

Low 25-hydroxyvitamin D₂ and 25-hydroxyvitamin D₃ levels are independently associated with macroalbuminuria, but not with retinopathy and macrovascular disease in type 1 diabetes: the EURODIAB Prospective Complications Study

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-Electronic Supplementary Material -

Additional file 1: Figure S1 Concentration of 25(OH)D₂ (A) and 25(OH)D₃ (B) according to latitude of the study centre. City, country code and, between brackets, latitude of the study centre, and the number of individuals per study centre are displayed on the x-axis. Boxes indicate medians and interquartile ranges and whiskers represent the minimum and maximum values.

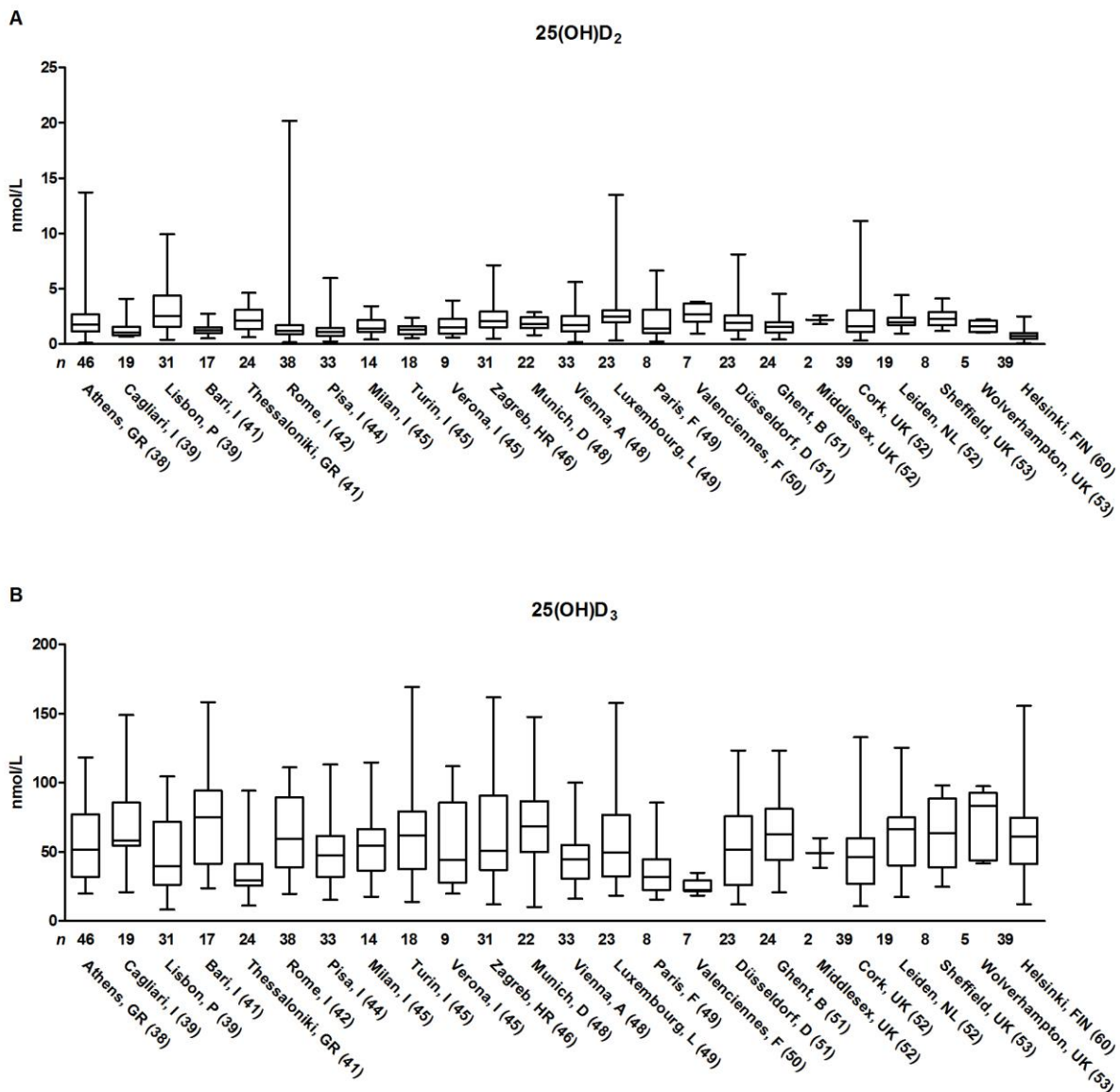


Figure S1

Table S1: Associations between 25-hydroxyvitamin D₂ tertiles and prevalent albuminuria, retinopathy and cardiovascular disease

	Model	25-hydroxyvitamin D ₂ tertiles					<i>p-trend</i>	
		T1		T2		T3		
		OR	OR	95% CI	OR	95% CI		
Microalbuminuria (n=83)	1	1.0 (reference)	0.87	0.47; 1.61	0.74	0.40; 1.37	0.34	
	2	1.0 (reference)	0.86	0.41; 1.80	0.53	0.25; 1.12	0.09	
	3	1.0 (reference)	0.92	0.44; 1.94	0.55	0.26; 1.17	0.10	
	4	1.0 (reference)	0.94	0.44; 2.00	0.54	0.25; 1.15	0.09	
Macroalbuminuria (n=124)	1	1.0 (reference)	0.29	0.18; 0.49	0.12	0.07; 0.23	<0.001	
	2	1.0 (reference)	0.39	0.19; 0.80	0.13	0.06; 0.30	<0.001	
	3	1.0 (reference)	0.41	0.20; 0.86	0.14	0.06; 0.31	<0.001	
	4	1.0 (reference)	0.42	0.20; 0.86	0.13	0.06; 0.30	<0.001	
Non-proliferative retinopathy (n=146)	1	1.0 (reference)	0.67	0.39; 1.13	0.60	0.35; 1.02	0.07	
	2	1.0 (reference)	0.70	0.35; 1.39	0.95	0.48; 1.89	0.95	
Proliferative retinopathy (n=152)	1	1.0 (reference)	0.46	0.27; 0.77	0.51	0.31; 0.85	0.009	
	2	1.0 (reference)	0.73	0.34; 1.55	1.60	0.76; 3.37	0.16	
CVD (n=131)	1	1.0 (reference)	1.88	1.11; 3.20	1.45	0.85; 2.49	0.18	
	2	1.0 (reference)	2.22	1.23; 4.01	1.64	0.89; 3.05	0.13	

OR, odds ratio indicates the odds of prevalent albuminuria, retinopathy and CVD, for 25-hydroxyvitamin D₂ tertile (T) 2 and T3 vs. T1. Model 1: adjusted for age and sex; Model 2: model 1 + BMI, smoking (never, ex, current), HbA_{1c}, total-HDL-cholesterol-ratio, systolic blood pressure, use of antihypertensive medication, eGFR, physical activity (0, ≤sex-specific median, >sex-specific median), alcohol intake (0, ≤70 g/wk, >70 g/wk), and prevalent CVD, albuminuria and retinopathy, as appropriate; Model 3: model 2 + low-grade inflammation score; Model 4: model 2 + endothelial dysfunction score.

Table S2: Associations between 25-hydroxyvitamin D₃ tertiles and prevalent albuminuria, retinopathy and CVD

	Model	month-specific 25-hydroxyvitamin D ₃ tertiles						<i>p-trend</i>
		T1		T2		T3		
		OR		OR	95% CI	OR	95% CI	
Microalbuminuria (n=83)	1	1.0	(reference)	1.26	0.91; 1.71	0.65	0.35; 1.22	0.16
	2	1.0	(reference)	1.81	0.89; 3.69	0.80	0.38; 1.72	0.56
	3	1.0	(reference)	2.02	0.97; 4.18	0.88	0.41; 1.91	0.73
	4	1.0	(reference)	1.80	0.88; 3.67	0.81	0.38; 1.75	0.60
Macroalbuminuria (n=124)	1	1.0	(reference)	0.61	0.47; 0.79	0.29	0.17; 0.51	<0.001
	2	1.0	(reference)	0.97	0.48; 1.94	0.32	0.14; 0.69	0.005
	3	1.0	(reference)	1.06	0.52; 2.16	0.34	0.15; 0.75	0.008
	4	1.0	(reference)	0.97	0.48; 1.94	0.32	0.15; 0.72	0.006
Non-proliferative retinopathy (n=146)	1	1.0	(reference)	0.69	0.41; 1.17	0.43	0.25; 0.74	0.002
	2	1.0	(reference)	0.77	0.40; 1.49	0.71	0.36; 1.39	0.32
Proliferative retinopathy (n=152)	1	1.0	(reference)	0.70	0.42; 1.17	0.50	0.30; 0.84	0.008
	2	1.0	(reference)	0.93	0.45; 1.91	0.95	0.46; 1.98	0.91
CVD (n=131)	1	1.0	(reference)	1.07	0.65; 1.77	0.77	0.46; 1.31	0.36
	2	1.0	(reference)	1.23	0.71; 2.12	0.94	0.52; 1.70	0.86

OR, odds ratio indicates the odds of prevalent albuminuria, retinopathy and CVD for month-specific 25-hydroxyvitamin D₃ tertile (T) 2 and T3 vs. T1. Model 1: adjusted for age and sex; Model 2: model 1 + BMI, smoking (never, ex, current), HbA_{1c}, total-HDL-cholesterol-ratio, systolic blood pressure, use of antihypertensive medication, eGFR, physical activity (0, ≤sex-specific median, >sex-specific median), alcohol intake (0, ≤70 g/wk, >70 g/wk), and prevalent albuminuria, retinopathy and CVD, as appropriate; Model 3: model 2 + low-grade inflammation score; Model 4: model 2 + endothelial dysfunction score.

Table S3: Associations between 25-hydroxyvitamin D₂ levels and markers of low-grade inflammation and endothelial dysfunction

	Model	25-hydroxyvitamin D ₂ per 1 nmol/L			25-hydroxyvitamin D ₂ tertiles						
		β	95% CI	p	T1		T2		T3		p-trend
					β	β	95% CI	β	95% CI		
LGI score	1	-0.050	-0.094; -0.006	0.027	0.0 (reference)	-0.307	-0.500; -0.115	-0.325	-0.517; -0.132	0.001	
	2	-0.026	-0.068; 0.015	0.22	0.0 (reference)	-0.239	-0.420; -0.058	-0.190	-0.371; -0.009	0.042	
Ln(CRP)	1	-0.015	-0.061; 0.031	0.52	0.0 (reference)	-0.056	-0.256; 0.144	-0.066	-0.266; 0.134	0.52	
	2	-0.005	-0.049; 0.039	0.82	0.0 (reference)	-0.078	-0.270; 0.114	-0.028	-0.221; 0.164	0.78	
Ln(IL-6)	1	-0.013	-0.059; 0.034	0.60	0.0 (reference)	-0.211	-0.416; -0.005	-0.151	-0.355; 0.052	0.15	
	2	-0.004	-0.051; 0.043	0.87	0.0 (reference)	-0.181	-0.386; 0.025	-0.093	-0.298; 0.112	0.38	
Ln(TNF-α)	1	-0.081	-0.125; -0.038	<0.001	0.0 (reference)	-0.405	-0.594; -0.216	-0.493	-0.683; -0.303	<0.001	
	2	-0.049	-0.087; -0.010	0.013	0.0 (reference)	-0.264	-0.432; -0.097	-0.294	-0.465; -0.124	0.001	
ED score	1	-0.036	-0.082; 0.011	0.13	0.0 (reference)	-0.456	-0.658; -0.254	-0.355	-0.553; -0.156	0.001	
	2	-0.016	-0.061; 0.028	0.48	0.0 (reference)	-0.363	-0.557; -0.170	-0.234	-0.426; -0.043	0.019	
sE-selectin	1	0.012	-0.035; 0.058	0.62	0.0 (reference)	-0.262	-0.467; -0.058	-0.066	-0.269; 0.136	0.53	
	2	0.009	-0.038; 0.055	0.72	0.0 (reference)	-0.249	-0.450; -0.048	-0.060	-0.260; 0.140	0.58	
sVCAM-1	1	-0.064	-0.111; -0.017	0.008	0.0 (reference)	-0.397	-0.601; -0.193	-0.446	-0.648; -0.244	<0.001	
	2	-0.032	-0.076; 0.012	0.16	0.0 (reference)	-0.276	-0.467; -0.084	-0.279	-0.469; -0.088	0.004	

β, regression coefficient indicates the increase in markers of inflammation and endothelial dysfunction (in SD) per 1 nmol/L higher 25-hydroxyvitamin D₂ (first column) or 25-hydroxyvitamin D₂ tertile (T) 2 and T3 vs. T1 (last three columns). Model 1: adjusted for age, sex and case-control status; Model 2: model 1 + BMI, smoking (never, ex, current), HbA_{1c}, total-HDL-cholesterol-ratio, systolic blood pressure, use of antihypertensive medication, eGFR, physical activity (0, ≤sex-specific median, >sex-specific median), alcohol intake (0, ≤70 g/wk, >70 g/wk). *LGI*, low-grade inflammation; *ED*, endothelial dysfunction.

Table S4: Associations between 25-hydroxyvitamin D₃ levels and markers of inflammation and endothelial dysfunction

	Model	25-hydroxyvitamin D ₃ per 10 nmol/L			month-specific 25-hydroxyvitamin D ₃ tertiles					
		β	95% CI	<i>p</i>	T1	T2		T3		<i>p-trend</i>
					β	β	95% CI	β	95% CI	
LGI score	1	-0.034	-0.066; -0.003	0.032	0.0 (reference)	-0.287	-0.480; -0.093	-0.215	-0.410; -0.020	0.031
	2	-0.019	-0.049; 0.011	0.21	0.0 (reference)	-0.206	-0.387; -0.024	-0.131	-0.317; 0.056	0.17
Ln(CRP)	1	-0.016	-0.048; 0.017	0.35	0.0 (reference)	-0.202	-0.402; -0.002	-0.122	-0.325; 0.081	0.24
	2	-0.010	-0.041; 0.022	0.55	0.0 (reference)	-0.165	-0.357; 0.026	-0.091	-0.288; 0.107	0.37
Ln(IL-6)	1	-0.029	-0.063; 0.004	0.09	0.0 (reference)	-0.275	-0.480; -0.070	-0.249	-0.456; -0.042	0.018
	2	-0.017	-0.051; 0.018	0.35	0.0 (reference)	-0.219	-0.426; -0.012	-0.175	-0.388; 0.037	0.10
Ln(TNF- α)	1	-0.030	-0.066; 0.002	0.07	0.0 (reference)	-0.151	-0.345; 0.044	-0.098	-0.294; 0.098	0.33
	2	-0.016	-0.045; 0.013	0.29	0.0 (reference)	-0.066	-0.238; 0.106	-0.020	-0.197; 0.157	0.83
ED score	1	-0.048	-0.082; -0.015	0.005	0.0 (reference)	-0.205	-0.407; -0.002	-0.272	-0.478; -0.066	0.010
	2	-0.040	-0.073; -0.007	0.017	0.0 (reference)	-0.149	-0.343; 0.046	-0.215	-0.416; -0.015	0.035
sE-selectin	1	-0.022	-0.056; 0.012	0.20	0.0 (reference)	-0.037	-0.242; 0.168	-0.155	-0.363; 0.053	0.14
	2	-0.020	-0.054; 0.014	0.24	0.0 (reference)	-0.002	-0.203; 0.200	-0.128	-0.336; 0.080	0.23
sVCAM-1	1	-0.048	-0.082; -0.014	0.006	0.0 (reference)	-0.258	-0.463; -0.053	-0.238	-0.446; -0.029	0.025
	2	-0.037	-0.069; -0.005	0.024	0.0 (reference)	-0.213	-0.404; -0.021	-0.183	-0.381; 0.015	0.07

β , regression coefficient indicates the increase in markers of inflammation and endothelial dysfunction (in SD) per 10 nmol/L higher 25-hydroxyvitamin D₃ (first column) or month-specific 25-hydroxyvitamin D₃ tertile (T) 2 and T3 vs. T1 (last three columns). Model 1: adjusted for age, sex and case-control status (and, in the analyses with continuous 25-hydroxyvitamin D₃ levels, season); Model 2: model 1 + BMI, smoking (never, ex, current), HbA_{1c}, total-HDL-cholesterol-ratio, systolic blood pressure, use of antihypertensive medication, eGFR, physical activity (0, \leq sex-specific median, $>$ sex-specific median), alcohol intake (0, \leq 70 g/wk, $>$ 70 g/wk).

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