Detection of Early Sympathetic Cardiovascular Neuropathy by Squatting Test in NIDDM

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OBJECTIVE — To determine the role of the squatting test in the detection of early sympathetic neuropathy in patients with non-insulin-dependent diabetes mellitus (NIDDM).

RESEARCH DESIGN AND METHODS — Three groups of nonsmoking, nonobese subjects were studied: 10 healthy subjects, 10 NIDDM patients without autonomic neuropathy (AN), and 10 NIDDM patients with AN defined by the presence of a pathological deep-breathing value. All subjects were given three postural tests: lying-to-standing, sitting-to-standing, and squatting test. Heart rate (HR) and finger arterial pressure were recorded with a noninvasive technique.

RESULTS — Blood pressure (BP) fall (expressed as decremental area) was not significantly different among the groups at standing up after sitting or lying. By contrast, a significantly greater BP drop occurred in NIDDM patients with AN (1,123 ± 245 mm^2) compared with NIDDM patients without AN (460 ± 232 mm^2) or normal subjects (429 ± 138 mm^2, $P < 0.001$). The HR increase after all the orthostatic maneuvers was smaller in diabetic patients with AN ($P < 0.01$) compared with that recorded in other groups. Significant correlations were observed between BP fall after squatting and either the expiration:inspiration ratio at deep breathing ($r = -0.77$, $P < 0.001$) or the duration of diabetes ($r = 0.76$, $P < 0.001$).

CONCLUSIONS — The intrinsic orthostatic load of the squatting test, which is greater than conventional postural maneuvers, makes the squatting test an easy and useful test to detect early orthostatic dysregulation in NIDDM.

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NIDDM, non-insulin-dependent diabetes mellitus; AN, autonomic neuropathy; BP, blood pressure; HR, heart rate; E:I ratio, expiration:inspiration ratio.

In the course of their lives, diabetic patients are frequently exposed to the risk of developing orthostatic hypotension. In a prospective study, older diabetic subjects and younger patients with diabetes of long duration showed an overall prevalence of orthostatic hypotension as high as 12% (1). Furthermore, clinical observations seem to indicate that diabetic patients with symptomatic orthostatic hypotension have a poor prognosis (2). Orthostatic hypotension is classically evaluated by measuring the blood pressure (BP) fall determined by supine-to-erect posture change. However, the defect of orthostatic regulation so detected indicates a severe involvement of the autonomic nervous system. The passage from squatting to standing represents a maneuver that produces an important orthostatic load (3), greater than the ones currently used for diagnosis of autonomic neuropathy (AN). This study compared three different orthostatic maneuvers (squatting, sitting-to-standing, and lying-to-standing) in NIDDM patients with and without AN.

RESEARCH DESIGN AND METHODS

— Three groups of nonsmoking subjects without symptoms of autonomic nervous system involvement volunteered for the study: healthy subjects (group 1, $n = 10$); NIDDM patients without AN (group 2, $n = 10$); NIDDM patients with AN, which was defined as having a pathological deep-breathing value (group 3, $n = 10$). An expiration: inspiration (E:I) ratio >1.16 was chosen as the limit of normality for our 50- to 60-year-old subjects (4). Because a high prevalence of cardiac autonomic dysfunction has been reported in obese people (5), we decided to investigate only normal-weight subjects. Control subjects had no family history of diabetes and normal glucose tolerance according to the criteria developed by the National Diabetes Data Group in 1979. Diabetic patients were in acceptable glycemic control and were treated with oral agents

Table 1—*Clinical and metabolic characteristics and BP and HR increase variations in the three groups of subjects during orthostatic maneuvers*

	Group 1	Group 2	Group 3
Age (years)	54.2 ± 3.5	51.2 ± 3.9	53.8 ± 5.4
Body mass index (kg/m^2)	25.9 ± 1	26.1 ± 1.6	25.3 ± 1.3
Duration of diabetes (years)	—	5.9 ± 3.6	6.8 ± 1.5
HbA_{1c} (%)	5.5 ± 0.6	7.5 ± 0.6	7.8 ± 0.5
BP (mmHg)			
Systolic	121 ± 9	126 ± 10	122 ± 9
Diastolic	76 ± 7	79 ± 7	78 ± 7
Deep breathing	1.24 ± 0.07	1.20 ± 0.06	1.06 ± 0.04
Valsalva maneuver	1.70 ± 0.04	1.51 ± 0.02	1.62 ± 0.04
Lying-to-standing	1.08 ± 0.01	1.09 ± 0.02	1.08 ± 0.02
Sitting-to-standing			
Decremental area (mm^2)	96 ± 9.6	109 ± 13.7	111 ± 12.2
HR increase (% of basal)	30 ± 6.5	38 ± 19.5	15 ± 3*
Lying-to-standing			
Decremental area (mm^2)	109 ± 10	123 ± 12	121 ± 14
HR increase (% of basal)	41 ± 7	40 ± 5	19 ± 5*
Squatting-to-standing			
Decremental area (mm^2)	429 ± 138†	460 ± 231†	1,123 ± 245†
HR increase (% of basal)	70 ± 10.6†	55 ± 9.7†	21 ± 15.5*

Data are means ± SD. The tests for AN are expressed as E:I ratios.
*$P < 0.01$–0.001 group 3 vs. groups 1 and 2.
†$P < 0.05$–0.001 squatting vs. sitting-to-standing and lying-to-standing.

($n = 12$) or insulin ($n = 8$). Insulin therapy was withheld on the morning of the test day. None of the subjects had clinical or biological signs of intrinsic heart, vascular, or lung disease. Any drug known to affect cardiovascular function was withdrawn at least 3 weeks before the study. Informed consent to participate in the study was obtained from all subjects. More detailed information of the clinical and metabolic characteristics of the three groups is given in Table 1.

To evaluate autonomic function, HR (R-R intervals) were automatically recorded by computerized ECG (Burdik Instruments, Milton) during deep breathing, Valsalva maneuver, and lying-to-standing (6). On another occasion, in late morning while in a quiet 20–24°C room, the subjects were submitted to the three postural tests—squatting, sitting-to-standing, and lying-to-standing. HR and finger arterial pressure were recorded with a noninvasive technique (Finapress, Finger Arterial Pressure, Ohmeda 2300, Englewood, CA) that uses the unloaded artery principle (7). The hand connected to the Finapress was placed at heart level and supported by a rubber band during postural changes. After maintaining the starting position for at least 2 min, patients were invited to stand up during the inspiratory phase. All tests were repeated 4 times with a 10-min rest in between. Subjects refrained from coffee on the morning of investigation. Data were elaborated with the aid of an original software developed at our request (Step Ahead, Naples, Italy) that allowed systolic, diastolic, and mean BP, as well as HR, to be expressed in graphs.

The orthostatic pressure fall was calculated as the decremental area between the negative deflection observed during the first 60 s after standing up and the baseline steady-state mean arterial pressure value. Statistical evaluation was conducted with the analysis of variance and nonparametric tests (Wilcoxon's test; sum of rank). All data are expressed as means ± SD.

RESULTS — The BP and HR variations observed during orthostatic tests in the three groups of subjects are reported in Table 1. BP decremental areas were not significantly different among the groups at standing after sitting or lying. By contrast, a significantly greater decremental area was found in group 3 compared with the other groups after squatting. In all groups, BP after the squatting test was significantly higher than after sitting-to-standing or lying-to-standing. The HR increase, expressed in the percentage of baseline after the orthostatic maneuvers, was smaller in group 3 patients compared with that recorded in other groups. In normal subjects and diabetic subjects without AN, squatting-induced HR stimulation was greater than that elicited by sitting-to-standing or lying-to-standing. A negative correlation was noted between BP decremental areas af-

ter squatting and the E:I ratio values ($r = -0.77$, $P < 0.001$) in all subjects and a positive one with duration of diabetes in group 3 ($r = 0.76$, $P < 0.001$). The coefficients of varation of the squatting test were 4.5 ± 1.4% for BP decremental area and 4.8 ± 1.5% for HR increase.

CONCLUSIONS— In this study, we have evaluated the cardiovascular events determined by the squatting test in diabetic subjects without postural hypotension in response to conventional orthostatic maneuvers. The passage from sitting or lying to the upright position produced BP variations in diabetic subjects with AN similar to those seen in either diabetic subjects without AN and nondiabetic subjects. By contrast, the passage from squatting to the standing posture caused a significantly greater pressure drop in diabetic subjects with AN, probably linked to the smaller HR response. In other words, the blunted HR increase in diabetic subjects with AN is still sufficient to avoid an excessive pressure fall in the passage from sitting or lying to standing, but not after a more sustained orthostatic load, such as squatting. The pathophysiological basis for this can be found in the leg artery compression occurring during the postural muscle contraction that lessens the orthostatic pressure fall at standing from sitting or lying (8). On the other hand, the pressure drop cannot be smoothed over by muscle contraction at standing from squatting because an adaptation of BP had already occurred during the steady state of the squatting position (9). Moreover, on standing erect from squatting, the hydrostatic fall in stroke output takes place while the vasodilatation of the previous phase is already present. Thus, a larger pressure fall is seen after squatting.

The significant correlations we found between both diabetes duration or E:I ratio and BP drop after squatting indicate that sympathetic involvement develops early in diabetes. The autonomic dysfunction of diabetes is categorically divided as parasympathetic or sympathetic depending on whether HR alone or HR and BP are affected. Contrasting the current opinion, which holds an earlier and more prominent parasympathetic damage in diabetic neuropathy (10), our results indicate that both parasympathetic and sympathetic fibers are affected at the same time, as also suggested by the data of spectral analysis (11). Although our data were obtained in normal-weight subjects and caution is needed concerning its generalization to the more common obese subset of NIDDM subjects, the passage from squatting to standing appears to be an effective stimulus for the baroregulatory reflex arc providing an easy and useful test to detect early orthostatic dysregulation in subclinical diabetic neuropathy.

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