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Autonomic dysfunction and hemodynamics in vitamin B_{12} deficiency

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Abstract

Orthostatic hypotension in patients with cobalamin (Cbl) deficiency has been reported previously in isolated cases but we are not aware of detailed systematic studies of hemodynamic and autonomic nervous system function in patients with cobalamin deficiency. We investigated hemodynamic and autonomic responses to 60° passive head up tilt (HUT) in 21 patients with vitamin B_{12} deficiency, 21 healthy age-matched control subjects and 9 age-matched patients with diabetes mellitus (DM) and established diabetic neuropathy. To systematically assess hemodynamic and autonomic nervous system function, we performed measurements of heart rate, beat-to-beat systolic and diastolic blood pressure, stroke index, cardiac index, total peripheral resistance index, total power, low (LF) and high (HF) frequency oscillatory component of heart rate variability, LF/HF ratio and spontaneous baroreflex sensitivity. As compared to controls, we found a significant fall of systolic blood pressure during 60 consecutive beats directly after head up tilt; furthermore, a significantly blunted fall of stroke index, cardiac index and a lack of increase of total peripheral resistance index for the duration of tilt in patients with diabetes mellitus and in patients with vitamin B_{12} deficiency. As compared to controls, we observed an altered response of spectral indices of sympathetic activation and vagal withdrawal and an impaired modulation of baroreflex sensitivity during head up tilt suggestive of a complex modification in the neural control activities in patients with cobalamin deficiency, which was comparable to that observed in patients with diabetes mellitus and established autonomic neuropathy. The results suggest that vitamin B_{12} deficiency causes autonomic dysfunction with similar hemodynamic consequences and patterns of autonomic failure as seen in diabetic autonomic neuropathy. Defective sympathetic activation may be the cause for orthostatic hypotension, which is occasionally seen in patients with vitamin B_{12} deficiency. It is concluded that patients with orthostatic hypotension should be screened for cobalamin deficiency.

Keywords: Cobalamin deficiency; Autonomic nervous system function; Stroke index; Peripheral resistance; Power spectral analyses; Heart rate variability; Baroreflex; Head up tilt

1. Introduction

The neurologic disorders associated with cobalamin (Cbl) deficiency are well described (Healton et al., 1991). Both the central nervous system and the peripheral nerves are affected. Orthostatic hypotension (Kalbfleisch and Woods, 1962; White et al., 1981; Eisenhofer et al., 1982; Mcombe and McLeod, 1984; Johnson, 1987; Lossos and Argov, 1991; Dündar and Yücel, 1988; Toru et al., 1999), urinary and faecal incontinence (Kurabayashi et al., 1992; Campellone et al., 1995; Lindenbaum et al., 1988) or impotence (see Healton et al., 1991; Lindenbaum et al., 1988) in singular patients with Cbl deficiency have been reported previously, but hemodynamic and autonomic nervous system function have not been investigated systematically in patients with vitamin B_{12} deficiency. We are aware of two studies that investigated heart rate variability (HRV) in patients with Cbl deficiency in the time and frequency domain (Sözen et el., 1998; Aytemir et al., 2000). Long- and short-term measurements of parameters of HRV were found to be significantly lower as compared to healthy control subjects. It was concluded that Cbl deficiency may cause autonomic dysfunction. In none of the two studies, however, modulation of hemodynamic and autonomic parameters after head up tilt (HUT) were assessed.

Since power spectral analyses of HRV have been introduced (Akselrod et al., 1981), the quantitative evaluation of beat-to-beat cardiovascular control has brought new insights into autonomic nervous system function in health and disease. Vagal activity is the major contributor of the

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high-frequency (HF) oscillatory component of HRV (Akselrod et al., 1981; Pomeranz et al., 1985; Malliani et al., 1991) while interpretation of the low-frequency (LF) component of HRV is still a matter of discussion. Some authors suggest LF to be a marker of sympathetic activity (Malliani et al., 1991; Montano et al., 1994) while other investigations view LF as reflecting both sympathetic and vagal activity (Akselrod et al., 1981; Appel et al., 1989).

The LF/HF ratio appears to be capable of providing evaluation of changes in the state of sympathovagal balance (see Malliani et al., 1991) especially during HUT (see Montano et al., 1994). Considering the limitations of frequency analyses of HRV, we not only performed short-term measurements of HRV but also systematically investigated hemodynamic responses of systolic (SBP) and diastolic (DBP) beat-to-beat blood pressure, stroke index (SI), cardiac index (CI) and total peripheral resistance index (TPRI) to passive 60° HUT in patients with diabetes mellitus (DM) and patients with Cbl deficiency. Furthermore, we assessed spontaneous baroreceptor reflex sensitivity (BRS) and its modulation by orthostasis.

2. Materials and methods

2.1. Subjects

The study population consisted of patients with Cbl deficiency (n=21) and two control groups: one group were healthy, age-matched subjects (n=21), the other age-matched patients with long standing Type II DM uncomplicated except established sensory diabetic neuropathy (n=9). The demographic and clinical characteristics in the three patient groups are given in Table 1. All patients underwent careful medical history, physical and neurological examination. All patients had chest X-rays, 12 lead ECG recordings, two-dimensional echocardiography and routine laboratory tests performed. There was no evidence of heart disease or pulmonary disease in any of the patients or control subjects. None of them had a major neurological deficit beside diabetic neuropathy or neurologic complications of Cbl deficiency. Serum creatinine was below 84 μmol/l (1.10 mg/dl) in all patients. None of the patients was receiving α- or β-adrenergic blockers, α- or β-mimetic agents or anticholinergic medication. None of the patients had paraproteinemia. All subjects had normal levels of serum folate (>7.1 ng/ml). There was no history of alcohol or substance abuse in any of the subjects. None of the patients developed clinical signs of orthostatic hypotension or syncope during HUT testing. All subjects were carefully instructed about the study and gave their informed consent. The experiments were approved by the responsible Ethical Committee.

Table 1
Demographic and clinical characteristics of patients with cobalamin deficiency (Cbl), patients with diabetes mellitus (DM) and control subjects (Contr.)[a]

Variable	Cbl	DM	Contr.
Age (years)	66.7±14.9	60.7±8.6	66.1±10.4
n (male/female)	21 (7/14)	9 (4/5)	21 (13/8)
Body weight (kg)	70±19	79±13	72±12
Height (cm)	166±11	170±9	169±9
Preprandial serum glucose (mmol/l)	5.31±0.63	13.13±5.16	5.11±0.7
HbA1c (%)	0.054±0.0028	0.088±0.012	0.056±0.0015
Serum creatinine (μmol/l)	63.4±11.3	61.3±10.7	69.4±9.2
Serum cobalamin (pmol/l)	83±4	279±97	278±108
Serum folate (nmol/l)	20.2±5.9	14.8±3.7	16.9±5.4

[a] Data are given as means±SD.

2.1.1. Patients with cobalamin deficiency

From June 1996 to August 2000, we investigated a total of 21 patients with newly detected and untreated Cbl deficiency (7 male, 14 female; mean age, 66.7±14.9 years; age range, 34–90 years) who presented themselves with gastroenterological, neurological or hematological symptoms (Table 1). Duration of symptoms attributable to vitamin B_{12} deficiency before diagnosis ranged from 2 months to 4 years (mean duration 13.7±13.4 months). To assure the diagnosis Cbl deficiency, all patients had to fulfil one or more of the following criteria in addition to subnormal serum Cbl levels (<138 pmol/l): an abnormal Schilling test result, signs associated with peripheral neuropathy, myelopathy and/or neuropathy (see Healton et al., 1991), involvement of the tongue, anemia or elevated mean corpuscular volume (MCV). Schilling's test was performed in 10 patients and results were abnormal in 7 patients. Ten of the 21 patients had biopsy proven atrophic gastritis, one had gastric resection. *Helicobacter pylori* associated gastritis was present in two patients. In the remainder of the patients, alimentary Cbl deficiency was assumed. All of the patients had normal serum glucose levels at repeated measurements and HbA1c<0.059%. Three patients had had neoplastic disease previously: one had uterine cancer, one had colon and one had breast cancer, which were operated 10, 2, and 1 year(s) before, respectively. At the time of investigation, all these patients were free of disease.

2.1.1.1. Neurological findings in patients with Cbl deficiency. Fourteen of the 21 patients had neurological symptoms associated with peripheral neuropathy, myelopathy and/or neuropathy (see Healton et al., 1991). Twelve patients had nonsegmental diminished vibration sense, five patients had diminished cutaneous sensations distally in the limbs, three were complaining of paresthesias (numbness and prickling, tingling sensations in the feet) and two patients had diminished tendon reflexes. One patient was complaining of limb weakness and one had gait ataxia.

Erectile failure was present in one of the seven male patients. None of the patients had evidence of urinary or rectal incontinence, gustatory sweating, spasticity or altered mental status. Three patients were complaining of dizzy- and light-headedness and two had experienced one singular episode of syncope 2 and 4 months before investigation. Five patients presented with symptoms suggestive of gastrointestinal neuropathy. In these cases, a gastrointestinal manifestation of autonomic neuropathy caused by Cbl deficiency was assumed. Two of them were suffering from diarrhoea, two had persistent constipation and one patient had disorders of gastric emptying.

2.1.1.2. Hematological findings in patients with Cbl deficiency. The hematological findings in the patients with Cbl deficiency were as follows: red blood cell count $4.34\pm0.83\times10^{12}$/l; hemoglobin (Hb) 134±21 g/l; hematocrit (Hct) 40.7±6.5%; MCV 95.2±7.8 fl; mean cell hemoglobin (MCH) 30.9±2.7 pg; white blood cell count $6.44\pm2.0\times10^{9}$/l; platelet count $261.4\pm73.6\times10^{9}$/l. Twelve of the 21 patients had high MCV (>95.0 fl). Coexisting iron deficiency may have influenced red cell size in three patients. Red blood cell counts were decreased in seven patients (two males, five females). Hct was decreased in three subjects (one male, two female) and Hb levels were decreased in four patients (one male, three female). Only 2 of the 21 patients showed typical features of pernicious anemia: high MCV, decreased Hb levels, decreased Hct, decreased red blood cell counts, elevated serum levels of lactate dehydrogenase (LDH), high serum bilirubin and neutrophilic hypersegmentation. At the time of investigation, only one of the patients was seriously anemic (Hb 75 g/l). White blood cell counts and platelet counts were normal in all 21 patients.

2.1.2. Patients with diabetes mellitus

Nine patients with long standing Type II DM (four male, five female; mean age, 60.7±8.6 years; age range, 49–72 years; mean duration of DM 16.6±8.4 years; mean HbA1c 0.088±0.012) and established sensory diabetic neuropathy served as a second control group (Table 1). Subjects with signs of sensory diabetic neuropathy were chosen as controls because of evidence of autonomic dysfunction in this population (Sundkvist, 1981; Kihara et al., 1998). None of the diabetic subjects had evidence of Cbl deficiency. Five of the nine patients were on treatment with insulin; four patients were receiving oral hypoglycaemic agents.

2.1.2.1. Neurological findings in patients with DM. All patients had signs of peripheral diabetic neuropathy, such as diminished thermal perception thresholds, paresthesias (numbness and prickling, tingling sensations in the feet), impaired touch and pain perception or painful neuropathy. In addition, all subjects in this group had diminished vibration sense determined over the medial malleoli as a criterion to participate in the study. Three of the four male patients were complaining about erectile failure. Five patients were complaining of dizzy- and light-headedness and two had experienced one singular episode of syncope in the last 6 months before investigation. None of the patients had evidence of gustatory sweating, bowel and bladder dysfunction or gastroparesis.

2.1.3. Control subjects

A total of 21 volunteer healthy control subjects (13 male, 8 female; mean age, 66.1±10.4 years; age range, 46–83 years) served as control group. All of them had normal serum glucose levels at repeated measurements and HbA1c<0.059%. All control subjects had normal serum Cbl levels (>138 pmol/l) (Table 1). None of them was complaining of dizzy- or light-headedness and none of them had experienced an episode of syncope before. None of them had neurological impairment.

2.2. Methods

2.2.1. Experimental protocol

Tilt table tests were performed between 9 and 12 am after a light breakfast to avoid hypoglycaemia during investigation in diabetic patients. Experiments were performed in a well tempered, quiet room where lightning was dim. Patients were positioned on a comfortable manually driven tilt table with foot board support. Each experimental session began with a 20-min supine rest period, during which equipment was adjusted (Benditt et al., 1996). The arm was positioned such that the lower arm remained near the hydrostatic indifference point at any body posture. After 20 min of supine rest, hemodynamic parameters were continuously recorded for 10 min in the supine position and for 10 min after 60° passive HUT.

2.2.2. Autonomic and hemodynamic measurements

To evaluate autonomic nervous system function, we used the Task Force Monitor® (CNSystems Medizintechnik, Graz, Austria) (Gratze et al., 1998, 1999; Fortin et al., 1998), which includes ECG, impedance cardiography (ICG), beat-to-beat blood pressure by the vascular unloading technique (Penaz, 1973) and oscillometric blood pressure recording performed on the contralateral upper arm. The ECG, impedance signal and beat-to-beat blood pressure was sampled with 1000 Hz each. These data were used to calculate online all hemodynamic parameters.

Reproducible measurements of SI and CI can be obtained using ICG (Veigl and Judy, 1983), which was performed by standard methods (Kubicek et al., 1966). A constant sinusoidal alternating current I_O of 400 μA and 40 kHz is passed through the thorax between an electrode placed around the neck and another electrode placed around the lower thorax aperture. The voltage $u(t)$ is acquired by two further electrodes placed between the admitting electrodes, each at a distance of at least 3 cm from the outer electrodes, in order to produce a homogeneous current field between them. The

detected voltage $u(t)$ is proportional to the thorax impedance ($Z(t)=u(t)*I_O$).

The electrocardiogram (ECG) is derived from two separate adhesive monitoring electrodes (CNSystems®), which are placed on the thorax, to give maximal amplitude of the R-wave. Continuous blood pressure is derived from the finger using the vascular unloading technique (see Penaz, 1973) and corrected to absolute values with oscillometric blood pressure measurement by the Task Force Monitor®. Beat-to-beat blood pressure reactions after HUT derived by the vascular unloading technique (see Penaz, 1973) were analysed by calculating the mean systolic and diastolic blood pressure of 60 consecutive beats directly after HUT. Standard formulas were used for calculation of SI and TPRI. Stroke volume was calculated according to Kubicek et al. (1966). TPRI was calculated according to Ohm's law; TPRI=MABP/CI (MABP is mean arterial blood pressure).

Spectral analysis of RR-interval was performed exactly as described by De Boer et al. (1984). Consecutive samples of 256 heart beats or multiples of them are used to calculate power spectra by Fast Fourier transformation. The program provides 3D power spectra by shifting the sampling window by increments of 10 heart cycles. TP and power in each oscillatory component as defined by others (Bellavere et al., 1992) is computed: (a) the very low frequency (VLF) component 0–0.05 Hz, (b) the LF component between 0.05 and 0.17 Hz and (c) the HF component between 0.17 and 0.40 Hz.

Parameters of HRV for LF and HF oscillatory components were expressed as normalised units (nu), obtained by dividing the absolute power of each oscillatory component by TP minus the VLF component and multiplying by 100 (Malliani et al., 1991; Pagani et al., 1986; Furlan et al., 1998). The normalisation tends to minimise the effect of the changes in TP on the values of LF and HF components.

The hemodynamic data were used for automatic calculation of BRS using the sequence method (Parati et al., 1995). Briefly, the algorithm searches for episodes of spontaneous activation of the baroreceptor reflex. Episodes of baroreceptor reflex activation are defined when blood pressure rises/falls for at least 1 mm Hg for at least four consecutive heart beats and when, simultaneously, decrements/increments of RR-interval of at least 4 ms/beat, respectively, occur. Linear regressions of increments/decrements in SBP and increments/decrements in RR-interval were computed. Only episodes with correlation coefficients $r>0.95$ were selected, and from all regressions, a mean slope of baroreceptor sensitivity was calculated for each steady state period.

2.2.3. Serum cobalamin levels

Serum cobalamin levels were measured using Enzyme-immune-assay methods (Abbott Axsym System®).

2.2.4. Vibration perception

A tuning fork (Rydel Seiffer®) was used for determining vibration perception.

2.2.5. Data analyses

After screening data for significant differences between the three groups using nonparametric Kruskal–Wallis test, Student's unpaired *t*-test and nonparametric Mann–Whitney *U*-test were performed for between-group comparison. Student's paired *t*-test and the nonparametric Wilcoxon's rank sum test were used for within-group comparison. Differences were considered significant if $P<0.05$ for the null hypothesis. Statistical analyses were carried out using StatView for Windows 95 Version 4.5 ©1992–1997. Data are expressed as means±standard deviation (SD).

3. Results

Fig. 1 shows representative examples of original trend recordings in (A) a healthy control subject, (B) a patient with Type II DM and (C) a patient with Cbl deficiency before and after HUT.

3.1. Hemodynamic measurements

Hemodynamic parameters in the three patient groups including the *P* values of significant differences are given in Table 2. Artefact-free recordings of HR and beat-to-beat blood pressure were available in all investigated subjects.

3.1.1. Heart rate

As seen from Table 2, patients with Cbl deficiency had significantly higher mean resting heart rates when compared with controls, even when anemic patients were excluded from analysis.

3.1.2. Blood pressure

The fall of SBP during 60 beats directly after HUT was significant in patients with DM and in patients with Cbl deficiency but not in control subjects.

3.1.3. Stroke index, cardiac index and total peripheral resistance index

Artefact-free recordings of impedance signals were available in 18 out of 21 patients with Cbl deficiency, in 9 out of 9 patients with DM and in 19 out of 21 control subjects. After HUT, TPRI rose highly significant in control subjects only. There was no significant change of TPRI in patients with DM and in patients with Cbl deficiency (see Fig. 2A). The decrease of SI induced by HUT (Delta SI) was significantly blunted in patients with DM and patients with Cbl deficiency when compared to controls (see Table 2).

3.2. Autonomic measurements

Autonomic parameters in the three patient groups including the *P* values of significant differences are given in Table 3.

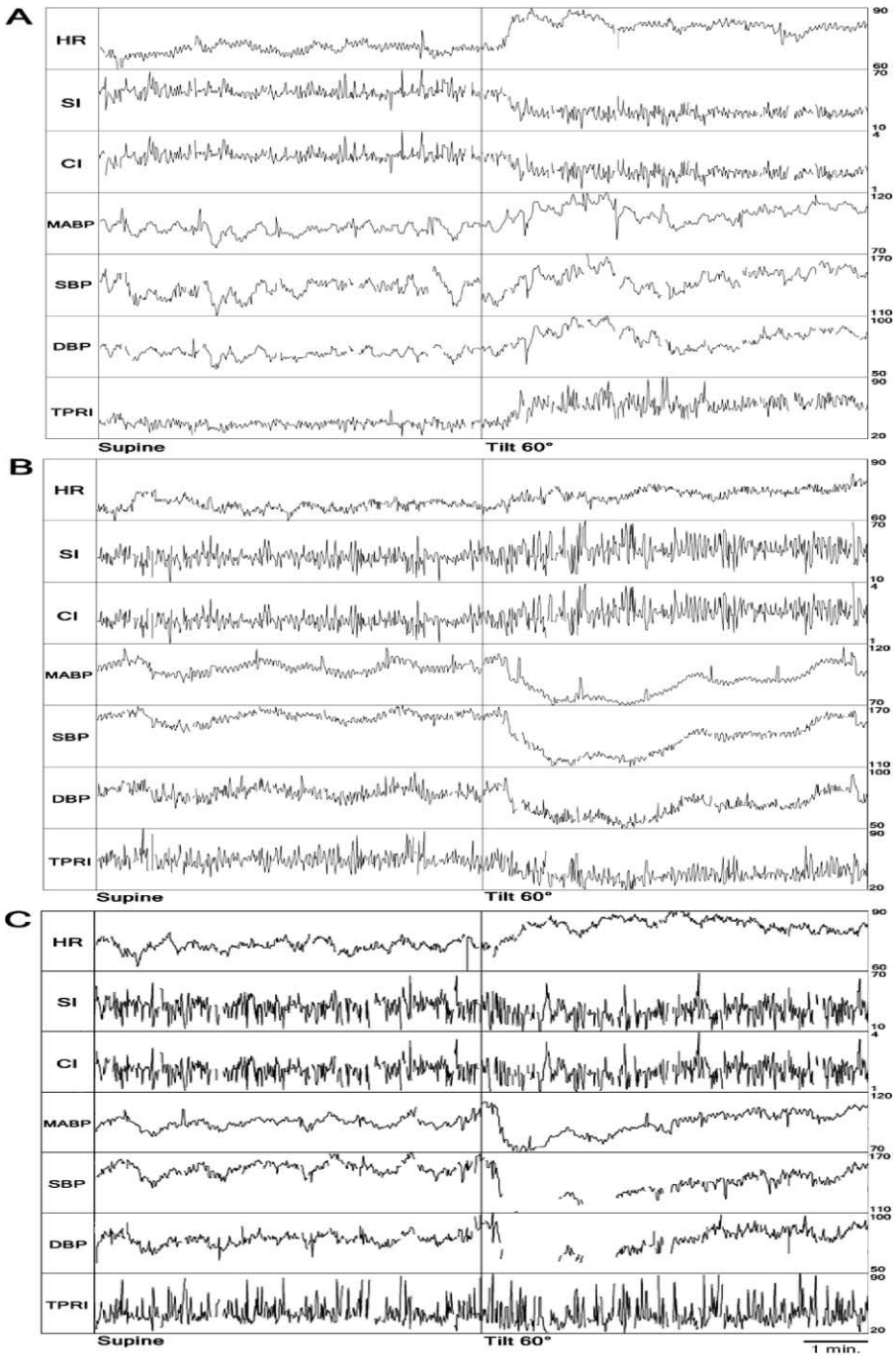

Fig. 1. Hemodynamic trend recordings of a healthy control subject (A), a patient with Type II diabetes mellitus (B) and a patient with cobalamin deficiency (C). HR, heart rate; SI, stroke index; CI, cardiac index; MABP, mean arterial blood pressure; SBP, systolic blood pressure; DBP, diastolic blood pressure; TPRI, total peripheral resistance index. Note the immediate but short lasting fall of blood pressure and the long lasting failure to increase TPRI in (B) and (C). Note that although HR rises in the patient with DM and the vitamin B_{12} deficient patient, in contrast to the healthy control subject, TPRI fails to rise after tilt. This may indicate that peripheral sympathetic failure may not be detected by measuring the sympathetic response at the level of the heart.

3.2.1. Spectral analyses of heart rate variability

Supine TP was significantly reduced in patients with DM. Patients with DM and patients with Cbl deficiency had significantly lower LF power after HUT when compared with controls. HF power decreased significantly after HUT in controls only but not in patients with DM and in patients with Cbl deficiency. A marked increase of LF/HF ratio after HUT was found in control subjects only (see Fig. 2B).

3.2.2. Baroreceptor reflex sensitivity

The decrease of BRS after HUT was highly significant in control subjects, significant in patients with Cbl

Table 2
Hemodynamic parameters in patients with cobalamin deficiency (Cbl), patients with diabetes mellitus (DM) and control subjects (Contr.) in the supine position and after head up tilt[a]

Variable	Unit	Cbl	DM	Contr.
HR supine	bpm	73±11**	70±14	64±7
HR tilt	bpm	81±13††	76±14††	72±9††
SBP during the steady state periods (SBP corrected to oscillometric values)				
SBP supine	mm Hg	127±15	147±24‡,**	125±16
SBP tilt	mm Hg	124±20	140±20	123±15
Supine SBP and SBP during 60 beats directly after tilt (SBP not corrected to oscillometric values)				
SBP supine	mm Hg	128±22	131±25	127±21
SBP tilt§	mm Hg	122±27†	119±25†	123±21
DBP during the steady state periods (DBP corrected to oscillometric values)				
DBP supine	mm Hg	68±8	77±10	68±7
DBP tilt	mm Hg	72±11†	78±9	73±7††
Supine DBP and DBP during 60 beats directly after tilt (DBP not corrected to oscillometric values)				
DBP supine	mm Hg	74±14	80±13	73±11
DBP tilt§	mm Hg	71±16	74±16	71±11
SI supine		34.6±11.9	28.4±10.5	40.9±16.9
SI tilt		29.6±9.8††	24.7±5.3	28.2±11.1††
Delta SI		−5.3±4.5***	−3.6±7.9**	−12.7±7.2
CI supine		2.6±1.0	2.0±0.8	2.6±1.1
CI tilt		2.35±0.8	1.9±0.5	2.0±0.8††
Delta CI		−0.2±0.4*	−0.1±0.5*	−0.6±0.4
TPRI supine		41.9±15.8	57.7±21.5	39.5±12.3
TPRI tilt		45.9±22.1	56.7±20.6	50.3±15.7††
Delta TPRI		3.9±8.8*	−1.0±18.9*	10.7±8.0

*$P<0.05$ vs. controls; **$P<0.01$ vs. controls; ***$P<0.001$ vs. controls.
†$P<0.05$ vs. supine; ††$P<0.001$ vs. supine.
‡$P<0.05$ vs. patients with Cbl deficiency.
§Blood pressure measured during 60 consecutive beats directly after tilt.
[a] Data are given as means±SD. HR, heart rate; bpm, beats per minute; SBP, systolic blood pressure; DBP, diastolic blood pressure; SI, stroke index; CI, cardiac index; TPRI, total peripheral resistance index; Delta, changes of hemodynamic parameters induced by tilt (calculated tilt minus supine values).

deficiency and not significant in diabetic patients (see Fig. 2C). The changes of BRS induced by HUT (Delta BRS) were significantly blunted in patients with Cbl deficiency when compared to controls (see Table 3). There was no statistically significant difference in episodes of activation of the baroreceptor reflex between the

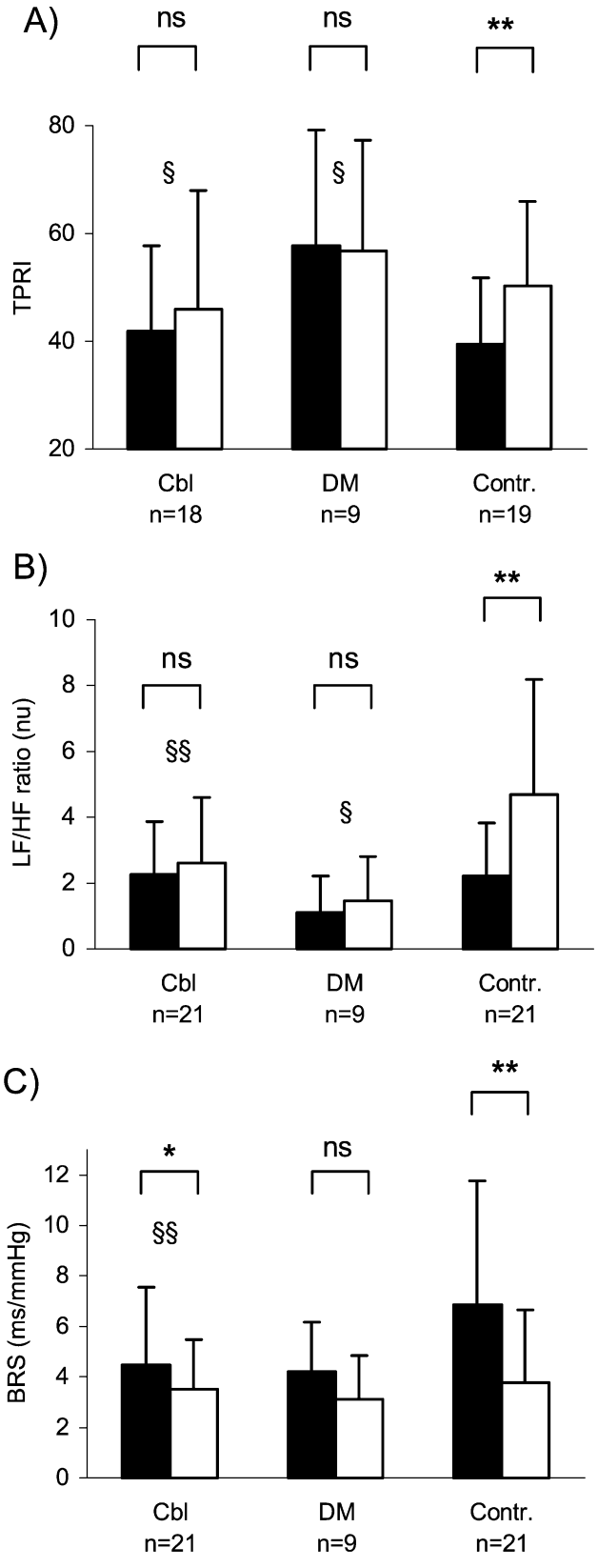

Fig. 2. Total peripheral resistance index (TPRI) (A), LF/HF ratio (B) and baroreceptor reflex sensitivity (BRS) (C) in the supine position (black bars) and after head up tilt (white bars) in patients with cobalamin deficiency (Cbl), patients with Type II diabetes mellitus (DM) and control subjects (Contr.). Data are given as means±SD. ns indicates not significant; * indicates significant intra-group differences. *$P<0.05$ vs. supine; **$P<0.001$ vs. supine. § indicates significant inter-group differences of the Delta values (calculated tilt minus supine values). §$P<0.05$ vs. controls; §§$P<0.01$ vs. controls.

Table 3
Autonomic parameters in patients with cobalamin deficiency (Cbl), patients with diabetes mellitus (DM) and control subjects (Contr.) in the supine position and after head up tilt[a]

Variable	Unit	Cbl (n=21)	DM (n=9)	Contr. (n=21)
TP supine	ms^2	939±1038	264±200†,***	1013±1047
TP tilt	ms^2	638±830	332±331	755±771
P vs. supine		$P<0.05$	ns	$P<0.01$
LF supine	nu	55±24	38±16	56±20
LF tilt	nu	53±25*	46±22**	73±22
P vs. supine		ns	ns	$P<0.005$
Delta LF	nu	−1.2±22**	8±25	17±23
HF supine	nu	34±17	50±18	35±15
HF tilt	nu	30±12	42±12†,**	25±14
P vs. supine		ns	ns	$P<0.001$
Delta HF	nu	−5±15	−8±19	−10±9
LF/HF ratio supine		2.25±1.62	1.10±1.12	2.21±1.61
LF/HF ratio tilt		2.61±2.0*	1.45±1.36**	4.69±3.49
P vs. supine		ns	ns	$P<0.001$
Delta LF/HF ratio		0.35±1.55**	0.35±1.44*	2.48±3.06
BRS supine	ms/mm Hg	4.48±3.07	4.21±1.97	6.85±4.92
BRS tilt	ms/mm Hg	3.52±1.96	3.11±1.73	3.78±2.87
P vs. supine		$P<0.05$	ns	$P<0.001$
Delta BRS	ms/mm Hg	−1.10±1.56**	−0.97±1.99	−3.07±2.88

*$P<0.05$ vs. controls; **$P<0.01$ vs. controls; ***$P<0.001$ vs. controls.
†$P<0.05$ vs. patients with Cbl deficiency.
[a] Data are given as means±SD. TP, total power; LF, low-frequency power; HF, high-frequency power; nu, normalised units; BRS, baroreceptor reflex sensitivity; Delta, changes of autonomic parameters induced by tilt (calculated tilt minus supine values); ns, not significant.

three groups neither in the supine position nor after HUT.

4. Discussion

To the best of our knowledge, the present study is the first which investigates systematically hemodynamic and autonomic responses to orthostatic challenge in a large group of patients with Cbl deficiency as compared to age-matched controls. The important finding of the present study is evidence of major hemodynamic and autonomic impairment comparable to that seen in patients with diabetic autonomic neuropathy.

Specifically, we found elevated mean resting heart rates in patients with Cbl deficiency. Increased heart rates have been reported not only in patients with severe diabetic autonomic neuropathy (Ewing, 1978; Page and Watkins, 1978; Clark et al., 1979) but also in patients with short duration of DM (see Sundkvist, 1981). Rapid heart rates due to parasympathetic damage may represent the initial early stage of cardiac autonomic involvement (Ewing et al., 1980).

A significant decrease of SBP during the first minute after HUT has been described in patients with sensory diabetic neuropathy (see Sundkvist, 1981). We demonstrated a significant fall of SBP during 60 consecutive beats directly after HUT not only in patients with DM but also in patients with Cbl deficiency.

In patients with autonomic failure, orthostatic hypotension results from an impaired capacity to increase vascular resistance during standing (Smit et al., 1999). We found a lack of rise of TPRI in patients with Cbl deficiency comparable to that observed in patients with diabetic autonomic neuropathy (see Fig 2A). Failure to increase TPRI during orthostasis may indicate damage of sympathetic fibres supplying peripheral resistance vessels. As compared to controls in patients with DM and patients with Cbl deficiency, SI and CI decrease less and may even increase during HUT (see Fig. 1B), which may represent a compensatory mechanism to prevent orthostatic hypotension in patients with autonomic failure. This is possibly being achieved by an augmented adrenergic drive to the heart. Measurements of plasma adrenaline in patients with autonomic failure during HUT could confirm or refute this hypothesis. These findings are consistent with our recently published work investigating a group of patients with autonomic failure (see Gratze et al., 1998), which was confirmed using the established Ewing battery (see Ewing et al., 1980).

When analysing autonomic responses in the frequency domain, we demonstrated decreased reduction of HF power and failure to increase LF during HUT. These impaired responses to HUT observed in patients with Cbl deficiency

are comparable in magnitude to those described in patients with DM and established autonomic neuropathy (Pagani et al., 1988a). The similarities in patients with DM and patients with Cbl deficiency are most impressive assessing the autonomic modulation by orthostasis. The shift of the LF/HF balance in favour of the LF component during HUT observed in healthy subjects (see Malliani et al., 1991; Montano et al., 1994; Pagani et al., 1986) is altered in patients with DM and in patients with Cbl deficiency (see Fig 2B). The impaired modulation of the two major components of HRV following orthostatic challenge indicates defective sympathetic activation in both patients with DM and patients with Cbl deficiency.

Calculation of the slope of the regression of heart interval changes, as a function of systolic arterial pressure changes, provides an index of the baroreceptor reflex arc, the afferent limb projecting from the baroreceptors to the medulla and the efferent limbs in the cardiac vagal and sympathetic nerves (Pagani et al., 1988b). The gain of the baroreflex control of heart rate is reduced during exercise (see Pagani et al., 1988b) and sympathetic activation (Pagani et al., 1982) while sleep increases BRS (Symth et al., 1969). Therefore, assessment of the modulation of baroreflex sensitivity during HUT appears to be a sensitive tool for the early detection of autonomic cardiac neuropathy. A study performed by Frattola et al. (1997) revealed that the slope of the regression of heart rate changes as a function of SBP changes was markedly reduced in diabetic patients with and without evidence of autonomic neuropathy. We demonstrated significantly impaired modulation of BRS during HUT in patients with DM and patients with Cbl deficiency (see Fig 2C). Together with the lack of rise of TPRI and failure to significantly increase LF after HUT, the impaired modulation of BRS may indicate defective sympathetic activation during orthostatic challenge in patients with DM and patients with Cbl deficiency.

Diabetic neuropathy is characterised by alteration of large and small peripheral nerve fibres. The damage is most pronounced in patients with severe autonomic involvement (Llewelyn et al., 1991). Disordered blood pressure control in diabetics correlates with pathological abnormalities in the sympathetic nervous system (Low et al., 1975). Toru et al. (1999) described reduction of sudomotor sympathetic unmyelinated fibres in a patient with Cbl deficiency and orthostatic hypotension. Mcombe and McLeod (1984) reported degeneration of unmyelinated fibres in a vitamin B_{12} deficient patient. Therefore, it is not unreasonable to assume that Cbl deficiency causes autonomic neuropathy also affecting small sympathetic postganglionic fibres supplying peripheral resistance vessels.

We are aware of the interpretational limitations of spectral analysis of HRV (Linden and Diehl, 1998). Therefore, we not only analysed the autonomic responses to orthostatic challenge as represented by spectral analysis of HRV but also performed analyses of BRS and included complete hemodynamic assessment in the present study.

The findings in patients with DM are consistent with the abnormalities in frequency HRV analyses associated with diabetic autonomic neuropathy described by the Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology (1996). Striking similarities of hemodynamic and autonomic modulation following HUT could be observed in patients with diabetic autonomic neuropathy and in nonanemic patients with Cbl deficiency. There was no change of the outcome of the study when the three patients with previous but presently cured malignant disease were excluded from the analysis.

5. Conclusions

We conclude that vitamin B_{12} deficiency should be taken into account in the evaluation of patients with symptoms of autonomic dysfunction such as orthostatic hypotension. Patients with orthostatic hypotension should be screened for Cbl deficiency even if they do not show obvious clinical neurologic impairment (see Karnaze and Carmel, 1990) or the typical hematological features (see Lindenbaum et al., 1988; Carmel, 1988) associated with Cbl deficiency.

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