Turkish Journal of Endocrinology and Metabolism, (2000) 1 : 19-25 **ORIGINAL ARTICLE**

The Association Between Autonomic Neuropathy and Microalbuminuria with Altered Diurnal Blood Pressure Variation in Type 2 Diabetic Patients

Oğuzhan Deyneli* H. Önder Ersöz* Dilek Gogas-Yavuz* Ali Serdar Fak Sema Akalın***

Marmara University, Istanbul, Turkey

** Department of Endocrinology and Metabolism*

*** Department of Cardiology*

Ambulatory blood pressure monitorization is a valuable tool for the assessment of diurnal variation of blood pressure and diagnosis of hypertension. Altered diurnal blood pressure variation is observed in diabetic patients with overt nephropathy. In this study, we planned to investigate the relationship between diabetic microvascular complications, mainly autonomic neuropathy and early stage nephropathy, and diurnal blood pressure variation in type 2 diabetic patients. We performed 24 hour ambulatory blood pressure monitoring, 24 hour urinary albumin excretion measurements, and autonomic neuropathy tests in healthy controls (C) (n=13), normoalbuminuric (N) (n=30) and microalbuminuric (M) (n=18) type 2 diabetic patients. Frequency of autonomic neuropathy was significantly higher in group M (45%) compared with group C (p<0.005). Although there was no significant difference between day and night blood pressures and heart rates between groups, the amount of dipping of sistolic, diastolic and mean arterial pressures in group M (5.5±3, 9.6±5, 7.1±4%) was decreased compared with group C (10±2, 14.9±3, 12.3±2%) (p<0.005) and group N (8.3±4, 11.9±5, 10.1±4 %) (p<0.05). Frequency of non-dippers was increased in groups M (83%; p<0.0005) and N (57%; p<0.01) compared with group C (18%). The frequency of non-dippers was found to be higher in patients with autonomic neuropathy (85% vs. 45.5%, p<0.05) and the frequency of autonomic neuropathy was found to be significantly higher in non-dipper patients (44.5% vs. 10%, p<0.05). In conclusion increased frequency of nondippers in patients with microalbuminuria and/or autonomic neuropathy demonstrates the relation between altered diurnal blood pressure rhythm and microvascular complications.

Key words: Type 2 diabetes mellitus, autonomic neuropathy, microalbuminuria

• *This work was supported by Eczacıbaşı Scientific. Research and Award Fund.*

Introduction

There is an increased risk of cardiovascular morbidity and mortality in type 2 diabetic patients (1,2). Microvascular and macrovascular complications of diabetes are associated with an increased cardiovascular risk. Microalbuminuria, the first clinical marker of diabetic nephropathy, is also a predictorof cardiovascular morbidity and mortality (3, 4). Prospective studies in diabetic patients have shown an increased mortality rate in patients with autonomic neuropathy (5, 6).

Ambulatory blood pressure monitorization is a valuable tool for the assessment of diurnal variation of blood pressure and diagnosis of hypertension. Absence of a natural decline in nocturnal blood pressure is associated with left ventricular hypertrophy (7-10), increased cardiovascular complications and hypertensive retinopathy (9, 11) in nondiabetic individuals. Diurnal blood pressure variation is also altered in diabetic patients with overtnephropathy (12, 13).

In this study, we planned to investigate the relationship between diabetic microvascular complications, mainly

Correspondence address:

Oguzhan Deyneli
Tophanelioğlu cad. Askorukent sitesi
C Blok No:4 Uskudar
Istanbul Turkey
Tel: +90 216 327 66 67
Fax: +90 216 428 00 13
e-mail: odeyneli@marun.edu.tr

autonomic neuropathy and early stage nephropathy, and diurnal blood pressure variation in type 2 diabetic patients. We proposed an association between impaired nocturnal blood pressure decline (non-dipping) and diabetic microvascular complications, thus one leading to another and also contributing to increased cardiovascular risk.

Materials and Method

Normotensive or mild hypertensive (diastolic blood pressure lower than 100 mmHg) type 2 diabetic patients were enrolled in the study. Patients on an antihypertensive drug were re-evaluated after the cessation of treatment and a two-week wash out period (7). The study group consisted of 20 patients with microalbuminuria (30-300 mg/day albuminuria in at least 2 measurements out of 3) and 31 normoalbuminuric patients, while 13 healthy subjects with no family history of hypertension and, diabetes served as the control group. Diabetes mellitus was diagnosed on the basis of at least 2 measurements of fasting blood glucose over 126 mg/dl or a second hour blood glucose level over 200 mg/dl during an oral glucose tolerance test. Patients with a diagnosis of diabetes after 35 years of age, on diet and/or oral hypoglycaemic drugs, or who had switched to insulin therapy after five years of oral hypoglycaemic drug treatment were accepted as type 2 diabetics.

All of the subjects were informed about the study protocol and gave written informed consent. This study was approved by Marmara University Human Studies Ethics Committee.

The subjects were evaluated with a complete history and physical examination including the measurement and calculation of body mass index prior to the study. The presence and grade of diabetic retinopathy was assessed by a specialist ophthalmologist with indirect ophthalmoscopy. Haemoglobin A1c, serum lipid profiles, creatinine and BUN levels, and urine analysis were obtained. Albuminuria was measured in 24-hour urine samples with a chemiluminesence method (Dade Behring, Newark, DE), and serum and urine creatinine levels with a calorimetric kit (Boehriger Mannheim GmBH, Germany) by the Jaffe method using Hitachi auto-analyser. Haemoglobin A1c levels were measured with an enzymatic immunoassay (ABBOTT-GmbH Diagnostika, Wiesbaden-Delkenheim, Germany) cholesterol and triglyceride levels with a spectrophotometric method by using a Dupond dimension (Boehriger Mannheim GmBH, Germany).

Autonomic nervous function was evaluated by:1. Beat-to-beat variation during deep breathing (abnormal:=<10 beats/min, borderline: 11-14 bpm normal:>15 bpm); 2. Orthostatic blood pressure measurements (change in systolic blood pressure 3 minutes after standing; abnormal: >= 30mmHg , borderline: 11-29 mmHg , normal: <10 mmHg); 3. Heart rate response to the Valsalva maneuvre (Valsalva Ratio is defined as the ratio of longest R-R interval after manoeuvre to shortest R-R interval during the manoeuvre; abnormal: =<1.10, border-line:1.11-1.20; normal: >=1.21); Valsalva manoeuvre was performed by the subjects blowing into a mouthpiece connected to a sphygmomanometer and holding it at a pressure of mmHg for 15 seconds while a continuous electrocardiogram was taken during tests; 4. Immediate heart rate response to standing (30:15 ratio) (abnormal: =<1.00; borderline: 1.01-1.03; normal: >=1.04) (14). Patients with at least two positive tests out of four were accepted as having autonomic neuropathy. Peripheral neuropathy was evaluated with a pinprick test for sensation and 128 Hz tuning fork for vibration.

Twenty-four hour ambulatory blood pressure monitorizations (ABPM) were performed with a Spacelab® 90207 device. Blood pressure measurements of this device were confirmed by a manual sphygmomanometer both at the beginning and at the end of the 24-hour monitorization period. Blood pressure measurements were performed and recorded at 20 minute intervals during the day-time (07:00 AM - 11:00 PM) and at 30 minute intervals during sleep (11:00 PM-07:00 AM). Subjects were asked to record their wake-up and sleeping times. Diurnal variation of blood pressure was assessed through these records. Mean blood pressure levels were calculated for day-time and sleeping periods. Dippers were defined as subjects with an average reduction in systolic and diastolic blood pressure of greater than 10% from day to night, while the others were classified as “non-dippers” (13, 15). Amount of dipping of blood perssure from day to nihth (%) was calculated as follows : (Mean B.P During Day-Mean Blood Presure During Sleep) * 100/Mean Blood Pressure During Day. (13)

Statistical Analysis

All calculations were made with a commercially available program (Instat II) for IBM computer. For normally distributed variables Tukey-Kramer ANOVA and for non-normally distributed variables Kruskall-Wallis ANOVA tests were done in order to analyse the differences between the three groups. Comparisons between two groups were performed with Student's test or Mann-Whitney U tests where appropriate. Frequencies were compared with Fisher's exact test and correlation analyses were calculated with Pearson test.

All the values are presented as mean ± standard deviation. A two-tailed p values less than 0.05 was considered as significant.

Results

The demographic and clinical characteristics of the subjects are summarised in Table 1. There was no significant difference between the three groups regarding age, gender, body mass index, serum lipid, and creatinine clearance levels. Diabetic microalbuminuric and normoalbuminuric groups were also found to be similar regarding diabetes duration, diabetic retinopathy and haemoglobin A1c levels. Urinary albumin excretion levels were similar control and normoalbuminuric groups but was significantly increased in the microalbuminuric group as expected (8.6±6.0 mg/day, 16.0±7.8 mg/day, and 124.7±84.6 mg/day, respectively, $p<0.001$).

The results of neuropathy tests are summarised in Table 2 and 3. Frequency of autonomic neuropathy was significantlyhigher in microalbuminuric diabetic patients (55%) (11 of 20 microalbuminuric type 2 diabetic patients had autonomic neuropathy) compared with healthy controls ($p<0.005$) but not different between microalbuminuric and normoalbuminuric diabetic patients or normoalbuminuric diabetic patients and healthy controls (Figure 1). Vibration sense was significantly impaired in the diabetic groups compared with healthy controls ($p<0.05$). There was no significant difference between the three groups regarding the loss of pinprick sensation or Achilles reflexes. Twenty-four hour blood pressure and heart rate

Table 1. Demographic and clinical characteristics of patients and controls.

		Tip 2 Diabetic Patients		
	Controls N=13	Normoalbuminuric N=31	Microalbuminuric N=20	
Male/Female	7/6	15/16	5/15	NS
Age (Year)	55±9.704	56.5±10.217	53.15±10.927	NS
BMI (Kg/m^2)	27.66±2.887	27.25±2.844	29.53±4.9	NS
Total Cholesterol (mg/dl)	210.91±33.38	231.58±47.02	237.85±49.49	NS
HDL (mg/dl)	41.33±4.43	47.70±8.35	52.55±15.08	NS
Triglyceride (mg/dl)	134.83±64.15	141.53±66.88	163.15±102.88	NS
LDL. (mg/dl)	142.75±34.61	155.77±42.82	148.2±34.13	NS
HbA_{1C} (%)	$5.23±0.32^{a,b}$	$7.36 ±1.139^{a}$	$8.32 ±2.042^{b}$	a,b
Creatinine clearance (ml/min)	104.5±26.41	94.581±29.594	84.275±30.49	NS
Diabetes Duration (Year)	-	5.96±5.743	7.65±6.798	NS
Microalbuminuria (mg/day)	$8.6±5.95^{b}$	$16±7.75^{c}$	$124.7±84.61^{b,c}$	b,c
Retinopathy (Absent/NonProlif./ Proliferative)		29 / 2 / 0	16 / 2 / 2	NS

a: $p<0.001$ Normoalbuminuric group vs Control group

b: $p<0.001$ Microalbuminuric group vs Control group

c: $p<0.001$ Microalbuminuric group vs Normoalbuminuric group

NS : Non significant

Table 2. Autonomic Neuropathy.

	Controls n=13	Type 2 Diabetic Patients Normo albuminuric n=31	Micro albuminuric n=20	
Heart rate variation during deep breathing (N/B/A)*	13/0/0 a, b	12/6/13 b	5/3/12 a	a,b
Valsalva Ratio (N/B/A)*	13/0/0 c	23/3/5	12/5/3 c	c
Immediate heart rate response to standing 30/15 ratio (N/B/A)*	12/1/0 d	20/2/9	9/2/9 d	d
Blood pressure response to standing 3rd min. (N/B/A)*	13/0/0	30/1/0	17/2/1	N.S. #
Frequency of autonomic neuropathy (Present/Absent)	0/13 e	7/24	9/11e	e
Pinprick sensation (Normal/Reduced/ Absent)	13/0/0	20/11/0	16/3/1	N.S. #
Vibration (Normal/Reduced/ Absent)	12/1/0	14/13/4	9/10/1	f,g
Achilles reflex (present/absent)	13/0	21/10	15/5	N.S. #

*N/B/A: Normal/Borderline/Abnormal
#: No significant difference p>0.05
a: Microalbuminuric group vs Control group, p<0.0001
b: Normoalbuminuric group vs Control group, p<0.005
c: Microalbuminuric group vs Control group, p<0.05
d: Microalbuminuric group vs Control group, p<0.01
e: Microalbuminuric group vs Control group, p<0.005
f: Microalbuminuric group vs Control group, p<0.05
g: Normoalbuminuric group vs Control group, p<0.05

Table 3. Number of Patients with Autonomic Neuropathy in Dipper and Non-dipper Diabetics.

	Controls n=13	Type 2 Diabetic Patients Dipper n=22	Non-dipper n=29	
Heart rate variation during deep breathing (N/B/A)*	13/0/0 a, b	8/3/8 b	10/5/17 a	a, b
Valsalva Ratio (N/B/A)*	13/0/0 c	16/2/1	19/6/7 c	c
Immediate heart rate response to standing 30/15 ratio (N/B/A)*	12/1/0 c	17/1/1	13/3/17 c	c
Blood pressure response to standing 3rd min. (N/B/A)*	13/0/0	19/0/0	28/3/1	N.S.
Frequency of autonomic neuropathy (Present/Absent)	0/13 d	3/16	13/19d	d

a: nondipper vs controls p<0.005
b: dipper vs controls p<0.01
c: Non-dipper vs controls p<0.05
d: Non-dipper vs dipper p<0.05
N.S.: Non significant

variations of the three groups are summarised in Table 4. There was no difference between the three groups regarding day-time or night-time systolic, diastolic, and mean arterial pressures or heart rates. Nocturnal reduction in systolic blood pressure was more prevalent in healthy controls (10.0%, p<0.001) and normoalbuminuric diabetic patients (8.3%, p<0.05) compared with the microalbuminuricgroup (5.5%). There was a 14.9% decline in nocturnal diastolic blood pressure in healthy controls, while this was only 9.6% in microalbuminuric patients group (p<0.05). Diurnal variation of mean arterial pressure was also blunted in microalbuminuric patients (7.1%) compared to normoalbuminuric (12.3%, p<0.001). Frequency of non-dippers was significantly higher in diabetic patients compared

with healthy controls. Non-dippers comprised 70% (14 of 20 patients) of the microalbuminuric group ($p<0.001$) and 48.3% (15 of 31 patients) of the normoalbuminuric diabetic group ($p<0.05$) while only 7.6% (1 of 13) of the healthy controls were non-dippers (Figure 1).

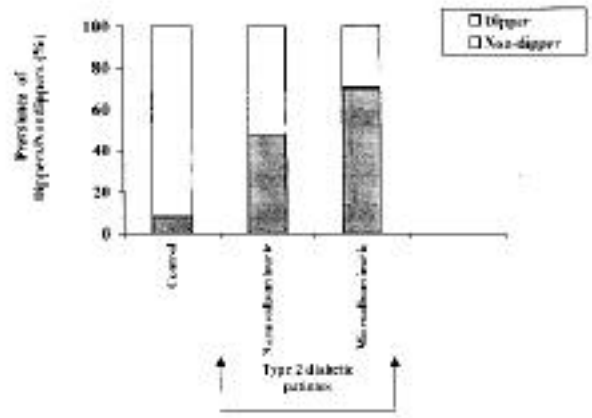

Figure 1. Comparisons of Prevalence of Dippers and Nondippers.

- Prevalence of non-dippers in microalbuminuric group>Prevalence of non-dippers in control group, $p<0.0004$
- Prevalence on non-dippers in normoalbuminuric group>Prevalence of non-dippers in control group, $p<0.02$

When diabetic patients were separated into two groups according to autonomic neuropathy status, the frequency of non-dippers was found to be higher in patients with autonomic neuropathy [13 of 16 AN(+) patients vs 19 of 35 AN(--) patients](81.25% vs. 45.7%, $p<0.05$). When diabetic patients were separated into two groups according to diurnal blood

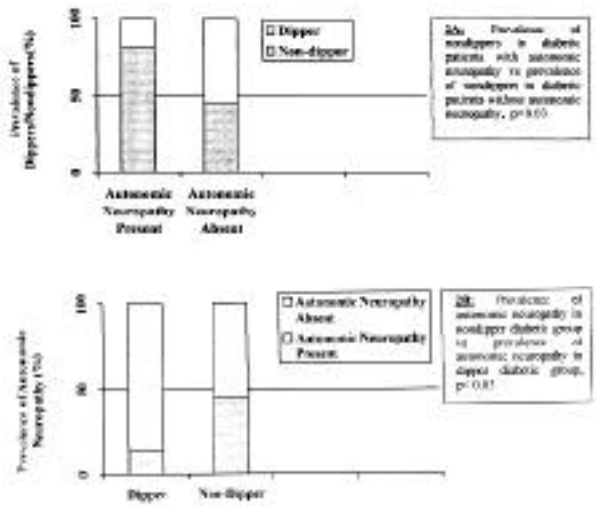

Figure 2. A: Autonomic nervous function in dippers and nondippers . B: Prevalence of Dippers and Nondippers in Patients with/ without Autonomic Neropathy

Table 4. Ambulatory blood pressure measurements and diurnal variations

	Daytime				Night-time				Amount of Blood Pressure Change From Day to Night			
	Systolic B.P. MmHg	Diastolic B.P. MmHg	MAP MmHg	Heart rate beat/min	Systolic B.P. MmHg	Diastolic B.P. MmHg	MAP MmHg	Heart rate beat/min	Systolic B.P	Diastolic B.P.	MAP	Heart rate
Control Group	127.94 ± 8.5	77.18±3.6	94.57±4.5	71.9±3.9	114.30±8.5	63.49±4.5	82.46±4.6	62.05±9.8	%10±2.3	%14.9±3.3	%12.3±2.1	%13.3±3.2
Normo albuminuric	123.76±9.3	77.01±7.6	94.08±7.6	77.34±9.8	114.40±8.3	67.41±6.4	84.08±6.1	64.26±8.9	%8.3±4.7	%11.9±3.6	%10.1±4.8	%13.8±6.8
Micro albuminuric	135.0±16.9	80.94±10.1	99.86±12.2	79.49±12.2	126.24±19.9	73.19±10.6	92.71± 13.8	68.69±11.3	%5.8±3.9	%9.6±5.4	%7.1±4.8	%13.1±6.0
	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	a,b	c	d, e	N.S.

a: Controls (%) vs Microalbuminuric group (%), $p<0.001$
b: Normoalbuminuric group (%) vs Microalbuminuric group(%), $p<0.05$
c: Controls (%) vs Microalbuminuric group(%), $p<0.005$
d: Controls (%) vs Microalbuminuric group (%), $p<0.001$
e: Normoalbuminuric group (%) vs Microalbuminuric group (%), $p<0.05$

pressure variations, the frequency of autonomic neuropathy was found to be significantly higher in non-dipper patients[13 of 32 nondipper patients vs 3 of 19 dipper patients] (44.5% vs. 15.7%, $p<0.05$) (Figure 3A and 3B).

There was a significant correlation between HbA1c and urinary albumin excretion levels in diabetic patients ($r=0.66$, $p<0.0001$). Other variables were not correlated with each other.

Discussion

In this study we found a significant association between autonomic neuropathy, early stage nephropathy and altered diurnal blood pressure variation in type 2 diabetic patients. Normal nocturnal decline of arterial blood pressure was found to be significantly blunted in the microalbuminuric group. Diurnal variation of blood pressure was not quite different from healthy controls in the normoalbuminuric diabetic group, indicating a correlation between microalbuminuria and altered diurnal variation of blood pressure. On the other hand, the frequency of non-dippers was significantly higher in both diabetic groups compared with healthy controls, although being more prevalent in the microalbuminuric group. A similar finding was observed by Nielsen et al previously in a group of type 2 diabetic patients with overt nephropathy (13). In that study, the nocturnal blood pressure reduction and frequency of dippers was found to be significantly lower in nephropathic type 2 diabetic patients compared with normoalbuminuric type 2 diabetic patients and healthy controls. But at the same time a less prominent difference was also observed between normoalbuminuric type 2 diabetic patients and healthy controls (13).

Impairment of nocturnal decline of blood pressure is a well-established finding for ovent nephropathic diabetic patients (12). But in this study, with the support of the recent literature (13), we demonstrated a similar pattern in microalbuminuric diametic patients. Although noctunal decline of blood pressure in mormoalbuminuric type 2 diabetic patients was not significantly different from controls, significantly increased prevalance of nondippers in the normoalbuninuric diabetic group might demonstrate impairment of nocturnal decline of blood pressure in this group of patients.

The correlation between hypertension and diabetic nephropathy is well understood in type 1 diabetic patients. Elevation of blood pressure has always followed the onset of diabetic nephropathy (16-18). But such a cause and effect relationship is not always present for type 2 diabetic patients. Hypertension may precede diabetic nephropathy in this group of patients, further aggravating and being aggravated by the nephropathy process (4,19). In this study we demonstrated a disturbed diurnal blood pressure pattern in normotensive and mild hypertensive type 2 diabetic patients before the onset of nephropathy. This disturbed pattern may be the result of hyperinsulinemia, insulin resistance, sympathetic hyperactivity, endothelial damage, or diabetes itself. Explanation of this pattern needs further investigation, but an evident explanation may be the effect of autonomic neuropathy.

In this study we demonstrated an increased frequency of non-dippers in diabetic patients with autonomic neuropathy and an increased frequency of autonomic neuropathy in non-dippers. An increased frequency of autonomic neuropathy in non-dippers. Similar data (20.21) had been reported previously suggesting a close relation between autonomic neuropathy and impaired nocturnal blood pressure decline. We also found an increased frequency of autonomic neuropathy in microalbuminuric diabetic patients. Spallone et al studied type 1 diabetic patients, excluding the ones with overt nephropathy, and reported a higher night-time blood pressure in patients with autonomic neuropathy. However night-time urinary albumin excretion rate was also elevated in patients with autonomic neuropathy, evidence of a complex relationship between autonomic neuropathy, elevated night-time urinary albumin excretion and a high night/day ratio of blood pressure (22). We have also demonstrated a similar pattern, which may be of prognostic importance. Alteration of diurnal blood pressure variation is proposed to be a predictor for the transition from normoalbuminuria to microalbuminuria in type 1 diabetic patients (23). Although it needs to be demonstrated with longitudinal studies, we suggest a similar hypothesis for type 2 diabetic patients.

References

1. Swerdlow AJ, Jones ME. Mortality during 25 years of follow-up of a cohort with diabetes. *Int J Epidemiol* **25:** 1250-1261, 1996.
2. Wang SL, Head J, Stevens LFuller JH. Excess mortality and its relation to hypertension and proteinuria in diabetic patients. The World Health Organization multinational study of vascular disease in diabetes. *Diabetes Care* **19**: 305-312,1996.
3. Mogensen CE. Prediction of clinical diabetic nephropathy in IDDM patients. *Diabetes* **39:** 761-766, 1990.
4. Ritz E, Stefanski A. Diabetic nephropathy in Type II diabetes. *Am J Kidney Dis* **27:** 167-194, 1996.
5. Ewing DJ, Campbell IW, Clarke BF. Mortality in diabetic autonomic neuropathy. *Lancet* **I:** 601-603, 1976.
6. Rathmann W, Ziegler D, Jahnke M, Haastert B, Gries FA. Mortality in diabetic patients with cardiovascular autonomic neuropathy. *Diab Med* **10:** 820-824, 1993.
7. Sixth report of the joint national committee on prevention, detection, evaluation, and treatment of high blood pressure: *Arch Int Med* **157:** 2413-2445, 1997.
8. Verdecchia P, Schillaci G, Guerri M, Gatteschi C, Benemio G, Boldrini F, Porcellati C. Circadian blood pressure changes and left ventricular hypertrophy in essential hypertension. *Circulation* **81:** 528-536, 1990.
9. Palatini P, Penzo M, Racioppa A, Zugno E, Guzzerdi G, Anacierio M, Pessina AC. Clinical relevance of nighttime blood pressure variability. *Arch Intern Med* **152:** 1855-1860, 1992.
10. Verdecchia P, Schillaci G, Gatteschi C, Zampi I, Battistelli M, Bartoccini C, Porcellati C. Blunted nocturnal fall in blood pressure in hypertensive women with future cardiovascular morbid events. *Circulation* **88:** 986-992, 1993.
11. Parati G, Pomidossi G, Albini F, Malaspina D, Mancia G. Relationship of 24-hour blood pressure mean and variability to severity of target organ damage in hypertension. *J Hypertens* **5:** 93-98, 1987.
12. Hansen KW, Mau Pedersen M, Marshall SM, Christiansen JS, Mogensen CE. Circadian variation of blood pressure in patients with diabetic nephropathy. *Diabetologia* **35:** 1074-1079, 1992.
13. Nielsen SF, Rossing P, Bang L, Svendsen TL, Gall MA, Smidt MU,Parving HP. On the mechanism of blunted nocturnal decline in arterial blood pressure in NIDDM patients with diabetic nephropathy. *Diabetes* **44:** 783-789, 1995.
14. Ewing DJ, Clarke BF. Diagnosis and management of diabetic autonomic neuropathy. *BMJ* **285:** 916-918, 1982.
15. Verdecchia P, Schillaci G, Boldrini F,Guerrieri M, Porcellati C. Sex, cardiac hypertrophy and diurnal variations in essential hypertension. *J of Hypertension* **10:**683-692, 1992.
16. American Diabetes Association, Clinical practice recommendations 1997: Diabetic Nephropathy. *Diab Care* 20 (Supp. 1): S67-S71, 1997.
17. Mathiesen ER, Oxenboll B, Johansen K, Svendsen PA, Deckert T. Incipient nephropathy in type I DM. *Diabetologia* **26:** 406-410, 1984.
18. Parving HH, Smidt UM, Friisberg B, Bonnevie-Nielsen V, Andersen AR. A prospective study of glomerular filtration rate and arterial blood pressure in IDDM with diabetic nephropathy. *Diabetologia* **20:** 457-461, 1981.
19. Molich ME. Management of early diabetic nephropathy. *Am J Med* **102:** 392-398, 1997.
20. Nakano S, Uchida K, Kigoshi T, Azukizawa S, Iwasaki R, Kaneko M, Marimoto S. Circadian rhythm of blood pressure in normotensive NIDDM subjects. Its relationship to microvascular complications. *Diab Care* **14:** 707-711, 1991.
21. Hornung RS, Mahler RF, Raftery EB. Ambulatory blood pressure and heart rate in diabetic patients: An assessment of autonomic function. *Diabetic Med* **6:** 579-585, 1989.
22. Spallone V, Gambardella S, Maiello MR, Barini A, Frontoni S, Menzinger G. Relationship between autonomic neuropathy, 24 hour blood pressure profile and nephropathy in normotensive IDDM patients. *Diabetes Care* **17:** 578-584, 1994.
23. Poulsen PL, Hansen KW, Mogensen CE. Ambulatory blood pressure in their transition from normo to microalbuminuria: a longitudinal study in IDDM patients. *Diabetes* **43:**1248-1253, 1994.