, whichever occurred first for incident cases, or the end of the study for non-cases. Given the highly skewed distributions of urinary metal concentrations, logarithmic transformations with base 2 were applied to all metal concentrations and the HRs and 95% CIs were interpreted as effects of a twofold increase in each urinary metal concentration. For metals with low detection rate (beryllium, chromium, uranium, vanadium, and tungsten; table 1), HRs were calculated comparing participants with metal concentration above the LOD with those with value below the LOD.

Potential confounders were adjusted progressively in the Cox models. Initial regression models included adjustment for age at baseline, race/ethnicity, study site, and specific gravity (log-transformed), while subsequent models further adjusted for education, annual household income, BMI and waist circumference at the time of urinary metal measurements, smoking, alcohol drinking, physical activity score, total energy intake, menopausal status, and hormone therapy (full model). We decided not to include time-varying BMI and waist circumference in our analysis because of their role as a potential diabetes risk factor and the fact that they could be affected by metal exposures at baseline.[12] For arsenic, cadmium and mercury, we additionally adjusted for seafood and rice intake in the full model, which have been identified as important determinants of their urinary concentrations

in our previous study.[1] For zinc, we additionally adjusted for total zinc intake from food and supplements in the full model to better capture the potential effect of urinary zinc excretion on diabetes that is independent of dietary zinc intake. For other essential metals, such as copper, no dietary intake was adjusted due to lack of data. We adjusted p values for multiple comparisons at a false discovery rate (FDR) of 0.05 using the Benjamini-Hochberg method.[17]

To quantify the differences in risk of diabetes between subgroups corresponding to different exposure patterns to metal mixtures, a non-parametric clustering method, k-means clustering, was applied. Briefly, this approach creates a single variable with k categories representing different clusters, where participants within the same cluster are as similar as possible and participants from different clusters are as dissimilar as possible, in terms of their urinary metal concentrations. The k-means clustering was performed for the metals of which the detection rate was ≥70%. All log-transformed specific gravity-adjusted urinary metal concentrations were standardized to make variables comparable before the k-means clustering. The number of optimal clusters (k) was determined based on cubic clustering criterion, elbow method, and interpretability. HRs of diabetes incidence were estimated between subgroups (clusters) with different exposure patterns to metal mixtures using the Cox models. We also calculated survival probability of diabetes of participants in different subgroups throughout 16 years of follow-up using adjusted survival curves recommended by Hernán[18] and displayed the results graphically. Briefly, a discrete-time hazards model with adjustment of confounding factors was fitted to estimate the conditional survival probability of diabetes under different exposure patterns to metal mixtures in a counterfactual causal framework.[18]

We recognized that selective participation into the SWAN-MPS and selective loss to follow-up that occurred after the metal measurements may have biased the estimates of associations between metals and diabetes. To mitigate these biases, we assigned weights to participants based on inverse probability weighting (IPW). Directed acyclic graphs illustrating the potential selection bias and details of estimation of IPW are described in online supplementary methods, online supplementary figure 2 and online supplementary figure 3.

In sensitivity analyses, we calculated HRs in relation to specific gravity-adjusted metal concentrations (urinary metal concentration × (1.017–1)/specific gravity–1), where 1.017 was the median level of specific gravity in our analytic sample. We also additionally adjusted for baseline levels of fasting glucose, systolic blood pressure, total cholesterol level, high-density lipoprotein cholesterol level, triglyceride level, and family history of diabetes. All analyses were conducted using SAS V.9.4.

RESULTS

Among 1237 SWAN participants free of diabetes at baseline, 102 developed diabetes during 17 005 person-years of follow-up, with an incidence of 6.0 per 1000 person-years. Women with incident diabetes were more likely to be black, from Michigan, to have higher BMI and waist circumference, of lower education level, and to be current or former smokers, and hormone users (table 2). The percentage of women with detectable concentrations of individual metals ranged from 15.7% to 100% (table 1). Participants with incident diabetes had higher copper, manganese, nickel, lead, tin, zinc and lower cesium concentrations than those without incident diabetes. Most metals were modestly and positively correlated with each other (online supplementary figure 4).

Table 3 summarizes the associations between urinary metal concentrations (detection rate ≥70%) and incident diabetes. In the initial models, significant associations were found for urinary tin (p=0.03) and zinc (p<0.0001). After full adjustment for covariates, HR for diabetes associated with each doubling of urinary metal concentration was 1.19 (95% CI 1.10 to 1.30, p<0.0001) for arsenic, 1.20 (95% CI 1.05 to 1.37, p=0.006) for lead, 1.11 (95% CI 1.01 to 1.22, p=0.04) for tin, and 1.31 (95% CI 1.11 to 1.53, p=0.001) for zinc. To adjust for multiple comparisons, a significance level of α=0.006 was used, which corresponded to an FDR of 5% using the Benjamini-Hochberg method. This adjustment for multiple comparisons left only arsenic, lead, and zinc as significant independent predictors (p<0.006) of diabetes. No significant association was detected between metals with detection rate <70% and diabetes (table 4).

Two distinct clusters of participants were identified based on the exposure profiles of metal mixtures through k-means clustering according to both the cubic clustering criterion and the elbow method, which was consistent with our previous finding.[1] These two clusters were labeled as 'high' (n=604) and 'low' (n=633) for the exposure patterns. Participants assigned to the 'high' cluster had higher overall exposures to all the metals compared with those classified into the 'low' cluster (online supplementary figure 5). Adjusted survival curves of diabetes by these two clusters are shown in figure 1. Adjusted HR for diabetes was 1.42 (95% CI 1.08 to 1.87, p=0.01) for women in the 'high' cluster compared with those in the 'low' cluster in the Cox model.

Similar results were observed when specific gravity-adjusted concentrations of urinary metals were used instead of adjusting for specific gravity in the Cox models for urine dilution adjustment (online supplementary table 1). Additional adjustment for baseline levels of fasting glucose, systolic blood pressure, total cholesterol level, high-density lipoprotein cholesterol level, triglyceride level, and family history of diabetes did not alter our results (online supplementary table 2).

DISCUSSION

In this multisite, multiethnic cohort study of women at midlife, urinary arsenic, lead, and zinc concentrations were prospectively associated with incidence of diabetes

Table 2 Characteristics at the time of metal measurements according to incident diabetes status: the Study of Women's Health Across the Nation Multi-Pollutant Substudy

	No diabetes (n=1135)	Diabetes (n=102)
Age (years)*	49.5 (47.4–51.6)	50.0 (47.7–52.2)
Race/ethnicity		
White	601 (53.0)	39 (38.2)
Black	202 (17.8)	35 (34.1)
Chinese	150 (13.2)	14 (13.7)
Japanese	182 (16.0)	14 (13.7)
Study site		
Michigan	172 (15.2)	33 (32.4)
Boston	187 (16.5)	7 (6.9)
Oakland	263 (23.2)	20 (19.6)
Los Angeles	327 (28.8)	19 (18.6)
Pittsburgh	186 (16.4)	23 (22.6)
Body mass index (kg/m^2)	25.3 (22.2–30.0)	31.6 (26.9–37.3)
Waist circumference (cm)	80.6 (72.9–91.2)	92.3 (82.3–108.3)
Education		
High school or less	189 (16.7)	20 (19.6)
Some college	343 (30.2)	43 (42.2)
College and above	603 (53.1)	39 (38.2)
Household income		
Less than $19 999	55 (5.0)	10 (0.10)
$20 000–$49 999	282 (25.8)	26 (0.25)
$50 000–$99 999	447 (40.9)	47 (0.46)
$100 000 or more	309 (28.3)	19 (0.19)
Smoking status		
Never	737 (64.9)	58 (56.9)
Former	299 (26.3)	29 (28.4)
Current	99 (8.7)	15 (14.7)
Alcohol consumption		
Infrequent	573 (50.5)	63 (61.8)
Moderate	271 (23.9)	25 (24.5)
Heavy	291 (25.6)	14 (13.7)
Physical activity score	8.0 (6.7–9.1)	7.4 (6.6–8.5)
Menopausal status		
Premenopausal	796 (70.1)	64 (62.8)
Postmenopausal	162 (14.3)	18 (17.6)
Unknown†	177 (15.6)	20 (19.6)
Hormone therapy	230 (20.3)	28 (27.5)
Family history of diabetes		
Yes	367 (32.3)	55 (53.9)
No	418 (36.8)	25 (24.5)
Unknown	350 (30.8)	22 (21.6)
Systolic blood pressure (mm Hg)	110.0 (101.0 to 120.0)	120.0 (109.0–128.0)
Total cholesterol (mg/dL)	197.0 (175.0–219.0)	190.0 (172.0–217.0)
HDL cholesterol (mg/dL)	61.0 (51.0–72.0)	52.0 (45.0–60.0)

Continued

Table 2 Continued

	No diabetes (n=1135)	Diabetes (n=102)
Triglyceride (mg/dL)	94.0 (71.0–135.0)	137.5 (93.0–195.0)
Fasting glucose (mg/dL)	84.9 (80.3–89.4)	96.3 (90.3–102.7)
Dietary seafood intake (times/week)	1.4 (0.8–2.5)	1.5 (0.8–2.8)
Dietary rice intake (times/week)	2.0 (1.0–5.5)	2.0 (0.6–5.5)
Total zinc intake (mg/day)	11.0 (7.6–20.5)	11.4 (8.5–21.5)
Total energy intake (kcal)	1661 (1324–2110)	1950 (1475–2440)

*Data are median (IQR) or n (%).
†Menopausal status unknown due to hormone therapy or hysterectomy.
HDL, high-density lipoprotein.

after adjusting for sociodemographic variables, lifestyle factors, BMI, waist circumference, menopausal status, use of hormones and dietary sources. These associations remained significant after further controlling for multiple comparisons. A metal mixtures analysis revealed that a 'high' overall exposure pattern to metals was associated with a higher incidence of diabetes.

Arsenic

We found a positive association between total arsenic in urine and incidence of diabetes. Inorganic arsenic is a toxicant and its common sources include drinking water and certain foods (eg, rice, seafood).[1] After absorption through the gastrointestinal tract, inorganic arsenic is metabolized into monomethylarsonate (MMA) and dimethylarsinate (DMA), which are excreted rapidly into the urine together with inorganic arsenic.[19] The sum of inorganic arsenic, MMA, and DMA in the urine mainly reflects inorganic arsenic exposure.[19] Epidemiologic evidence has supported a possible role of arsenic in diabetes. High exposure to arsenic in drinking water (≥50 µg/L) has been associated with increased risk of diabetes in areas such as Taiwan and Bangladesh, where historical problems of arsenic contamination exist.[20] Association between arsenic and diabetes has also been reported in populations with low-moderate exposure (<50 µg/L in drinking water). In the USA, urinary

Table 3 HR for incident diabetes with twofold increase in urinary metal concentrations

	Initial model*		Full model†	
Metals	HR (95% CI)	P value	HR (95% CI)	P value‡
Arsenic	1.06 (0.98 to 1.15)	0.13	1.19 (1.10 to 1.30)	<0.0001
Barium	0.98 (0.88 to 1.09)	0.70	0.96 (0.85 to 1.09)	0.53
Cadmium	1.00 (0.90 to 1.10)	0.92	0.96 (0.86 to 1.07)	0.42
Cobalt	0.97 (0.85 to 1.10)	0.60	1.01 (0.88 to 1.15)	0.90
Cesium	1.12 (0.92 to 1.37)	0.25	1.23 (0.98 to 1.50)	0.06
Copper	1.06 (0.91 to 1.23)	0.47	0.96 (0.82 to 1.13)	0.65
Mercury	0.83 (0.75 to 0.92)	0.03	0.92 (0.82 to 1.03)	0.12
Manganese	1.14 (0.94 to 1.37)	0.18	1.10 (0.90 to 1.35)	0.33
Molybdenum	0.93 (0.81 to 1.07)	0.29	1.04 (0.90 to 1.21)	0.58
Nickel	1.08 (0.93 to 1.25)	0.33	1.15 (0.98 to 1.35)	0.08
Lead	1.12 (0.99 to 1.27)	0.07	1.20 (1.05 to 1.37)	0.006
Antimony	1.03 (0.91 to 1.17)	0.61	1.07 (0.93 to 1.22)	0.36
Tin	1.10 (1.01 to 1.20)	0.03	1.11 (1.01 to 1.22)	0.04
Thallium	1.05 (0.95 to 1.16)	0.38	1.04 (0.93 to 1.16)	0.52
Zinc	1.48 (1.27 to 1.74)	<0.0001	1.31 (1.11 to 1.53)	0.001

All models were constructed by Cox proportional hazards model.
*Initial model: adjustment for age, race/ethnicity, study site, and specific gravity (log-transformed).
†Full model: initial model with additional adjustment for education, household income, body mass index (baseline level), waist circumference (baseline level), smoking status, alcohol consumption, physical activity score, total energy intake, menopausal status, and use of hormone. In full model, seafood and rice intake was additionally adjusted for arsenic, cadmium, and mercury models; zinc intake from diets and supplements was additionally adjusted for zinc model.
‡Significance level at α=0.006 corresponding to a false discovery rate of 0.05 using the Benjamini-Hochberg method.

Table 4 HR for incident diabetes comparing participants with urinary beryllium, chromium, uranium, vanadium, and tungsten concentrations above the limits of detection with those below the limits of detection

	Initial model*		Full model†	
Metals	HR (95% CI)	P value	HR (95% CI)	P value
Beryllium	1.03 (0.74 to 1.41)	0.86	0.93 (0.66 to 1.30)	0.67
Chromium	0.77 (0.55 to 1.06)	0.11	0.71 (0.50 to 1.01)	0.06
Uranium	0.84 (0.61 to 1.14)	0.27	0.95 (0.68 to 1.33)	0.76
Vanadium	0.85 (0.64 to 1.12)	0.24	0.77 (0.58 to 1.04)	0.08
Tungsten	1.02 (0.73 to 1.40)	0.92	1.07 (0.77 to 1.49)	0.68

All models were constructed by Cox proportional hazards model.
Detection rate: beryllium, 15.7%; chromium, 24.3%; uranium, 33.5%; vanadium, 37.3%; tungsten, 29.2%.
*Initial model: adjustment for age, race/ethnicity, study site, and specific gravity (log-transformed).
†Full model: initial model with additional adjustment for education, household income, body mass index (baseline level), waist circumference (baseline level), smoking status, alcohol consumption, physical activity score, total energy intake, menopausal status, and use of hormone.

arsenic was noted to be positively associated with diabetes prevalence in the general population[21] and in American Indian adults.[22] A diabetogenic effect of arsenic has been supported by mechanistic evidence. Arsenic had been linked with insulin resistance by altering gene expression of a variety of diabetes-related factors and by affecting insulin-stimulated glucose uptake in adipocytes and skeletal muscle cells.[10 23] In the pancreas, arsenic may increase amyloid formation and apoptotic death/damage of pancreatic β cells through the generation of oxidative stress.[7] Additionally, arsenic has been suggested to substitute phosphate and to interact with sulfhydryl groups, which could impair the glucose transport, interrupt the production of energy, and interfere with the ATP-dependent insulin secretion of β cells.[24]

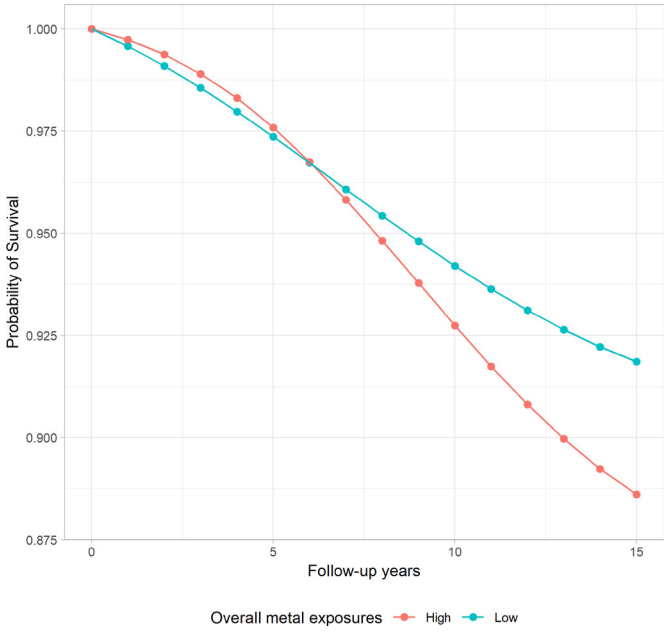

Figure 1 Adjusted survival curves of diabetes by two distinct exposure patterns to metal mixtures, adjusting for age, race/ethnicity, study site, education, household income, body mass index (baseline level), waist circumference (baseline level), smoking status, alcohol consumption, physical activity score, total energy intake, menopausal status, use of hormone, dietary intake of seafood and rice, and zinc intake from diets and supplements.

Lead

We found a significant association between urinary lead concentration and incidence of diabetes. Bone lead stores accrued from cumulative environmental exposures for decades are the major endogenous source of lead.[25 26] Bone lead has been considered a proxy for cumulative exposure to lead and found to be a better biomarker of lead dose than blood lead in recent studies of the relationship between lead exposure and chronic health outcomes such as cardiovascular disease.[27] Urinary lead adjusted for urine dilution has been found to closely reflect lead mobilized from the bone.[25] Given the fact that midlife women may experience an increased bone turnover rate,[28] the observed association could be attributed in part to a greater mobilization of lead from bone into the circulation. Existing evidence on the influence of lead exposure on diabetes risk has been limited and inconsistent: higher lead concentrations in different biological matrices have been observed in patients with diabetes compared with referents in case–control studies.[29 30] On the contrary, no association has been found in two cross-sectional studies in both the USA and South Korea.[5 31] One recent study in China found that higher blood lead concentration was associated with an increased risk of non-alcoholic fatty liver disease, which commonly coexists with type 2 diabetes and has been suggested as a predictor of diabetes risk.[32] Lead is a well-known toxicant that can induce oxidative stress through reactive oxygen species (ROS) generation, where the ROS pathway has been suggested in the pathogenesis of diseases including diabetes.[33] Lead is also thought to disrupt a variety of intracellular signaling pathways by interfering with calcium homeostasis and calcium cellular uptake, and modulating the activity of protein kinase C.[33]

Zinc

Zinc is an essential nutrient that is necessary for biochemical pathways and required by thousands of proteins for

catalytic functions. The human body has no specialized zinc storage system and humans rely on a daily intake of zinc to maintain health. Zinc leaves the body mainly in feces and urine.[11] Zinc intake has been associated with a lower risk of type 2 diabetes in women.[34] In our study, zinc status was assessed from both zinc intake and urinary excretion. We observed a positive association between urinary zinc concentration and risk of diabetes after adjustment for zinc intake from diets and supplements, suggesting urinary zinc excretion independent of dietary sources as a predictor of diabetes. The average intake levels in our participants were greater than the recommended dietary allowance, which is 8 mg/day for women.[35] Our results suggest that women with excess zinc in urine may be at elevated risk of diabetes regardless of the amount of dietary zinc intake. In pancreatic β cells, zinc has been known to be necessary for insulin synthesis, storage and secretion, and has accounted for the conformational integrity of insulin in its hexameric crystalline form.[11] Excessive urinary excretion of zinc was found to lead to a loss of zinc in β cells, which accounted for reduced insulin secretion.[11] Certain zinc complexes showed an insulin-like effect including attenuating hyperglycemia and increasing lipogenesis in animal models.[11] Zinc has also been shown to enhance tyrosine kinase phosphorylation in insulin signal transduction, improving binding of insulin to its receptor and glucose transportation.[11] Zinc is a structural part of antioxidant enzymes such as superoxide dismutase that could protect insulin and β cells from being attacked by free radicals.[11] Despite this evidence, hyperglycemia, on the other hand, was suggested to interfere with the active transportation of zinc back to renal cells, leading to a loss of this mineral in the urine.[36] This raised the possibility that the observed association could also be explained by the increased urinary excretion of zinc in women who already had relatively high glucose levels at baseline. However, we still observed a positive association between urinary zinc and incident diabetes when we additionally excluded women with fasting glucose levels from 100 to 125 mg/dL (impaired fasting glucose) at the study baseline (data not shown), which reduces the likelihood of our findings being a result of reverse causation.

Other metals

Our data provided modest evidence on an association between tin and diabetes. Tin is commonly used in coatings for cans and containers, and in electrical, construction, and transportation. Environmental exposure to tin occurs through food, consumer products, and ambient air. One recent study in the US general population found that urinary tin was positively associated with diabetes prevalence.[37] Experimental research suggested the potential role of tin in glucose tolerance and insulin resistance through induction of hepatic inflammation and excess hepatic fat accumulation.[38] In pancreatic β cells, tin was demonstrated to interfere with glucose-induced insulin secretion, due to its inhibitory effect on the cellular calcium response in triggering exocytosis of insulin granules.[39]

Our data did not provide evidence to suggest an association between cadmium and diabetes. Previous studies concerning cadmium exposure and diabetes have yielded inconsistent results.[4 40 41] It is notable that cigarette smoking was less prevalent in our study population of midlife women compared with participants investigated in previous studies. Cigarette smoking has been found to be a major source of cadmium exposure[1] and has been associated with an increased risk of developing diabetes by triggering free radicals, increasing inflammation, oxidative stress and dyslipidemia, and directly damaging β cells.[42] However, no significant association between urinary cadmium concentration and diabetes was observed in never smokers, former smokers, or current smokers when we stratified our analysis by smoking status (data not shown). Further investigations aimed at confirming the association and explaining the inconsistency between populations are warranted.

In previous studies in US adults, urinary cobalt, molybdenum, uranium and tungsten have been positively associated with prevalence of diabetes.[5] Urinary barium has been associated with higher odds of impaired fasting glucose,[43] and urinary nickel has been associated with higher odds of prevalent diabetes, higher fasting glucose, hemoglobin A1c (HbA1c), insulin levels, and increased insulin resistance.[44] A large longitudinal study in US young adults suggested that people with high mercury exposure in young adulthood may have an elevated risk of diabetes and decreased β cell function later in life.[45] On the contrary, mercury exposure was not associated with diabetes risk in both the Health Professionals Follow-up Study and the Nurses' Health Study, the two other large longitudinal studies in US adults.[46] In a recent longitudinal study of Chinese senior adults, plasma antimony was inversely associated with diabetes incidence.[47] Our study did not provide enough evidence to suggest associations of urinary barium, beryllium, cobalt, cesium, mercury, manganese, molybdenum, nickel, antimony, uranium, and tungsten with diabetes.

Metal mixtures

Metals are widely dispersed in the environment and people could be exposed to a myriad of metals simultaneously throughout their lifetime. In this study, we identified two clusters of women with distinct metal concentration profiles, suggesting different exposure patterns to mixtures of metals in the environment. Our previous study using the same clustering approach reported significant differences in sociodemographic, lifestyle, and dietary characteristics between women with different exposure profiles.[1] In the present study, higher overall exposure to metal mixtures was associated with an increased risk of diabetes after adjustment for all these factors, suggesting a potential role of exposure to metal mixtures in diabetes. Notably, each exposure pattern showed homogeneous distributions of individual

metals (standardized concentrations). No patterns had particularly high or low concentrations of specific metals including arsenic, lead, and zinc, of which associations with diabetes were identified individually. This indicates that there may be other components of metal mixtures distinct from arsenic, lead, and zinc that affect diabetes risk but may not be adequately captured by the single-pollutant approach possibly due to relatively small or non-linear effects. It should be acknowledged that the associations between the exposure to metal mixtures represented by k-means clusters and diabetes risk do not provide an insight into which metals were responsible for these associations or allow for dose–response characterization. Ultimately, future research adopting advanced statistical approaches is needed to quantify the diabetogenic impact of exposure to metal mixtures with high degrees of correlation while disentangling the potential low-dose, non-linear effects, and metal–metal interactions.

Strengths and limitations

The primary strength of our study is that diabetes events, as well as other potential confounding factors including sociodemographic factors, lifestyle factors, and metabolic quantitative traits, were assessed annually or biannually over 16 years of follow-up. The prospective design minimized the possibility of reverse causation. The ethnically diverse population as well as comparable metal concentrations in the SWAN cohort compared with women of the same age in the US general population also increase the generalizability of our findings.[1] Another advantage is that we systematically examined a suite of 20 metals in urine samples with high-quality laboratory methods. To the best of our knowledge, the associations between most of the metals included in our study and diabetes have never been investigated in a prospective cohort study.

Our study also has several limitations. First, metals included in the current analysis have very different half-lives in the human body. Urinary concentrations of metals with short half-lives such as arsenic mainly reflect recent exposures.[21] In contrast, metals such as cadmium are not rapidly excreted and have half-lives of years to decades. Therefore, diabetes risk is likely impacted by metal exposures over time periods longer than a few days, and information on the temporal variability of urinary metal concentrations, especially for those with short half-lives, is needed to characterize cumulative metal exposures. Second, we measured all metal concentrations in urine, and urinary concentrations may not unanimously reflect exposure levels because they are influenced by renal clearance. We acknowledge that information on renal function is not available in SWAN, although renal clearance is considered relatively stable in this age group.[48] Third, in our study, only total arsenic concentration was measured in urine sample, and data on arsenic speciation were not available. Exposure to inorganic arsenic has been associated with increased risk of diabetes. In contrast, organic arsenic is generally considered to have low toxicity and a small impact on risk of diabetes.[21 49] Arsenic metabolites may also influence diabetes, as shown in a recent prospective cohort study where a lower proportion of urinary MMA relative to urinary DMA was associated with an increased incidence of diabetes.[50] In future studies, arsenic speciation will be critical to providing a better understanding of arsenic exposures and associated health risks. Fourth, in this study, urinary zinc was adjusted for dietary intake of zinc and zinc supplements in the regression analysis to better capture renal clearance and excretion of zinc. However, the dietary intake of other essential metals was not measured, and we were unable to distinguish between the metals from dietary sources (or other external sources) and the metals from internal sources. Fifth, the use of fasting glucose to determine incident diabetes may have missed some cases who would have been considered to have diabetes based on other tests such as HbA1c and oral glucose tolerance test. However, the use of self-reported physician diagnosis and antidiabetic medication use in diabetes ascertainment reduce the possibility of misclassification. Finally, our results may be subject to selection bias at enrollment into the SWAN-MPS for selective attrition during follow-up. To minimize the possibility of bias in effect estimates, we assigned weights to participants at each follow-up visit using an IPW approach.

In conclusion, this prospective cohort study provides evidence of positive associations of urinary concentrations of arsenic and lead, increased urinary excretion of zinc, as well as a high overall exposure to metal mixtures with the risk of diabetes among midlife women. Our findings may have important public health implications as increasing and widespread exposure to environmental toxicants and their mixtures may be a key contributor to the worldwide epidemics of type 2 diabetes. Our findings also provide impetus to further investigate the underlying mechanisms by which metals and their mixtures may influence risk of diabetes.

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ORCID iDs
Xin Wang http://orcid.org/0000-0002-0851-6605
Sung Kyun Park http://orcid.org/0000-0001-9981-6250

REFERENCES

1 Wang X, Mukherjee B, Batterman S, *et al*. Urinary metals and metal mixtures in midlife women: the study of women's health across the nation (Swan). *Int J Hyg Environ Health* 2019;222:778–89.
2 Hanna-Attisha M, LaChance J, Sadler RC, *et al*. Elevated blood lead levels in children associated with the Flint drinking water crisis: a spatial analysis of risk and public health response. *Am J Public Health* 2016;106:283–90.
3 Maull EA, Ahsan H, Edwards J, *et al*. Evaluation of the association between arsenic and diabetes: a national toxicology program workshop review. *Environ Health Perspect* 2012;120:1658–70.
4 Li Y, Zhang Y, Wang W, *et al*. Association of urinary cadmium with risk of diabetes: a meta-analysis. *Environ Sci Pollut Res Int* 2017;24:10083–90.
5 Menke A, Guallar E, Cowie CC. Metals in urine and diabetes in U.S. adults. *Diabetes* 2016;65:164–70.
6 Ercal N, Gurer-Orhan H, Aykin-Burns N. Toxic metals and oxidative stress Part I: mechanisms involved in metal-induced oxidative damage. *Curr Top Med Chem* 2001;1:529–39.
7 Lu T-H, Su C-C, Chen Y-W, *et al*. Arsenic induces pancreatic β-cell apoptosis via the oxidative stress-regulated mitochondria-dependent and endoplasmic reticulum stress-triggered signaling pathways. *Toxicol Lett* 2011;201:15–26.
8 Patra RC, Rautray AK, Swarup D. Oxidative stress in lead and cadmium toxicity and its amelioration. *Vet Med Int* 2011;2011:1–9.
9 Han JC, Park SY, Hah BG, *et al*. Cadmium induces impaired glucose tolerance in rat by down-regulating GLUT4 expression in adipocytes. *Arch Biochem Biophys* 2003;413:213–20.
10 Abdul KSM, Jayasinghe SS, Chandana EPS, *et al*. Arsenic and human health effects: a review. *Environ Toxicol Pharmacol* 2015;40:828–46.
11 Jansen J, Karges W, Rink L. Zinc and diabetes--clinical links and molecular mechanisms. *J Nutr Biochem* 2009;20:399–417.
12 Wang X, Mukherjee B, Park SK. Associations of cumulative exposure to heavy metal mixtures with obesity and its comorbidities among U.S. adults in NHANES 2003-2014. *Environ Int* 2018;121:683–94.
13 Sowers MF, Crawford SL, Sternfeld B, *et al*. SWAN: a multi-center, multi-ethnic, community-based cohort study of women and the menopausal transition. In: Lobo RA, Kelsey J, Marcus R, *et al*, eds. *Menopause: biology and pathobiology*. Academic Press, 2000: 175–88.
14 Ding N, Harlow SD, Batterman S, *et al*. Longitudinal trends in perfluoroalkyl and polyfluoroalkyl substances among multiethnic midlife women from 1999 to 2011: the study of women's health across the nation. *Environ Int* 2020;135:105381.
15 CDC. *Laboratory procedure manual, Multi-Element in urine. NHANES 2011-2012*, 2012.
16 Sternfeld B, Cauley J, Harlow S, *et al*. Assessment of physical activity with a single global question in a large, multiethnic sample of midlife women. *Am J Epidemiol* 2000;152:678–87.
17 Benjamini Y, Hochberg Y. Controlling the false discovery rate: a practical and powerful approach to multiple testing. *J R Stat Soc Ser B* 1995;57:289–300.
18 Hernán MA. The hazards of hazard ratios. *Epidemiology* 2010;21:13–15.
19 Jones MR, Tellez-Plaza M, Vaidya D, *et al*. Estimation of inorganic arsenic exposure in populations with frequent seafood intake: evidence from MESA and NHANES. *Am J Epidemiol* 2016;184:590–602.
20 Wang W, Xie Z, Lin Y, *et al*. Association of inorganic arsenic exposure with type 2 diabetes mellitus: a meta-analysis. *J Epidemiol Community Health* 2014;68:176–84.
21 Navas-Acien A, Silbergeld EK, Pastor-Barriuso R, *et al*. Arsenic exposure and prevalence of type 2 diabetes in US adults. *JAMA* 2008;300:814.
22 Gribble MO, Howard BV, Umans JG, *et al*. Arsenic exposure, diabetes prevalence, and diabetes control in the strong heart study. *Am J Epidemiol* 2012;176:865–74.
23 Walton FS, Harmon AW, Paul DS, *et al*. Inhibition of insulin-dependent glucose uptake by trivalent arsenicals: possible mechanism of arsenic-induced diabetes. *Toxicol Appl Pharmacol* 2004;198:424–33.
24 Petrick JS, Jagadish B, Mash EA, *et al*. Monomethylarsonous acid (MMA(III)) and arsenite: LD(50) in hamsters and in vitro inhibition of pyruvate dehydrogenase. *Chem Res Toxicol* 2001;14:651–6.
25 Wang X, Kim D, Tucker KL, *et al*. Effect of dietary sodium and potassium intake on the mobilization of bone lead among middle-aged and older men: the Veterans Affairs normative aging study. *Nutrients* 2019;11:2750.
26 Wang X, Ding N, Tucker KL, *et al*. A Western diet pattern is associated with higher concentrations of blood and bone lead among middle-aged and elderly men. *J Nutr* 2017;147:1374–83.
27 Ding N, Wang X, Tucker KL, *et al*. Dietary patterns, bone lead and incident coronary heart disease among middle-aged to elderly men. *Environ Res* 2019;168:222–9.
28 Hernandez-Avila M, Villalpando CG, Palazuelos E, *et al*. Determinants of blood lead levels across the menopausal transition. *Arch Environ Health* 2000;55:355–60.
29 Afridi HI, Kazi TG, Brabazon D, *et al*. Comparative metal distribution in scalp hair of Pakistani and Irish referents and diabetes mellitus patients. *Clin Chim Acta* 2013;415:207–14.
30 Nagaraj G, Sukumar A, Nandlal B, *et al*. Tooth element levels indicating exposure profiles in diabetic and hypertensive subjects from Mysore, India. *Biol Trace Elem Res* 2009;131:255–62.
31 Moon S-S, lead Aof. Association of lead, mercury and cadmium with diabetes in the Korean population: the Korea National health and nutrition examination survey (KNHANES) 2009-2010. *Diabet Med* 2013;30:e143–8.
32 Zhai H, Chen C, Wang N, *et al*. Blood lead level is associated with non-alcoholic fatty liver disease in the Yangtze River delta region of China in the context of rapid urbanization. *Environ Health* 2017;16:93.

33 Leff T, Stemmer P, Tyrrell J, *et al*. Diabetes and exposure to environmental lead (Pb). *Toxics* 2018;6:54.
34 Vashum KP, McEvoy M, Shi Z, *et al*. Is dietary zinc protective for type 2 diabetes? Results from the Australian longitudinal study on women's health. *BMC Endocr Disord* 2013;13:40.
35 Maret W, Sandstead HH. Zinc requirements and the risks and benefits of zinc supplementation. *J Trace Elem Med Biol* 2006;20:3–18.
36 Chausmer AB, Zinc CAB. Zinc, insulin and diabetes. *J Am Coll Nutr* 1998;17:109–15.
37 Liu B, Sun Y, Lehmler H-J, *et al*. Association between urinary tin concentration and diabetes in nationally representative sample of US adults. *J Diabetes* 2018;10:977–83.
38 Bertuloso BD, Podratz PL, Merlo E, *et al*. Tributyltin chloride leads to adiposity and impairs metabolic functions in the rat liver and pancreas. *Toxicol Lett* 2015;235:45–59.
39 Miura Y, Matsui H. Triphenyltin impairs a protein kinase A (PKA)-dependent increase of cytosolic Na+ and Ca2+ and PKA-independent increase of cytosolic Ca2+ associated with insulin secretion in hamster pancreatic beta-cells. *Toxicol Appl Pharmacol* 2006;216:363–72.
40 Barregard L, Bergström G, Fagerberg B. Cadmium exposure in relation to insulin production, insulin sensitivity and type 2 diabetes: a cross-sectional and prospective study in women. *Environ Res* 2013;121:104–9.
41 Swaddiwudhipong W, Limpatanachote P, Mahasakpan P, *et al*. Progress in cadmium-related health effects in persons with high environmental exposure in northwestern Thailand: a five-year follow-up. *Environ Res* 2012;112:194–8.
42 Śliwińska-Mossoń M, Milnerowicz H. The impact of smoking on the development of diabetes and its complications. *Diab Vasc Dis Res* 2017;14:265–76.
43 Feng W, Cui X, Liu B, *et al*. Association of urinary metal profiles with altered glucose levels and diabetes risk: a population-based study in China. *PLoS One* 2015;10:e0123742.
44 Liu G, Sun L, Pan A, *et al*. Nickel exposure is associated with the prevalence of type 2 diabetes in Chinese adults. *Int J Epidemiol* 2015;44:240–8.
45 He K, Xun P, Liu K, *et al*. Mercury exposure in young adulthood and incidence of diabetes later in life: the cardia trace element study. *Diabetes Care* 2013;36:1584–9.
46 Mozaffarian D, Shi P, Morris JS, *et al*. Methylmercury exposure and incident diabetes in U.S. men and women in two prospective cohorts. *Diabetes Care* 2013;36:3578–84.
47 Yuan Y, Xiao Y, Yu Y, *et al*. Associations of multiple plasma metals with incident type 2 diabetes in Chinese adults: the Dongfeng-Tongji cohort. *Environ Pollut* 2018;237:917–25.
48 Murphy D, McCulloch CE, Lin F, *et al*. Trends in prevalence of chronic kidney disease in the United States. *Ann Intern Med* 2016;165:473–81.
49 Thayer KA, Heindel JJ, Bucher JR, *et al*. Role of environmental chemicals in diabetes and obesity: a national toxicology program workshop review. *Environ Health Perspect* 2012;120:779–89.
50 Kuo C-C, Howard BV, Umans JG, *et al*. Arsenic exposure, arsenic metabolism, and incident diabetes in the strong heart study. *Diabetes Care* 2015;38:dc141641–7.