

Supplementary information

Automated measurement method for the determination of vitamin E in plasma lipoprotein classes.

Yuji Hirowatari ^{1*}, Hiroshi Yoshida ^{2*}, Hideo Kurosawa ³, Daisuke Manita ¹, Norio Tada ⁴.

¹ Bioscience Division, TOSOH Corporation, Kanagawa, Japan

² Department of Laboratory Medicine, Jikei University Kashiwa Hospital, Chiba, Japan

³ Clinical Laboratory Department, Inzai General Hospital, Chiba Japan.

⁴ Clinical Medicine Research Institute, Jikei University Kashiwa Hospital, Chiba, Japan

Address of corresponding authors

Yuji Hirowatari, Ph.D.

Bioscience Division, TOSOH CORPORATION

2743-1 Hayakawa, Ayase, Kanagawa 252-1123, Japan

Phone: +81-467-76-9911

FAX: +81-467-76-9932

E-mail: yuuji-hirowatari-bx@tosoh.co.jp

Hiroshi Yoshida, MD, PhD, FAHA, FACP

Professor, Department of Laboratory Medicine,

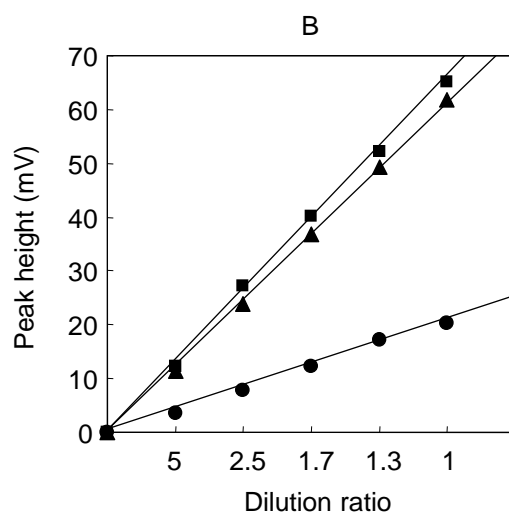
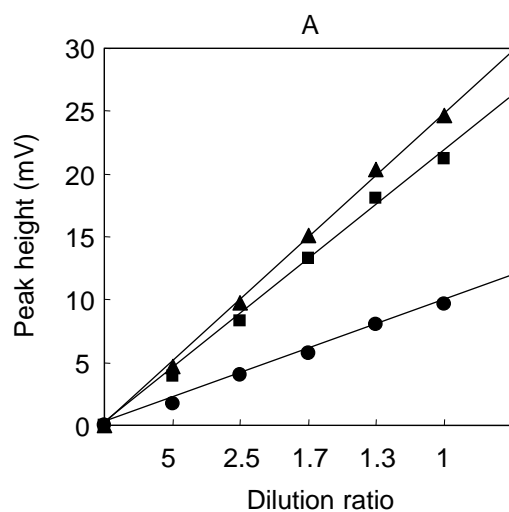
Jikei University Kashiwa Hospital

163-1 Kashiwashita, Kashiwa Chiba 277-8567, Japan

Phone: +81-4-7164-1111 (ext. 2270)

FAX: +81-4-7164-1126

E-mail: hyoshida@jikei.ac.jp



Supplementary Figure S1. Linearity of γ - and α -tocopherol in lipoprotein classes.

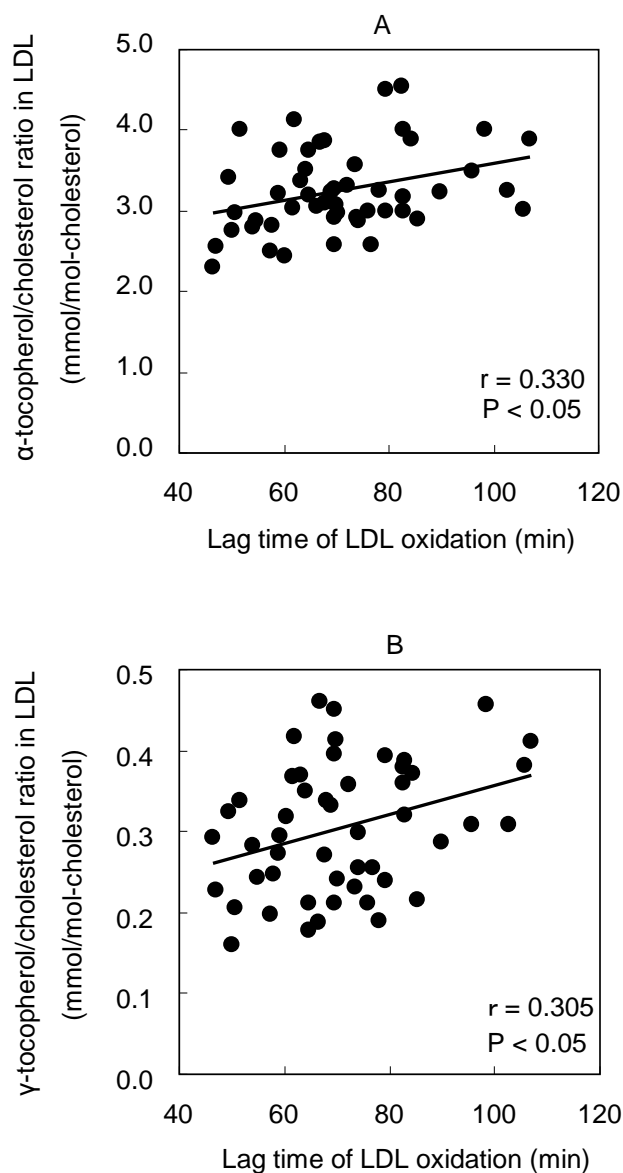
A and B show the data of γ - and α -tocopherol in lipoprotein classes. Tocopherols in HDL, LDL, and VLDL are represented by circles, triangles, and squares, respectively.

Supplementary Table S1. Precision data for tocopherol concentrations in lipoproteins

		Between assay (n=10)			Within assay (n=10)		
		Mean	SD	CV	Mean	SD	CV
		($\mu\text{mol/L}$)	($\mu\text{mol/L}$)	(%)	($\mu\text{mol/L}$)	($\mu\text{mol/L}$)	(%)
Subject 4	γ -tocopherol in HDL	0.74	0.07	9.15	0.78	0.05	5.94
	α -tocopherol in HDL	6.70	0.47	7.00	7.09	0.46	6.53
	γ -tocopherol in LDL	0.68	0.06	9.19	0.70	0.05	7.51
	α -tocopherol in LDL	7.04	0.58	8.21	7.27	0.45	6.23
	γ -tocopherol in VLDL	0.30	0.04	14.73	0.32	0.04	12.84
	α -tocopherol in VLDL	2.52	0.23	8.99	2.69	0.22	8.20
Subject 5	γ -tocopherol in HDL	0.78	0.08	9.73	0.74	0.04	5.54
	α -tocopherol in HDL	5.29	0.50	9.44	5.22	0.26	5.05
	γ -tocopherol in LDL	1.05	0.09	8.79	1.04	0.05	4.73
	α -tocopherol in LDL	8.73	0.69	7.93	9.07	0.57	6.30
	γ -tocopherol in VLDL	0.73	0.07	9.15	0.73	0.05	6.31
	α -tocopherol in VLDL	6.37	0.60	9.39	6.65	0.39	5.84

Supplementary Table S2. Recovery data of tocopherol.

	Original concentration ($\mu\text{mol/L}$)	Added amount ($\mu\text{mol/L}$)	Amount found ($\mu\text{mol/L}$)	Recovery rate (%)
γ -tocopherol in HDL	0.53	0.21	0.20	92
		0.32	0.33	101
		0.43	0.44	102
α -tocopherol in HDL	3.86	2.22	2.01	90
		3.33	3.74	112
		4.44	4.92	111
γ -tocopherol in LDL	0.80	0.52	0.49	93
		0.79	0.77	98
		1.05	1.14	109
α -tocopherol in LDL	9.33	7.27	6.91	95
		10.90	11.68	107
		14.54	16.64	114
γ -tocopherol in VLDL	0.45	0.59	0.54	92
		0.88	0.92	104
		1.18	1.23	105
α -tocopherol in VLDL	5.64	14.81	16.07	108
		22.41	24.10	108
		32.42	32.14	99



Supplementary Figure S2. Correlation between LDL oxidation lag-time and γ - and α -tocopherol/cholesterol ratio in LDL.

Oxidation lag-times of LDL fraction obtained from plasma of 50 subjects by ultracentrifugation are analyzed. The levels of γ - and α -tocopherol/cholesterol ratio in LDL are determined by the new estimated method. The data of 50 subjects are as follows: male/female = 37/13, age = 52.8 ± 13.8 year, TC = 5.06 ± 0.67 mmol/L, TG = 1.26 ± 0.61 mmol/L.

Supplementary Table S3. Comparison of the sum of tocopherols in lipoprotein fractions with the plasma level.

Plasma No.	γ -tocopherol					α -tocopherol						
	HDL ($\mu\text{mol/L}$)	LDL ($\mu\text{mol/L}$)	VLDL ($\mu\text{mol/L}$)	Sum (HDL+LDL+VLDL) ($\mu\text{mol/L}$)	Plasma level ($\mu\text{mol/L}$)	Recovery rate (%)	HDL ($\mu\text{mol/L}$)	LDL ($\mu\text{mol/L}$)	VLDL ($\mu\text{mol/L}$)	Sum (HDL+LDL+VLDL) ($\mu\text{mol/L}$)	Plasma level ($\mu\text{mol/L}$)	Recovery rate (%)
1	1.18	0.93	0.37	2.49	2.84	87	6.18	6.66	2.00	14.83	14.98	99
2	0.90	0.78	0.20	1.88	2.35	80	8.65	9.95	2.34	20.94	22.86	92
3	1.81	1.20	0.35	3.36	4.39	76	9.68	7.34	1.75	18.77	20.55	91
4	0.45	0.79	0.41	1.65	1.73	95	3.47	8.86	4.20	16.53	17.28	96
5	0.93	0.84	0.29	2.05	2.16	95	6.99	10.09	2.67	19.75	20.32	97
6	1.38	0.88	0.28	2.54	3.40	75	11.66	9.43	3.50	24.59	26.71	92
7	1.11	1.02	0.40	2.53	2.82	90	8.84	10.25	2.83	21.93	22.58	97
8	1.13	1.22	0.46	2.81	3.01	93	7.45	10.22	3.21	20.88	21.16	99
9	0.93	1.09	0.43	2.45	3.21	76	7.20	10.59	3.35	21.14	24.67	86
10	0.77	0.89	0.71	2.37	2.90	82	4.61	8.56	5.43	18.60	21.81	85
Mean	1.06	0.96	0.39	2.41	2.88	85	7.47	9.19	3.13	19.79	21.29	93
SD	0.37	0.16	0.14	0.48	0.73	8	2.40	1.33	1.09	2.78	3.38	5