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Serum Thrombomodulin Concentration Is a Warning Marker for Diabetic Complications

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Summary. **Serum thrombomodulin (sTM) concentration, a marker for the endothelium, is increased by endothelial damage or by decreased renal clearance. In this study, we followed up 65 diabetic patients for more than five years to assess the relationship between complications and sTM concentrations. At the beginning, 31 of 65 patients had a high sTM concentration (≧25.9 ng/ml). In this group (the HsTM Group), prevalences of diabetic retinopathy and nephropathy amounted to 14 cases (45%) with 9 cases (29%) at the beginning, and 17 cases (55%) and 7 cases (23%) at the end of follow-up period, respectively. On the other hand, in 34 patients with normal sTM concentrations (the NsTM Group), there were 11 cases (32%) and 4 cases (12%) at the beginning, and 14 cases (41%) and 6 cases (18%) at the end of the period, respectively. There was a negative correlation between sTM and TM clearance (CTM) both for those with and without diabetic nephropathy; the sTM levels with nephropathy were higher than those without nephropathy at the same CTM, suggesting more marked endothelial damage in the patients with nephropathy than those without nephropathy. Furthermore, 11 out of 24 patients in the HsTM Group were diagnosed to have ischemic heart diseases, and 3 patients later died of myocardial infarction. On the other hand, only four out of 31 patients in the NsTM Group had ischemic heart disease.**

In conclusion, sTM concentration seems to be a warning marker for developing diabetic complications, especially coronary heart disease. Increased endothelial damage seems to result in increased sTM levels even in the patients with diabetic nephropathy.

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INTRODUCTION

Thrombomodulin (TM), a thrombin receptor on the endothelial cell, is released to the bloodstream when endothelial cells are damaged. From this view, the measurement of serum thrombomodulin (sTM) seems to reveal the extent of the endothelial cell damage. Many researches have reported that sTM levels are increased in collagen diseases or thrombotic diseases[1,2] in which endothelial cell damage is present. Serum TM is also known to be cleared by the liver and kidney, and sTM levels have been found to increase in patients with renal failure.[3]

In diabetes mellitus, some authors[2,4,5,6] have reported that sTM concentration is increased in diabetic patients and that sTM levels correlate with the severity of diabetic complications including diabetic retinopathy and diabetic nephropathy. Some workers[7] have suggested that increased sTM concentrations in diabetic patients are caused by reduced renal function due to diabetic nephropathy, but whether thrombomodulin clearance is decreased in these patients or not remains to be proven.

This study investigated which diabetic complication is most related to an increased sTM concentration. Whether or not increased sTM is a mere result of diabetic nephropathy was assessed in this five-year follow-up study.

PATIENTS AND METHODS

Sixty-five patients (all aged under 70 years) with diabetes mellitus were involved in the present study. During the follow-up period (1988-1995), serum and urine samples were taken and measured annually or biannually. The serum and urinary TM was measured using an enzyme-linked immunosorbent assay kit (Thrombomodulin "MGCC" EIA) established by Mitsubishi Gas Chemical Co. Ltd. (Tokyo, Japan). In this kit, the normal range of serum TM was 14.5-25.9 U/ml (95% range) and intra- and inter assay error was < 5%. The upper serum creatinin and HbAlc limits were 1.3 mg/dl and 5.4%, respectively. Diabetic retinopathy was diagnosed by ophthalmologists. Evaluation of diabetic nephropathy was performed by albumin excretion rates using overnight urine, and the grading of diabetic nephropathy was classified by Mogensen's classification.[8] Evaluation of ischemic heart disease was performed by an exercise test (ergometer or treadmill) or ^{201}Tl myocardial scintigram (with dipyridamole or exercise) in 55 out of 65 cases. Statistical analysis was performed using the unpaired t-test, with a p-value <0.05 considered significant.

RESULTS

At the beginning (1989-1990) of the follow-up period, sTM concentrations were above the upper limit of the normal range (25.9 U/ml) in 31 of 65 patients (high sTM Group (HsTM)), but normal in the remaining patients (NsTM). The mean age of the HsTM Group was 60.9±6.14 years, which was significantly higher than that of NsTM group (56.6±8.43, $p<0.05$). There was no significant difference in the mean HbAlc levels between the two groups (the HsTM Group 7.04±1.40% and the NsTM Group 6.89±1.20%, respectively) (Table 1).

Data of glycemic controls and grades of complications are also summarized in Table 1. At the beginning of the follow-up period, 14 cases of the HsTM Group of 31 patients (45%) had diabetic retinopathy (6 simple, and 8 proliferative), and the prevalence of diabetic retinopathy increased from 14 (45%) to 17 (55%) during the follow-up period of five years. In the NsTM Group, 11 out of 34 patients (32%) had diabetic retinopathy (9 simple, 2 proliferative), and the prevalence of diabetic retinopathy was increased from 11 (32%) to 14 (41%) during the follow-up period.

In the HsTM Group, 9 had diabetic nephropathy (7 microalbuminuria, and 2 macroalbuminuria) at the beginning of the follow-up and 7 at the end of the follow-up period (4 microalbuminuria, 1 macroalbuminuria, and 2 hemodialysis). Two patients with macroalbuminuria at the beginning underwent hemodialysis therapy during the follow-up period. On the other hand, in the NsTM Group, 4 had diabetic nephropathy (all microalbuminuria) at the beginning and only 6 at the end of the follow-up period (5 microalbuminuria, and 1 macroalbuminuria).

As for coronary heart disease, 11 out of 24 patients examined in the HsTM Group were diagnosed as having ischemic heart diseases, with 3 patients succumbing to mycardial infarction. On the other hand, only four out of 31 patients examined in NsTM group

Table 1. Clinical features of HsTM and NsTM groups

		HsTM	NsTM	
No. of case (M/F)		31 (12/19)	34 (12/22)	
Age (y.o.)		60.9±6.14	56.6±8.43	$p<0.05$
Duration of diabetes (y)		8.6±6.54	6.6±5.30	$p<0.05$
mean HbAlc (%)		7.04±1.40	6.89±1.20	n.s.
Hypertension		13/31 (41.2%)	13/34 (38.2%)	n.s.
Hyperlipidemia		8/31 (25.8%)	12/34 (35.3%)	n.s.
Smoking		8/28 (28.6%)	9/29 (31.0%)	n.s.
Retinopathy	none	17	23	
	simple	6	9	
	proliferative	8	2	
Nephropathy	normoalbuminuria	22	30	
	microalbuminuria	7	4	
	macroalbuminuria	2	0	
Ischemic heart disease		11/24 (45.8%)	6/31 (19.4%)	$p<0.05$

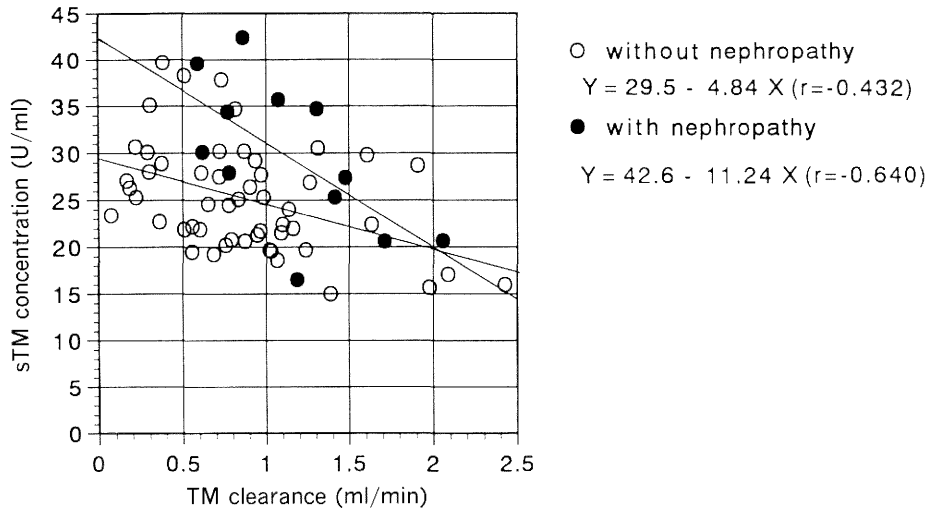

Fig. 1. Correlation between sTM concentration and TM clearance.

were diagnosed as having ischemic heart diseases.

Fig. 1 shows the correlation between sTM and TM clearance (CTM) with and without diabetic nephropathy. There was a negative correlation between sTM and CTM both with and without diabetic nephropathy (with nephropathy, $sTM=42.1-11.2\times CTM$, $r=-0.640$: without nephropathy, $sTM=29.6-5.06\times CTM$, $r=-0.441$); the sTM concentrations with nephropathy were higher than those without nephropathy with the same CTM.

DISCUSSION

In this study, 31 out of 65 diabetic patients were shown to have high sTM levels (the HsTM Group). The mean age of the HsTM Group was significantly higher than that of the NsTM Group. However, it is known that the sTM concentrations in the old healthy men are remained within normal range, although the sTM concentrations increase higher with aging. As the duration of diabetes in the HsTM Group was longer than that of the NsTM Group, it is likely that diabetic complications in the HsTM Group were more frequent and more severe than in the NsTM Group. In fact, Table 1 shows diabetic complications in the HsTM Group to be more frequent and severe than in the NsTM Group in this study.

As for the mechanisms of increased concentrations of sTM, there seem to be two possibilities: one is the increase of the endothelial damage, and the other is the reduced renal function due to diabetic nephropathy. However, as shown in Fig. 1, the sTM concentrations with nephropathy were higher than those without nephropathy with the same CTM. Furthermore, even in the HsTM Group, 22 patients did not have diabetic nephropathy. These findings cannot be explained by a decreased renal excretion caused by diabetic nephropathy, and seem to suggest that a higher sTM concentration is a result of increased endothelial damage.

Concerning another diabetic complication, patients of the HsTM Group showed with more ischemic heart diseases detected by loading ECG or scintigraphy than those of the NsTM Group. In diabetic clinics, it is impossible to perform the loading ECG test for many outpatients to detect ischemic heart disease because of the enormous number of patients and of limited number of medical staff. In this respect, the serum thrombomodulin concentration can serve as a hallmark in predicting ischemic heart disease.

In conclusion, a high sTM concentration in diabetic patients seems to be a warning marker for detecting diabetic complications, both micro- and macroangiopathies. Though the patients might exhibit diabetic nephropathy, increased endothelial damage seems to be the main cause of the increased sTM levels.

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