


ORIGINAL ARTICLE

Cardiovascular autonomic neuropathy in adolescents and young adults with type 1 and type 2 diabetes: The SEARCH for Diabetes in Youth Cohort Study

Mamta Jaiswal¹  | Jasmin Divers² | Elaine M Urbina³ | Dana Dabelea⁴ | Ronny A Bell⁵ | David J Pettitt⁶ | Giuseppina Imperatore⁷ | Catherine Pihoker⁸ | Lawrence M Dolan⁹ | Angela D Liese¹⁰ | Santica Marcovina¹¹ | Barbara Linder¹² | Eva L Feldman¹ | Rodica Pop-Busui¹³ | for the SEARCH for Diabetes in Youth Study Group

¹Department of Neurology, University of Michigan, Ann Arbor, Michigan

²Department of Biostatistical Sciences, Wake Forest School of Medicine, Winston-Salem, North Carolina

³Department of Preventive Cardiology, Cincinnati Children's Hospital Medical Center, Cincinnati, Ohio

⁴Department of Epidemiology, Colorado School of Public Health, Aurora, Colorado

⁵Department of Epidemiology and Prevention, Wake Forest School of Medicine, Winston-Salem, North Carolina

⁶Santa Barbara, California

⁷Division of Diabetes Translation, Centers of Disease Control and Prevention, Atlanta, Georgia

⁸Department of Pediatrics, University of Washington, Seattle, Washington

⁹Division of Endocrinology, Children's Hospital Medical Center, Cincinnati, Ohio

¹⁰Department of Epidemiology and Biostatistics and Center for Research in Nutrition and Health Disparities, Arnold School of Public Health, University of South Carolina, Columbia, South Carolina

¹¹Northwest Lipid Research Laboratory, University of Washington, Seattle, Washington

¹²Division of Diabetes, Endocrinology and Metabolic Diseases, National Institute of Diabetes and Digestive and Kidney Diseases, Bethesda, Maryland

¹³Division of Metabolism, Endocrinology and Diabetes, Department of Internal Medicine, University of Michigan, Ann Arbor, Michigan

Correspondence

Dr Mamta Jaiswal, MBBS, PhD, Division of

Objective: To estimate the prevalence of and risk factors for cardiovascular autonomic neuropathy (CAN) in adolescents and young adults with type 1 and type 2 diabetes enrolled in the SEARCH for Diabetes in Youth Study.

Methods: The study included 1646 subjects with type 1 diabetes (age 18 ± 4 years, diabetes duration 8 ± 2 years, HbA1c $9.1 \pm 1.9\%$, 76% non-Hispanic Whites) and 252 with type 2 diabetes (age 22 ± 4 years, diabetes duration 8 ± 2 years, HbA1c $9.2 \pm 3.0\%$, 45% non-Hispanic Blacks). Cross-sectional and longitudinal risk factors were assessed at baseline and follow-up visits. Area under the curve (AUC) was used to assess the longitudinal glycemic exposure and cardiovascular risk factors. CAN was assessed by time and frequency domain indices of heart rate variability (HRV). CAN was defined as the presence of ≥ 3 of 5 abnormal HRV indices.

Results: The prevalence of CAN was 12% in adolescents and young adults with type 1 diabetes and 17% in those with type 2 diabetes. Poor long-term glycemic control (AUC HbA1c), high blood pressure, and elevated triglyceride levels were correlates of CAN in subjects with type 1 diabetes. In those with type 2 diabetes, CAN was associated with elevated triglycerides and increased urinary albumin excretion.

Conclusions: The prevalence of CAN in this multiethnic cohort of adolescents and young adults with type 1 and type 2 diabetes are comparable to those reported in adults with diabetes. Sub-optimal glycemic control and elevated triglycerides were the modifiable risk factors associated with CAN.

KEYWORDS

cardiovascular autonomic neuropathy, heart rate variability, prevalence, type 1 and type 2 diabetes

Metabolism, Endocrinology and Diabetes,
University of Michigan, 5300 Brehm Tower,
1000 Wall Street, Ann Arbor, MI 48105.
Email: jaiswalm@med.umich.edu

Funding information

National Institute of Diabetes and Digestive
and Kidney Diseases, Grant/Award number:
PA numbers 00097, DP-05-069, and DP-10-
001; Centers for Disease Control and
Prevention, Grant/Award number: PA numbers
00097, DP-05-069, and DP-10-001

1 | INTRODUCTION

Cardiovascular autonomic neuropathy (CAN) is a serious complication of diabetes affecting the autonomic nerves innervating the heart and blood vessels, with subsequent sympathovagal imbalance and impact on heart rate regulation and cardiac performance.¹⁻³ Although asymptomatic in earlier stages, CAN has been shown to be an independent predictor of cardiovascular disease (CVD) mortality risk,^{4,5} silent myocardial ischemia⁶ and/or major CVD events,⁷ cardiac remodeling and left ventricular dysfunction,⁸ and progression of diabetic nephropathy and chronic kidney disease.^{9,10} Yet, CAN is one of the least recognized complications of diabetes, especially in youth.

Reduction in the heart rate variability (HRV) parameters is the earliest manifestations of CAN. Evaluation of HRV with time and frequency domain indices are non-invasive methods to assess the presence and severity of CAN.² Several small cross-sectional studies in various pediatric populations, using diverse definitions, have reported CAN prevalence estimates between 18% and 75%.¹¹⁻¹⁵ In one of the largest pediatric epidemiological studies assessing the burden of diabetes-related complications in an Australian cohort of adolescents with diabetes, Eppens et al found strikingly high prevalence of autonomic neuropathy (using pupillometry) in adolescents with type 1 and type 2 diabetes (61% and 57%, respectively).¹⁵

Apart from the SEARCH study, there have been no systematic efforts to assess the burden of neuropathy, including CAN, in adolescents and young adults with diabetes in the United States. We previously reported that youth with type 1 diabetes have reduced HRV as compared to age-matched healthy controls enrolled in the SEARCH CVD study.¹⁶ Although, we have recently reported the age-adjusted prevalence of several diabetic complications (including CAN) in the SEARCH participants as part of the SEARCH Cohort Study,¹⁷ the specific objective of this study was to examine the prevalence of CAN more closely by age group, diabetes duration, gender and race/ethnicity. Moreover, we were specifically interested in examining the cross-sectional and longitudinal risk factors (since diagnosis of diabetes to present) in adolescents and young adults with type 1 and type 2 diabetes to better understand the underlying risk factors and the pathological processes that drive CAN in this young cohort.

The overall objectives of the current study were: (1) to estimate the prevalence of CAN in a large, ethnically diverse cohort of adolescents and young adults with type 1 and type 2 diabetes enrolled in

the SEARCH for Diabetes in Youth Study by age, gender, diabetes duration and race/ethnicity; and (2) to identify the cross-sectional and longitudinal risk factors for CAN.

2 | METHODS

2.1 | SEARCH for Diabetes in Youth Study

SEARCH for Diabetes in Youth Study is a prospective cohort study following children and adolescents of diverse racial and ethnic backgrounds diagnosed with diabetes at less than 20 years of age in the United States.¹⁷ SEARCH participants are incident cases of diabetes identified at 4 geographically defined populations in Ohio, Washington, South Carolina, and Colorado, from health plan enrollees in California, and from Indian Health Service beneficiaries from American Indian populations in Arizona and New Mexico.

2.2 | Study population

Adolescents and young adults with diabetes diagnosed at <20 years of age were identified from a population-derived incident registry network at 5 US sites by the SEARCH for Diabetes in Youth Registry Study.¹⁷ Cases with newly diagnosed type 1 or type 2 diabetes in 2002-2006 or 2008, who completed a SEARCH baseline examination (on average 9.3 ± 6.4 months from diagnosis) and had at least 5 years of diabetes duration between 2011 and 2015, were recruited into the SEARCH Cohort Study (2011-2015) (on average at 7.9 ± 1.9 years from diagnosis) (Figure A1). Although the parent SEARCH Cohort Study enrolled 2777 individuals, we excluded children <10 years of age ($n = 134$) at the cohort visit, those with no diabetes antibody measures for the etiological definition of diabetes ($n = 440$), and those with incomplete neuropathy assessment ($n = 305$), which reduced the analytic sample size to 1898 individuals (Figure A2).

Prior to protocol implementation, local institutional review board approval was obtained for each center. Written informed consent was obtained from participants 18 years and older, while assent with parental written informed consent was obtained for participants younger than 18 years.

2.3 | Baseline and cohort visits

The SEARCH baseline and cohort visits included a participant survey; measurement of height, weight, waist circumference, blood pressure;

and blood and urine collection. Race and ethnicity were self-reported and categorized as non-Hispanic White (NHW), non-Hispanic Black (NHB), Hispanic, Asian or Pacific Islander, and Other. Current cigarette smoking was defined as having smoked cigarettes on ≥ 1 of the 30 days preceding the visit. Individuals who had tried smoking or smoked regularly (at least 1 cigarette every day for 30 days) but were not current smokers were considered former smokers. Subjects who had never smoked a whole cigarette were considered non-smokers.

Waist circumference was measured using the natural waist location¹⁷ and divided by height in centimeters to calculate the waist-to-height ratio (WHR). Body mass index (BMI) was defined as weight (kg) divided by height² (m). For participants <20 years of age, the Centers for Disease Control and Prevention (CDC)-derived BMI z20 scores were used; for those ≥ 20 years the observed mean and SD were used to standardize their BMI z20 values.

Resting systolic blood pressure (SBP) and diastolic blood pressure (DBP) were measured 3 times using an aneroid sphygmomanometer while the participants were seated for at least 5 minutes, and the average of the 3 measurements was taken.

A blood draw occurred after an 8-hour overnight fast, and medications, including short-acting insulin, were withheld the morning of the visit. Blood samples were obtained under conditions of metabolic stability, as defined by no episodes of diabetic ketoacidosis in the prior month. Specimens were processed locally at the sites and shipped within 24 hours to the central laboratory (Northwest Lipid Metabolism and Diabetes Research Laboratories, University of Washington), where they were analyzed for measurement of high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), triglycerides, and glycated hemoglobin (HbA1c) as previously described.¹⁷ Urinary albumin and creatinine levels were assessed on a random spot urine sample to evaluate renal function using the albumin-to-creatinine ratio (ACR). The accuracy of HbA1c data was monitored by participation in the National Glycohemoglobin Standardization Program, and the accuracy and consistency of lipid data were monitored regularly by comparing results obtained by enzymatic methods with those obtained by CDC reference methods (CDC Reference Laboratory).¹⁸

In addition to a SEARCH baseline and a cohort visit, 57% of participants ($n = 1082$) had one or more intermediate visits (at 1, 2, or 5 years after the baseline visit) at which the same cardio-metabolic risk factors were measured, including HbA1c, lipids, waist circumference, WHR, and BMI. The assay of biological samples has remained consistent over time.

2.4 | Diabetes type

Diabetes type was defined using an etiological classification developed by SEARCH¹⁹ based on diabetes autoantibodies (glutamate decarboxylase-2 [GAD-65], insulinoma-associated-2 antibodies [IA-2A], and zinc-T8 autoantibody) and estimated insulin sensitivity score (validated equation including waist circumference, HbA1c, and triglyceride levels) at the baseline visit.¹⁹ Type 1 diabetes was defined as at least 1 positive antibody, regardless of insulin sensitivity, or no positive antibodies and insulin sensitivity (score > 8.15). Type 2 diabetes was defined as negative antibodies and insulin resistance (score < 8.15).¹⁹

2.5 | Assessment of CAN

CAN was assessed by HRV testing using the SphygmoCor (AtCor, Philadelphia, PA). SEARCH staff from each center was centrally trained and certified to perform the HRV test. The HRV tests were performed under standardized conditions that included overnight fasting, avoidance of caffeine and tobacco products for 8 hours before the test, and withholding prescriptions and over-the-counter medicines (except for basal insulin) until testing was completed. Participants underwent a 5-minute continuous electrocardiogram (ECG) recording while supine after a 10-minute rest. All traces were reviewed and analyzed to ensure that R-waves were adequately identified from artifacts and ectopic beats. The term "NN interval" is used instead of RR interval of the ECG to emphasize that the processed beats are normal sinus rhythm (ie, every QRS complex preceded a P-wave). We analyzed the following time- and frequency-domain HRV parameters from the SphgmoCor device: standard deviation of NN interval (SDNN), root mean square difference of the successive NN interval (RMSSD), high frequency (HF) power, low frequency (LF) power, and LF:HF ratio. SDNN is a measure of overall HRV, while the RMSSD and HF power represent the parasympathetic component of the autonomic system and LF power the sympathetic component. HRV test was considered abnormal if the values were below the 5th percentile observed in 206 age- and sex-matched healthy controls (age 10-28 years, 54% females) from the SEARCH CVD study.¹⁶ CAN, as our primary outcome measure, was defined as the presence of ≥ 3 abnormal HRV indices.

2.6 | Statistical analysis

2.6.1 | Cross-sectional data

Anthropometric, demographic, and metabolic data collected at the cohort visit were used to compare the characteristics of adolescents and young adults with and without CAN stratified by diabetes type.

Student's *t* test and Wilcoxon 2-sample tests were used to compare the distribution of normally and non-normally (triglycerides, SDNN, RMSSD) distributed continuous variables, respectively, and the χ^2 test was used for categorical variables separately for type 1 and type 2 diabetes participants. Fisher's exact test was used whenever a cell count for a particular test was less than 5. The data were presented as mean \pm SD for normally distributed variables and as median (interquartile range, IQR) for non-normally distributed variables such as triglycerides and log transformation was done for others such as ACR.

The prevalence of CAN was estimated overall and based on the age at diagnosis (≥ 10 and <10 years) and duration of diabetes (5, 6-10, and >10 years) separately for persons with type 1 or type 2 diabetes.

2.6.2 | Longitudinal data

In addition to the data collected at the baseline and cohort visit, the area under the curve (AUC) was computed to summarize the longitudinal trajectory of HbA1c and other continuous variables, such as lipids, blood pressure, and BMI collected over time (at the baseline,

1-, 2-, or 5-year follow-up and cohort visits), adjusting for the time interval between the first and last measurement.

To assess the association of long-term glycemic control with CAN, logistic regression models treating the presence of CAN as the outcome were fitted separately for participants with type 1 and type 2 diabetes. These models were adjusted for potential confounders (collected at current SEARCH cohort visit) such as age and sex (Model 2), BMI (Model 3), blood pressure (Model 4), triglycerides (Model 5), and ACR (Model 6). A fully adjusted model that included all of these variables as covariates was also fitted (Model 7). Models were stratified by diabetes type to limit confounding effects of age and adiposity. Diagnostic tests were performed to ensure that modeling assumptions were satisfied. The data were analyzed using SAS 9.4 (SAS Institute, Cary, NC).

3 | RESULTS

Characteristics of adolescents and young adults with type 1 ($n = 1646$) and type 2 diabetes ($n = 252$) stratified by their CAN status are shown in Table 1. The prevalence of CAN was 12% in adolescents and young adults with type 1 diabetes, and 17% among those with type 2 diabetes (Table 1). Subjects with type 1 diabetes and CAN were older (mean age: 19 ± 4 vs 18 ± 4 years), more likely to have developed diabetes at age 10 or older, and had a larger waist circumference (81 ± 12 vs 78 ± 12 cm), higher blood pressure (SBP: 110 ± 11 vs 106 ± 11 and DBP 72 ± 9 vs 69 ± 9 mm Hg), poorer glycemic control (HbA1c: $9.6 \pm 2.1\%$ vs $9.1 \pm 1.8\%$), and elevated levels of triglycerides (median [IQR] 82 [61 120] vs 74 [55 104] mg/dL) than those without CAN (all $P < .05$; Table 1). Males with type 1 diabetes had a higher prevalence of CAN as compared to females (15% vs 10%, $P = .001$). Subjects with type 2 diabetes and CAN had higher DBP (80 ± 12 vs 75 ± 10 mm Hg) and elevated triglyceride levels (median [IQR] 151 [102 254] vs 110 [78 183] mg/dL) and ACR (3.2 ± 1.5 vs 2.7 ± 1.7 mg/g) compared to those without CAN (all $P < .05$; Table 1). Hispanic and NHW subjects with type 2 diabetes had a higher prevalence of CAN (29% and 27%, respectively) as compared to NHB (7%) and other minority groups (12%) ($P = .001$).

The AUC for cardio-metabolic risk factors for CAN is depicted in Table 2. Long-term poor glycemic control, summarized as AUC for HbA1c, triglyceride levels (AUC triglycerides), and blood pressure (AUC SBP and AUC DBP), were significantly higher among type 1 diabetes subjects with CAN as compared to those without CAN. Only higher triglycerides over time were significantly associated with CAN among subjects with type 2 diabetes (Table 2).

Table 3 summarizes the results from the multiple logistic regression analyses for the association between longitudinal glycemic control (AUC HbA1c as the independent variable) and CAN (dependent variable) adjusted sequentially for covariates. Long-term poor glycemic control (AUC HbA1c) was associated with CAN independent of age, sex, blood pressure, BMI, triglyceride levels, and ACR in subjects with type 1 diabetes, but not in those with type 2 diabetes (Table 3).

Since nearly 32% ($n = 879$) of the 2777 SEARCH participants were excluded from the analysis dataset due to various reasons

(age < 10 years, missing etiological definition of diabetes type, incomplete CAN assessment), we examined whether there were any significant differences in the anthropometric, demographic, and metabolic characteristics of the participants who were excluded vs included in this dataset that could potentially affect the prevalence estimates. Individuals excluded ($n = 879$) from the analysis were more likely to have a longer duration of diabetes (9 vs 8 years, $P < .001$) and were younger at the time of diabetes diagnosis (9 vs 11 years, $P < .001$), as compared to those included in the analytic sample ($n = 1898$) (Table A1).

4 | DISCUSSION

This study evaluating a large, multiethnic cohort of adolescents and young adults in the United States found high prevalence of CAN in subjects with type 1 and type 2 diabetes. Poor glycemic control and higher triglyceride levels over time were consistently associated with CAN. This is the first population-derived study in the United States that carefully characterized differences in the cross-sectional and longitudinal risk factors for CAN in a large, racially/ethnically diverse cohort of adolescents and young adults with type 1 and type 2 diabetes.

Placing our findings in perspective, in a meta-analysis including 3943 participants from 19 studies, the prevalence of subclinical CAN defined by either cardiovascular reflex tests or baroreflex sensitivity in young people (age < 24 years) with type 1 diabetes varied between 16% and 75% depending on the outcomes reported.²⁰ For instance, in this meta-analysis pooled prevalence of CAN defined by measures of HRV was 21%, and ranged from 4% to 11% if CAN was defined by a single cardiovascular reflex test such as deep breathing or Valsalva.²⁰ In contrast, a relatively small study that assessed HRV (LF and HF powers) and included only 20 pediatric patients with type 1 diabetes (mean diabetes duration: 7 years, mean HbA1c: 8.2%) reported prevalence rates for CAN as high as 75%.¹¹ The prevalence estimates for CAN we found in the SEARCH cohort are lower compared with those reported by Eppens et al in an Australian cohort that included 1433 youth with type 1 and 68 type 2 diabetes who found that 61% of youth with type 1 diabetes and 57% of those with type 2 diabetes had evidence of autonomic neuropathy.¹⁵ However, in that cohort autonomic neuropathy was defined as an abnormal pupillometry test (assessed by measuring the pupil size before and 3 seconds after a light stimulus was delivered using an infrared pupillometer).¹⁵ The difference in the method of assessment (pupillometry vs HRV testing) likely explains the higher prevalence of autonomic neuropathy in the Australian cohort, in spite of that cohort being younger, with shorter diabetes duration and better glycemic control compared with the SEARCH cohort.¹⁵ Although pupillometry was considered in the past a simple non-invasive test to assess parasympathetic autonomic function, it has not been widely used in the research setting because of the lack of standardization in the techniques employed, and the lack of validation studies. Thus, differences in outcome definitions, type of autonomic dysfunction evaluated, and the methods of assessments are the reasons accounting for the high

TABLE 1 Clinical characteristics of adolescents and young adults with type 1 and type 2 diabetes stratified by cardiovascular autonomic neuropathy status at the current SEARCH Cohort research visit (2011-2015)

Variable	Type 1 diabetes (n = 1646)			Type 2 diabetes (n = 252)		
	No CAN	CAN	P-value	No CAN	CAN	P-value
N (%)	1443 (88%)	203 (12%)		209 (83%)	43 (17%)	
Age (years)	18 ± 4	19 ± 4	<.0001	22 ± 4	22 ± 3	.99
Age at diagnosis (years)	10 ± 4	11 ± 3	<.0001	14.1 ± 3	14 ± 3	.82
Age at diagnosis ≥10 years	723 (84%)	137 (16%)	<.0001	200 (83%)	40 (17%)	.45
Age at diagnosis <10 years	720 (92%)	66 (8%)		9 (75%)	3 (25%)	
Diabetes duration (years)	7.8 ± 1.8	7.9 ± 1.9	.44	7.9 ± 1.9	7.8 ± 2.0	.82
Diabetes duration (years)						
5-10	1233 (88%)	167 (12%)	.24	174 (82%)	38 (18%)	.49
≥10	210 (85%)	36 (15%)		35 (87%)	5 (13%)	
Sex						
Female	746 (90%)	79 (10%)	.001	141 (83%)	28 (17%)	.77
Male	697 (85%)	124 (15%)		68 (82%)	15 (18%)	
Race/ethnicity						
Non-Hispanic White	1089 (87%)	158 (13%)	.63	48 (73%)	18 (27%)	.001
Asian/Pacific Islander	24 (92%)	2 (8%)		3 (100%)	0 (0%)	
Non-Hispanic Black	143 (91%)	14 (9%)		105 (93%)	8 (7%)	
Hispanic	175 (86%)	28 (14%)		37 (71%)	15 (29%)	
Others	12 (87%)	1 (13%)		16 (88%)	2 (12%)	
Smoking						
Never	967 (89%)	122 (11%)	.15	80 (84%)	15 (16%)	.76
Former	259 (85%)	46 (15%)		61 (80%)	15 (20%)	
Current	188 (86%)	30 (14%)		63 (84%)	12 (16%)	
Oral diabetes medication						
Yes	68 (87.2%)	10 (12.8%)	.40	109 (82.6%)	23 (17.4%)	.90
No	1372 (87.7%)	192 (12.3%)		99 (83.2%)	20 (16.8%)	
Insulin dose	56.84 ± 29.09	64.22 ± 30.97	.0007	66.19 ± 38.48	76.35 ± 36.26	.20
Insulin dose, per kg	0.85 ± 0.40	0.90 ± 0.40	.06	0.73 ± 0.39	0.86 ± 0.36	.19
Waist circumference (cm)	78 ± 12	81 ± 12	.00011	104 ± 19	111 ± 20	.06
Waist-to-height ratio	0.5 ± 0.1	0.5 ± 0.1	.48	0.7 ± 0.1	0.7 ± 0.1	.33
BMI (kg/m ²)	24 ± 5	25 ± 5	.12	35 ± 9	37 ± 9	.38
SBP (mm Hg)	106 ± 11	110 ± 11	<.0001	118 ± 13	121 ± 17	.36
DBP (mm Hg)	69 ± 9	72 ± 9	<.0001	75 ± 10	80 ± 12	.01
HbA1c (%)	9.1 ± 1.8	9.6 ± 2.1	.001	9.2 ± 3	9.6 ± 2.8	.42
HbA1c (mmol/mol)	75 ± 7	80 ± 8	.001	77 ± 9	80 ± 9	.42
HDL-C (mg/dL)	55 ± 14	54 ± 13	.15	42 ± 12	39 ± 10	.16
LDL-C (mg/dL)	96 ± 28	99 ± 32	.50	106 ± 38	104 ± 37	.88
Triglycerides (mg/dL)	74 (55 104)	82 (61 120)	.005	110 (78 183)	151 (102 254)	.002
CRP (mg/dL)	0.2 ± 0.5	0.4 ± 1.5	.74	0.6 ± 0.8	0.7 ± 0.7	.16
Log ACR (μg/mg)	2.0 ± 0.93	2.1 ± 0.95	.33	2.7 ± 1.7	3.2 ± 1.5	.014

Abbreviations: ACR, albumin:creatinine ratio; BMI, body mass index; CAN, cardiovascular autonomic neuropathy; CRP, C-reactive protein; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; SBP and DBP, systolic and diastolic blood pressure. All data are presented as mean ± SD or n (%) or median (IQR).

variability in the prevalence reported in the few pediatric populations.

Poor glycemic control, longer duration of diabetes, increasing age, microalbuminuria, DBP, and dyslipidemia (lower HDL, increased

triglycerides) are some of the established risk factors for CAN in adults with diabetes.^{1,21-25} This study found that poor glycemic control and hypertriglyceridemia over time were the strongest risk factors associated with CAN in adolescents and young adults with type 1 diabetes.

TABLE 2 Cardio-metabolic risk factor burden over time by cardiovascular autonomic neuropathy status in adolescents and young adults with type 1 and type 2 diabetes

Variable	Type 1 diabetes (n = 1646)			Type 2 diabetes (n = 252)		
	No CAN	CAN	P-value	No CAN	CAN	P-value
AUC BMI z-score	20.6 ± 0.9	20.6 ± 0.9	.75	21.9 ± 0.7	22 ± 0.6	.50
AUC DBP	65.9 ± 7.2	68 ± 7.6	<.001	73.1 ± 6.7	75.6 ± 9.7	.18
AUC SBP	103.5 ± 9	107.5 ± 9	<.001	116.6 ± 9.3	119.7 ± 11.8	.22
AUC HbA1c	8.5 ± 1.3	8.8 ± 1.5	.005	8.4 ± 2.4	8.6 ± 2.3	.65
AUC LDL-C	93.5 ± 22.3	95.1 ± 24.2	.53	104.1 ± 30.7	99.6 ± 26.6	.54
AUC triglycerides	79.2 ± 43.8	92 ± 59.6	.001	149.6 ± 121.7	225.3 ± 221.8	.005
AUC HDL-C	56.1 ± 11.8	54.7 ± 10.6	.10	42.4 ± 10.9	40.1 ± 10.4	.13

Abbreviations: AUC, area under the curve derived from data collected at baseline, 1-, 2-, 5-year follow-up and the current cohort visits; BMI, body mass index; CAN, cardiovascular autonomic neuropathy; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; SBP and DBP, systolic and diastolic blood pressure. All data are presented as mean ± SD.

TABLE 3 Multivariable regression models for association between long-term glycemic control and cardiovascular autonomic neuropathy collected at the current SEARCH Cohort Study (2011-2015)

Dependent variable: CAN Independent variable: AUC A1c adjusted for time between measures	Type 1 diabetes		Type 2 diabetes	
	OR (95% CI)	P-value	OR (95% CI)	P-value
Model 1 = AUC A1c	1.28 (1.11; 1.48)	.0006	1.07 (0.77; 1.48)	.69
Model 2 = Model 1 + age, sex	1.3 (1.13; 1.5)	.0002	1.07 (0.77; 1.49)	.68
Model 3 = Model 1 + BMI	1.29 (1.11; 1.49)	.0006	1.11 (0.79; 1.56)	.54
Model 4 = Model 1 + SBP, DBP	1.26 (1.09; 1.47)	.002	1 (0.71; 1.41)	1.00
Model 5 = Model 1 + Trig	1.22 (1.05; 1.43)	.010	0.93 (0.65; 1.33)	.69
Model 6 = Model 1 + ACR	1.26 (1.07; 1.47)	.005	1.27 (0.86; 1.88)	.22
Model 7 = Model 1 + age, sex, BMI, SBP, DBP, Trig, ACR	1.24 (1.04; 1.46)	.01	1.08 (0.69; 1.7)	.72

Abbreviations: ACR, albumin:creatinine ratio; AUC, area under the curve; BMI, body mass index; CAN, cardiovascular autonomic neuropathy; CI, confidence interval; OR, Odds ratio; SBP and DBP, systolic and diastolic blood pressure; Trig, triglycerides.

There is ample biologic plausibility and evidence for the causal role of hyperglycemia in the development and progression of chronic complications, including CAN.^{1,26-29} Hyperglycemia induces abnormal signaling of the autonomic neurons via accumulation of advanced glycation end products and microangiopathy, causing ischemic atrophy of autonomic nerve fibers innervating cardiac and vascular tissues.³⁰ In this study, glucose control was quite poor in adolescents and young adults with type 1 or type 2 diabetes, with a mean HbA1c of ~9%, far exceeding the target HbA1c (<7.5%) recommended by the American Diabetes Association.³¹ These data further confirm that there is an urgent need for efforts focused at improving glycemic control among adolescents and young adults with diabetes to mitigate the elevated risk of the adverse outcomes associated with CAN and its downstream consequences, including increased CVD risk.

Dyslipidemia has also been implicated in the pathogenesis of diabetic neuropathy in a non-glucocentric paradigm involving linked metabolic and inflammatory insults that trigger neurodegeneration.³²⁻³⁶ However, there is a close link between glucose and lipid metabolism, as hypertriglyceridemia and reduced HDL commonly occur in poorly controlled type 1 and type 2 diabetes.³³ Hyperglycemia and dyslipidemia are also known as pro-inflammatory triggers to the neurodegenerative processes.²³ In a prior analysis of youth participating in the SEARCH CVD study, we found an atherogenic lipid profile in youth with type 1 diabetes and reduced HRV as compared to age-matched healthy controls.¹⁶

The findings from this study have important clinical implications. We observed that youth and young adults with type 1 and type 2 diabetes have evidence of CAN, as documented by changes in HRV at a mean diabetes duration of 8 years. Emerging epidemiological evidence also suggests that CAN is associated with increased arterial stiffness in adolescents³⁷ and adults³⁸ with type 1 diabetes, and thus could have an additive effect on the risk of future cardiovascular events, which occur earlier and with poorer prognosis in individuals with diabetes compared with the general population.^{4,39,40}

Considering that SEARCH participants are a representative cohort of US youth with diabetes and have suboptimal glycemic control (mean HbA1c well above the American Diabetes Association recommended target of <7.5%),¹⁹ these data provide evidence that screening for CAN, as is recommended for adults, may be beneficial in adolescents and young adults,¹ especially in those individuals who have additional risk factors associated with CAN. Recent recommendations from the American Heart Association call for a multifactorial approach to mitigate the increased CVD risk in youth with diabetes.⁴¹ Thus, targeting poor glycemic control and dyslipidemia in adolescents and young adults with diabetes as early as possible, which is also in line with current standard of care in diabetes,³¹ could help mitigate the increased CVD risk in this young population.^{28,36}

The large sample size, multiethnic composition of the cohort, use of a non-invasive, simple, validated instrument to assess CAN, and evaluation of the longitudinal and cross-sectional risk factors are

among the strengths of our study. The limited power to examine the association between long-term glycemic control and CAN among persons with type 2 diabetes (despite similar levels of HbA1c to those with type 1 diabetes) could be due to a comparatively small sample size (although the association was in the same direction as that of the type 1 diabetes group) is one of the limitations of our study. The lack of longitudinal measures of CAN is also a limitation of our study, although a subset of this cohort (2002-2012 incidence cases ≥ 10 years of age with least 5 years of duration of diabetes) will be re-evaluated for CAN as part of the next phase of SEARCH (2016-2020). Finally, although the SEARCH Cohort Study is drawn from population-based registries of youth with diabetes, those excluded from the analytic sample were more likely to be older at time of diagnosis and had a longer duration of diabetes. Each of these variables is associated with increased prevalence of CAN and may influence our estimates of CAN prevalence in youth and young adults with diabetes.

Overall, the current data support the fact that good glycemic control and better approaches to manage dyslipidemia are needed to prevent the development and worsening of CAN in this young population. Given the independent risk of CAN for cardiovascular events and death, health care providers should motivate pediatric patients to reach and maintain optimal glucose control and better management of other risk factors including triglycerides to ameliorate the risk of premature cardiovascular events.

ACKNOWLEDGEMENTS

The SEARCH for Diabetes in Youth Study is indebted to the many youth and their families, and their health care providers, whose participation made this study possible. Grant Support: SEARCH for Diabetes in Youth is funded by the Centers for Disease Control and Prevention (PA numbers 00097, DP-05-069, and DP-10-001) and supported by the National Institute of Diabetes and Digestive and Kidney Diseases. Site Contract Numbers: Kaiser Permanente Southern California (U48/CCU919219, U01 DP000246, and U18DP002714), University of Colorado Denver (U48/CCU819241-3, U01 DP000247, and U18DP000247-06A1), Children's Hospital Medical Center (Cincinnati) (U48/CCU519239, U01 DP000248, and U18DP002709), University of North Carolina at Chapel Hill (U48/CCU419249, U01 DP000254, and U18DP002708), University of Washington School of Medicine (U58/CCU019235-4, U01 DP000244, and U18DP002710-01), Wake Forest University School of Medicine (U48/CCU919219, U01 DP000250, and 200-2010-35171). The authors wish to acknowledge the involvement of the South Carolina Clinical & Translational Research Institute at the Medical University of South Carolina, NIH/National Center for Advancing Translational Sciences (NCATS) grant number UL1 TR000062; the Seattle Children's Hospital and the University of Washington, NIH/NCATS grant number UL1 TR00423; the University of Colorado Pediatric Clinical and Translational Research Center, NIH/NCATS grant number UL1 TR000154; the Barbara Davis Center at the University of Colorado at Denver (DERC NIH grant number P30 DK57516); the University of Cincinnati, NIH/NCATS grant number UL1 TR000077; and the Children with Medical Handicaps program managed by the

Ohio Department of Health. The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the Centers for Disease Control and Prevention and the National Institute of Diabetes and Digestive and Kidney Diseases.

Conflict of interest

All authors report no conflict of interest.

Author contributions

M.J., J.D., R.P.B., and D.D. designed the study and analysis plan. M.-J. wrote the manuscript. J.D. performed the analysis and provided critical input to the manuscript. R.P.B. reviewed data, provided input in study design, and critically reviewed the manuscript for intellectual content. E.M.U., D.D., R.A.B., D.J.P., G.I., C.P., L.M.D., A.D.L., S.M., B.L., and E.L.F. critically reviewed the manuscript and provided input. J.D. is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

ORCID

Mamta Jaiswal  <http://orcid.org/0000-0003-1727-6225>

REFERENCES

1. Pop-Busui R, Boulton AJ, Feldman EL, et al. Diabetic neuropathy: a position statement by the American Diabetes Association. *Diabetes Care*. 2017;40(1):136-154.
2. Spallone V, Ziegler D, Freeman R, et al. Toronto consensus panel on diabetic neuropathy. Cardiovascular autonomic neuropathy in diabetes: clinical impact, assessment, diagnosis, and management. *Diabetes Metab Res Rev*. 2011;27(7):639-653.
3. Pop-Busui R. What do we know and we do not know about cardiovascular autonomic neuropathy in diabetes. *J Cardiovasc Transl Res*. 2011;5(4):463-478.
4. Pop-Busui R, Evans GW, Gerstein HC, et al. Effects of cardiac autonomic dysfunction on mortality risk in the action to control cardiovascular risk in diabetes (ACCORD) trial. *Diabetes Care*. 2010;33(7):1578-1584.
5. Maser RE, Mitchell BD, Vinik AI, Freeman R. The association between cardiovascular autonomic neuropathy and mortality in individuals with diabetes: a meta-analysis. *Diabetes Care*. 2003;26(6):1895-1901.
6. Young LH, Wackers FJ, Chyun DA, et al. Cardiac outcomes after screening for asymptomatic coronary artery disease in patients with type 2 diabetes: the DIAD study—a randomized controlled trial. *JAMA*. 2009;301(15):1547-1555.
7. Pop-Busui R, Braffett BH, Zinman B, et al. Cardiovascular autonomic neuropathy and cardiovascular outcomes in the diabetes control and complications trial/epidemiology of diabetes interventions and complications (DCCT/EDIC) study. *Diabetes Care*. 2017;40(1):94-100.
8. Pop-Busui R, Cleary PA, Braffett BH, et al. Association between cardiovascular autonomic neuropathy and left ventricular dysfunction: DCCT/EDIC study (diabetes control and complications trial/ epidemiology of diabetes interventions and complications). *J Am Coll Cardiol*. 2013;61(4):447-454.
9. Orlov S, Cherney DZ, Pop-Busui R, et al. Cardiac autonomic neuropathy and early progressive renal decline in patients with nonmacroalbuminuric type 1 diabetes. *Clin J Am Soc Nephrol*. 2015;10(7):1136-1144.
10. Wheelock KM, Jaiswal M, Martin CL, et al. Cardiovascular autonomic neuropathy associates with nephropathy lesions in American Indians with type 2 diabetes. *J Diabetes Complications*. 2016;30(5):873-879.

11. Boysen A, Lewin MAG, Hecker W, Leichter HE, Uhlemann F. Autonomic function testing in children and adolescents with diabetes mellitus. *Pediatr Diabetes*. 2007;8:261-264.
12. Dalla Pozza R, Bechtold S, Bonfig W, et al. Impaired short-term blood pressure regulation and autonomic dysbalance in children with type 1 diabetes mellitus. *Diabetologia*. 2007;50:2417-2423.
13. Mohsin F, Craig ME, Cusumano J, et al. Discordant trends in microvascular complications in adolescents with type 1 diabetes from 1990 to 2002. *Diabetes Care*. 2005;28:1974-1980.
14. Riihimaa PH, Suominen K, Knip M, Tapanainen P, Tolonen U. Cardiovascular autonomic reactivity is decreased in adolescents with type 1 diabetes. *Diabet Med*. 2002;19:932-938.
15. Eppens MC, Craig ME, Cusumano J, et al. Prevalence of diabetes complications in adolescents with type 2 compared with type 1 diabetes. *Diabetes Care*. 2006;29(6):1300-1306.
16. Jaiswal M, Urbina EM, Wadwa RP, et al. Reduced heart rate variability among youth with type 1 diabetes: the SEARCH CVD study. *Diabetes Care*. 2013;36(1):157-162.
17. Dabelea D, Stafford JM, Mayer-Davis EJ, et al. Association of type 1 diabetes vs type 2 diabetes diagnosed during childhood and adolescence with complications during teenage years and young adulthood. *JAMA*. 2017;317(8):825-835.
18. Myers GL, Kimberly MM, Waymack PW, Smith SJ, Cooper GR, Sampson EJ. A reference method laboratory network for cholesterol: a model for standardization and improvement of clinical laboratory measurements. *Clin Chem*. 2000;46:1762-1772.
19. Dabelea D, Pihoker C, Talton JW, et al. Etiological approach to characterization of diabetes type: the SEARCH for diabetes in youth study. *Diabetes Care*. 2011;34(7):1628-1633.
20. Tang M, Donaghue KC, Cho YH, Craig ME. Autonomic neuropathy in young people with type 1 diabetes: a systematic review. *Pediatr Diabetes*. 2013;14:239-248.
21. Pop-Busui R, Low PA, Waberski BH, et al. Effects of prior intensive insulin therapy on cardiac autonomic nervous system function in type 1 diabetes mellitus: the diabetes control and complications trial/epidemiology of diabetes interventions and complications study (DCCT/EDIC). *Circulation*. 2009;119:2886-2893.
22. Kempler P, Tesfaye S, Chaturvedi N, et al. Autonomic neuropathy is associated with increased cardiovascular risk factors: the EURODIAB IDDM Complications Study. *Diabet Med*. 2002;19:900-909.
23. Gottsäter A, Ahmed M, Fernlund P, Sundkvist G. Autonomic neuropathy in type 2 diabetic patients is associated with hyperinsulinaemia and hypertriglyceridaemia. *Diabet Med*. 1999;16:49-54.
24. Witte DR, Tesfaye S, Chaturvedi N, Eaton S, Kempler P, Fuller JH. Risk factors for cardiac autonomic neuropathy in type 1 diabetes mellitus. *Diabetologia*. 2005;48:164-171.
25. Larsen JR, Sjøholm H, Berg TJ, et al. Eighteen years of fair glycemic control preserves cardiac autonomic function in type 1 diabetes. *Diabetes Care*. 2004;27:963-966.
26. The Diabetes Control and Complications Trial Research Group. The effect of intensive therapy of diabetes on the development and progression of long-term complications in insulin-dependent diabetes mellitus. *N Engl J Med*. 1993;329:977-986.
27. Nathan DM, Lachin J, Cleary P, et al. Intensive diabetes therapy and carotid intima-media thickness in type 1 diabetes mellitus. *N Engl J Med*. 2003;348(23):2294-2303.
28. Gaede P, Vedel P, Larsen N, Jensen GV, Parving HH, Pedersen O. Multifactorial intervention and cardiovascular disease in patients with type 2 diabetes. *N Engl J Med*. 2003;348(5):383-393.
29. Ang L, Jaiswal M, Martin C, Pop-Busui R. Glucose control and diabetic neuropathy: lessons from recent large clinical trials. *Curr Diab Rep*. 2014;14(9):528.
30. Prasad A, Bekker P, Tsimikas S. Advanced glycation end products and diabetic cardiovascular disease. *Cardiol Rev*. 2012;20:177-183.
31. Standards of Medical Care in Diabetes—2016. *Diabetes Care*. 2016;39(suppl 1):S86-S89.
32. Vincent AM, Hinder LM, Pop-Busui R, Feldman EL. Hyperlipidemia: a new therapeutic target for diabetic neuropathy. *J Peripher Nerv Syst*. 2009;14:257-267.
33. Wiggin TD, Sullivan KA, Pop-Busui R, Amato A, Sima AA, Feldman EL. Elevated triglycerides correlate with progression of diabetic neuropathy. *Diabetes*. 2009;58(7):1634-1640.
34. Hur J, Sullivan KA, Pande M, et al. The identification of gene expression profiles associated with progression of human diabetic neuropathy. *Brain*. 2011;134(11):3222-3235.
35. Ginsberg HN. Diabetic dyslipidemia: basic mechanisms underlying the common hypertriglyceridemia and low HDL cholesterol levels. *Diabetes*. 1996;45(3):S27-S30.
36. Fried LF, Forrest KY, Ellis D, et al. Lipid modulation in insulin-dependent mellitus: effect on microvascular outcomes. *J Diabetes Complications*. 2001;15(3):113-119.
37. Jaiswal M, Urbina EM, Wadwa RP, et al. Reduced heart rate variability is associated with increased arterial stiffness in youth with type 1 diabetes: the SEARCH CVD Study. *Diabetes Care*. 2013;36(8):2351-2358.
38. Prince CT, Secrest AM, Mackey RH, Arena VC, Kingsley LA, Orchard TJ. Cardiovascular autonomic neuropathy, HDL cholesterol, and smoking correlate with arterial stiffness markers determined 18 years later in type 1 diabetes. *Diabetes Care*. 2010;33(3):652-657.
39. Liao D, Carnethon M, Evans G, Cascio W, Heiss G. Lower heart rate variability is associated with the development of coronary heart disease in diabetics: the ARIC Study. *Am J Epidemiol*. 2000;151:S35.
40. Valensi P, Sachs RN, Harfouche B, et al. Predictive value of cardiac autonomic neuropathy in diabetic patients with or without silent myocardial ischemia. *Diabetes Care*. 2001;24:339-343.
41. Maahs DM, Daniels SR, de Ferranti SD, et al. Cardiovascular disease risk factors in youth with diabetes mellitus: a scientific statement from the American Heart Association. *Circulation*. 2014;130(17):1532-1558.

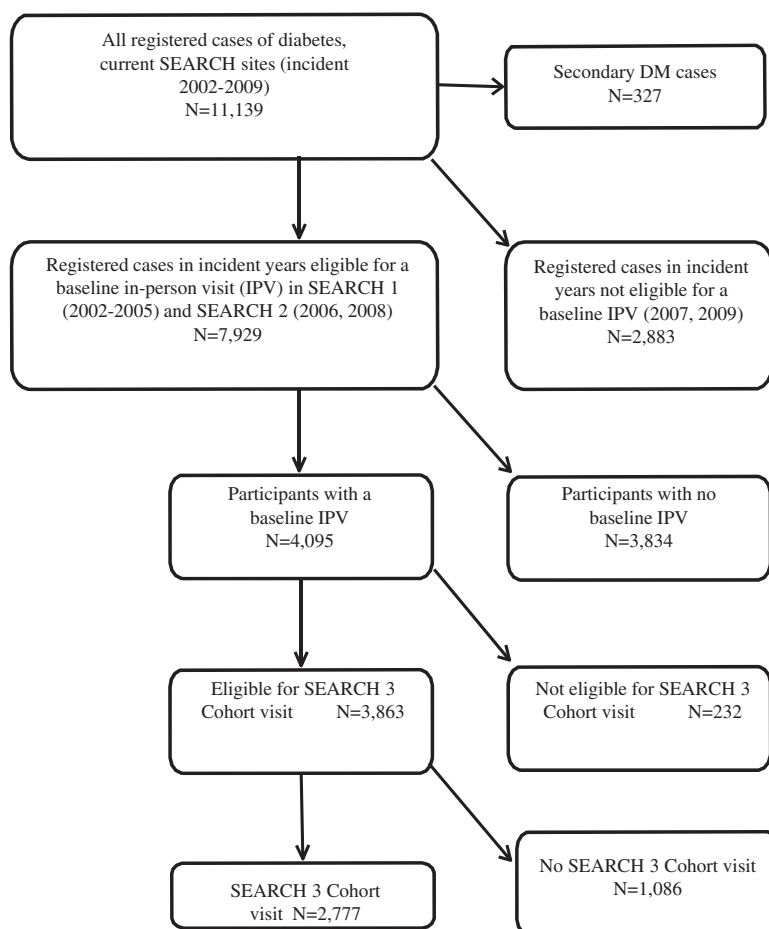
How to cite this article: Jaiswal M, Divers J, Urbina EM, et al. Cardiovascular autonomic neuropathy in adolescents and young adults with type 1 and type 2 diabetes: The SEARCH for Diabetes in Youth Cohort Study. *Pediatr Diabetes*. 2018;19:680-689. <https://doi.org/10.1111/pedi.12633>

APPENDIX

TABLE A1 Characteristics of the participants excluded and included in the cardiovascular autonomic neuropathy analysis dataset at the current SEARCH Cohort visit (2011-2015)

Variable	Excluded (n = 879)	Included (n = 1898)	P-value
Age at cohort visit (years)	18 ± 5	19 ± 4	<.001
Age at diagnosis (years)	9 ± 5	11 ± 4	<.001
Sex (female, %)	53	52	.74
Diabetes duration (months)	104 ± 24	95 ± 23	<.001
Race/ethnicity			.080
Non-Hispanic White (%)	63.8	69.3	
Non-Hispanic Black (%)	18.7	14.1	
Hispanic (%)	13.7	13.4	
Asian/Pacific Islander (%)	1.9	1.5	
Native Americans (%)	1.5	1.3	
Other (%)	0.4	0.3	
BMI z-score	0.8 ± 1.2	0.7 ± 1.1	.29
SBP (mm Hg)	102 ± 14	102 ± 13	.62
DBP (mm Hg)	64 ± 11	64 ± 10	.86
HbA1c (%)	9.2 ± 2.1	9.2 ± 2.1	.90
Triglycerides (mg/dL)	102 ± 100	105 ± 107	.43
LDL-C (mg/dL)	97 ± 30	98 ± 30	.49
HDL-C (mg/dL)	55 ± 15	53 ± 14	.06

Abbreviations: BMI, body mass index; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; SPB and DBP, systolic and diastolic blood pressure. All data are presented as mean ± SD or n (%).

**FIGURE A1** SEARCH 3 Consort diagram

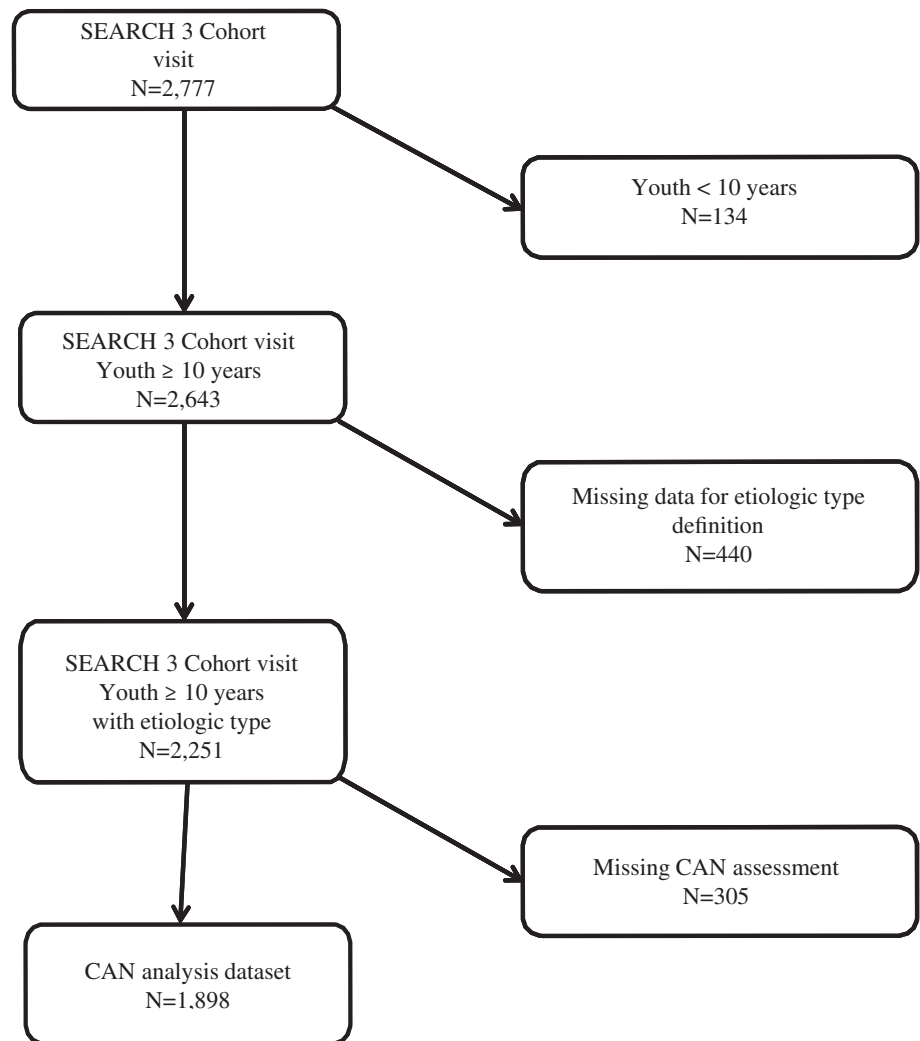


FIGURE A2 Exclusion and inclusion criteria