Delayed Vascular Reactivity to Ischemia in Diabetic Microangiopathy

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To investigate vascular responses in insulin-dependent diabetic patients both with and without retinopathy, we have assessed vasodilation by forearm transcutaneous pO_2 measurement after 10 min of ischemia produced by a sphygmomanometer cuff. Diabetic patients with proliferative retinopathy had a delayed vasodilatory response at 60 s (mean ± SD pO_2 = 9 ± 3 mm Hg) compared with those having diabetes without retinopathy (15 ± 4 mm Hg, $P < 0.01$) and matched normal subjects (14 ± 4 mm Hg, $P < 0.01$). Recently diagnosed insulin-dependent diabetic patients had a very similar response (15 ± 5 mm Hg) to matched normal subjects (15 ± 3 mm Hg). The diminished vascular reactivity may be a consequence of microangiopathy and neuropathy, although patients with an impaired vascular response might be particularly at risk from the development of capillary closure. DIABETES CARE 7: 47–51, JANUARY–FEBRUARY 1984.

Diabetic microangiopathy is not detected before it manifests itself as retinopathy, nephropathy, or neuropathy. Nevertheless, it is likely that the vascular system, being uniformly exposed to the diabetic milieu, suffers damage in a generalized manner. The phenomenon of postischemic hyperemia might be used as a test of vascular integrity.[1] We have studied this response in the forearm of patients with insulin-dependent diabetes who have developed diabetic microangiopathy and in control groups of (1) patients with long-standing diabetes but without any evidence of microangiopathy and (2) recently diagnosed diabetic patients.

METHODS

Patients. In a cross-sectional study, two groups with long-standing insulin-dependent diabetes were studied (Table 1). Eleven patients had proliferative retinopathy documented by color photographs and fluorescein angiography. A second group of 11 patients had nil/minimal evidence of retinopathy (fewer than five microaneurysms in a 30° color photograph field) in spite of a slightly longer duration of diabetes, and no proteinuria. Sensory neuropathy of patients was assessed by measurement of vibration sensory threshold with a Biothesiometer,[2] the assay variability being SD ± 1.3. The patients with proliferative retinopathy were less sensitive than those without retinopathy ($P < 0.05$). Autonomic neuropathy was assessed by standing subjects and measuring the maximal R-R interval shortening at about 15 beats and lengthening at about 30 beats.[3] The R-R variability in response to five deep breaths was also measured.[3] The patients with proliferative retinopathy had a slight but not significant impairment compared with those without retinopathy.

A control group of 13 nondiabetic subjects of similar age and body weight was studied during the same period (Table 1). No patients had angina, myocardial infarct, or other major vascular event.

A group of 11 patients with relatively recently diagnosed type I insulin-dependent diabetes was studied, with a separate parallel control group of 11 age- and sex-matched nondiabetic subjects (Table 2).

The study was approved by the Ethics Committee of the Oxford Area Health Authority, and all patients gave informed consent.

Blood flow monitoring. The tests were done in the morning after a normal breakfast, and all patients rested for at least 30 min in a room where the ambient temperature was kept between 20.5°C and 24.5°C.

At the start of the test, a blood sample was taken for assessment of glycosylated hemoglobin—assayed by the thiobarbituric method[4]—plasma glucose, and creatinine.

Using a sphygmomanometer cuff, forearm ischemia was

TABLE 1
Details of diabetic proliferative retinopathy, diabetic subjects without retinopathy, and nondiabetic groups*

	Normal (mean ± 1 SD)	Diabetic groups: Nil/minimal retinopathy (mean ± 1 SD)	Diabetic groups: Proliferative retinopathy (mean ± 1 SD)	Signif. between diabetic groups
N	13	11	11	
Age (yr)	47.3 ± 7.6	49.2 ± 8.9	44.5 ± 9.3	NS
Duration (yr)	—	27.2 ± 4.6	24.3 ± 4.9	NS
Hemoglobin A_{1c} (%)	7.4 ± 0.7	9.2 ± 1.7	8.8 ± 1.3	NS
Plasma glucose (mmol/L)	4.5 ± 1.2	12.7 ± 5.9	13.0 ± 7.1	NS
Room temperature (°C)	23.2 ± 0.8	22.5 ± 0.6	22.6 ± 1.3	NS
Ideal body weight (%)	118.6 ± 15.9	111.0 ± 7.4	114.5 ± 12.3	NS
Vibration sensory threshold		13.7 ± 5.1	22.0 ± 7.9	$P < 0.01$
Deep breathing heart rate change		1.31 ± 0.31	1.17 ± 0.16	NS
Standing pulse rate change		1.10 ± 0.08	1.06 ± 0.06	NS

The three groups consisted of (1) insulin-dependent diabetic patients who had developed proliferative retinopathy, (2) patients with diabetes of similar or slightly longer duration who had either no or minimal evidence of retinopathy, and (3) a control group of normal subjects.

achieved with a pressure of 200 mm Hg. After 10 min, the cuff pressure was released, and blood flow measurements were made over the next 5 min.

Cutaneous blood flow at the microvascular level was assessed using a Hellige Recommed transcutaneous oxygen monitor.[5] The oxygen electrode, warmed to 37°C, was placed on the ventral surface of the forearm 3–4 in. distal to the antecubital fossa, away from the brachial artery or any visible vein. The electrode was calibrated between ambient atmospheric oxygen and zero, using a metabisulfate solution.

To assess reproducibility of the measurements, one normal subject was studied on nine separate occasions, with a mean ± SD at 30, 60, and 90 s and a peak of 2.5 ± 2.1, 13.7 ± 4.7, 22.7 ± 3.9, and 25.6 ± 3.2 mm Hg, respectively. Four subjects with nil/minimal retinopathy and four with retinopathy were studied on two separate occasions. The reproducibility (±1 SD) at 60 s was ±2.2 and ±1.8 mm Hg, respectively. An effect of exposure to cold was shown by studying the hyperemic response after the arterial cuff in six normal subjects on a warm day, and repeating with the other arm immediately after a period of 30 min, in shirt sleeves, in a 2–3°C cold room. The pO_2 values were significantly reduced—e.g., at 60 s 15.4 ± 2.6 to 6.8 ± 5.6, and peak from 30.0 ± 2.7 to 23.4 ± 4.2 mm Hg—with a response similar to that of the diabetic patients with proliferative retinopathy. Because of this effect, we had subjects rest at 22.8 ± 0.9°C for at least ½ h before study.

Forearm pulsatile blood flow was measured using electrical impedance plethysmography.[6,7] Aluminum electrodes (3M) were placed nearly circumferentially on the forearm. A small constant amplitude alternating current (2 mA rms, 100 kHz) was applied between outer electrodes, and the voltage dif-

TABLE 2
Details of recently diagnosed diabetic patients and a matched group of normal subjects

	Normal	Diabetic
N	9	10
Age (yr)	24.9 ± 3.0	26.9 ± 6.5
Duration (yr)		1.3 ± 0.8
Hemoglobin A_{1c} (%)	6.0 ± 0.6	9.2 ± 2.9
Glucose	5.2 ± 1.0	10.3 ± 4.0
Ideal body weight (%)	101.1 ± 7.9	102.3 ± 11.6

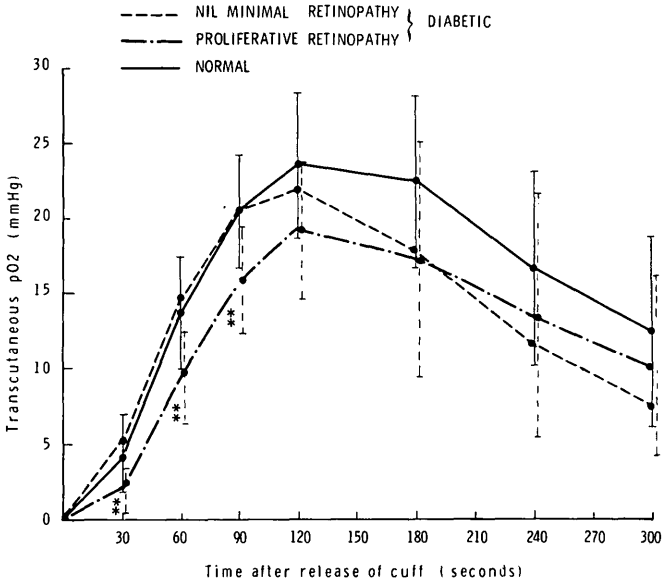

*FIG. 1. Forearm transcutaneous pO_2 after a 10-min period of ischemia by sphygmomanometer cuff at 200 mm Hg (median ± SD equivalent). Diabetic patients without retinopathy had a value similar to that of normal subjects, whereas diabetic patients with proliferative retinopathy had a delayed hyperemic response (*P = <0.01 from normal subjects and nil/minimal retinopathy patients).*

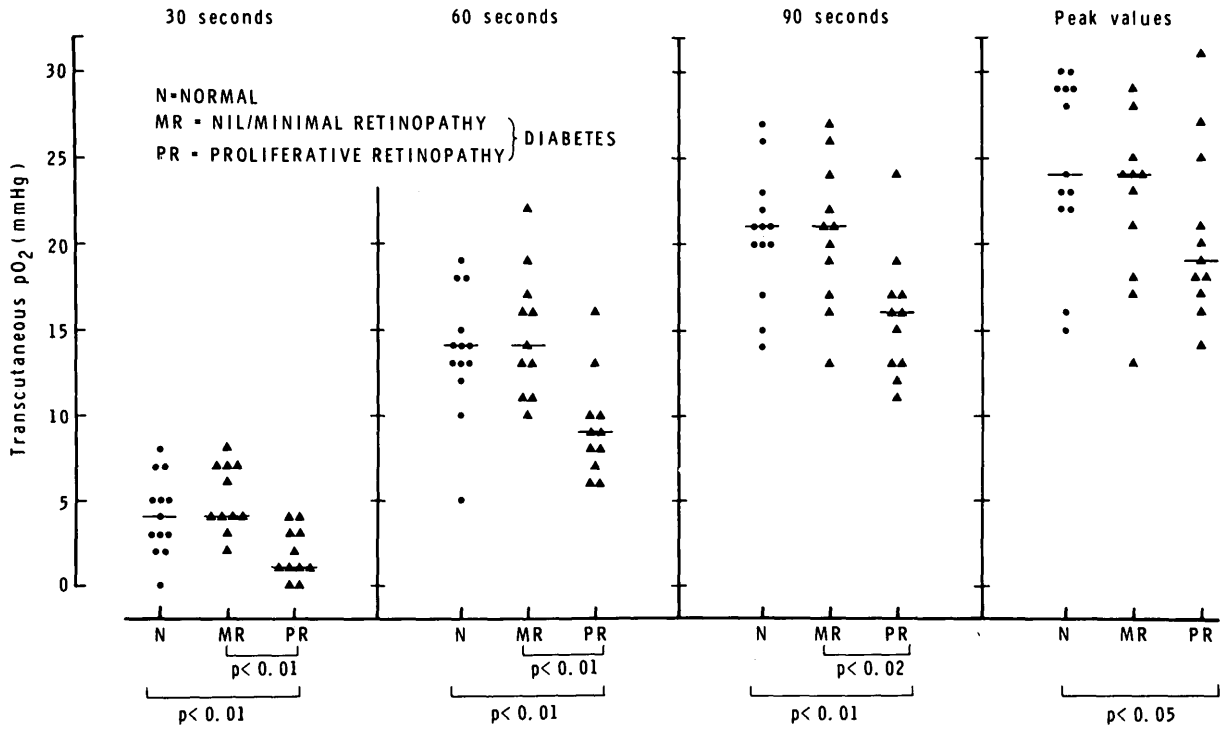

FIG. 2. Individual values of transcutaneous pO_2 at 30, 60, and 90 s after a 10-min period of forearm ischemia, together with the peak values. The median is shown.

ference was measured by separate band electrodes 15 cm apart inside the current electrodes with an instrument of 13-kΩ output impedance. The baseline amplitude of cardiac-synchronous limb impedance was measured from the amplitude of 10 pulse beats, and changes after ischemia were expressed relative to the baseline.

Statistics included ±1 SD, Student's two-tailed *t* tests and correlation coefficient r, except for nonparametric data, which were expressed as medians, with the 66% range equivalent to ±1 SD, and the Mann-Whitney U-test and Spearman rank correlation.[8]

RESULTS

Figure 1 shows the transcutaneous pO_2 after arterial occlusion in the diabetic patients with and without retinopathy and in controls. There was no significant difference between the normal controls and diabetic patients without retinopathy, but the group with proliferative retinopathy had a significantly diminished response compared both with the normal controls and with the nil/minimal retinopathy group at 30, 60, and 90 s after cuff release (Figure 2). The time taken to reach a pO_2 of 10 mm Hg was significantly longer ($P < 0.01$) for the proliferative retinopathy group (63 ± 11 s) than for the nil/minimal retinopathy group (46 ± 8 s) and the normal group (50 ± 10 s). The peak of the response was not different between the two diabetic groups, but the proliferative retinopathy group had a significantly lower peak than did the control subjects ($P < 0.05$).

The group of relatively newly diagnosed diabetic patients had a transcutaneous pO_2 response similar to that of their matched control group (Figure 3).

Blood glucose, glycosylated hemoglobin, percentage ideal body weight, and room temperature are shown in Table 1. There was no significant difference between the diabetic groups with and without retinopathy. The transcutaneous pO_2 results of the diabetic and control groups did not significantly correlate (all $r < 0.1$ for 60-s transcutaneous pO_2) with room temperature, plasma glucose, or glycosylated hemoglobin. The transcutaneous oxygen values at 60 s in all the diabetic patients correlated significantly ($P < 0.05$) with the vibration sensory threshold ($r = -0.54$) and the change in pulse rate upon standing ($r = 0.44$) but not with the change in heart rate on deep breathing ($r = 0.18$).

The electrical impedance plethysmography showed similar basal values in all three groups (normal subjects 18.9 ± 4.4, nil/minimal diabetic 21.8 ± 3.2, and proliferative retinopathy diabetic 21.1 ± 1.8 mΩ). The diabetic patients with proliferative retinopathy had a significantly impaired relative increase in impedance in response to ischemia compared with normal subjects (Figure 4). Diabetic patients with nil/minimal retinopathy were not significantly different from normal subjects or diabetic patients with retinopathy.

DISCUSSION

The hyperemic vascular response to ischemia has been found to be abnormal in patients with insulin-dependent diabetes

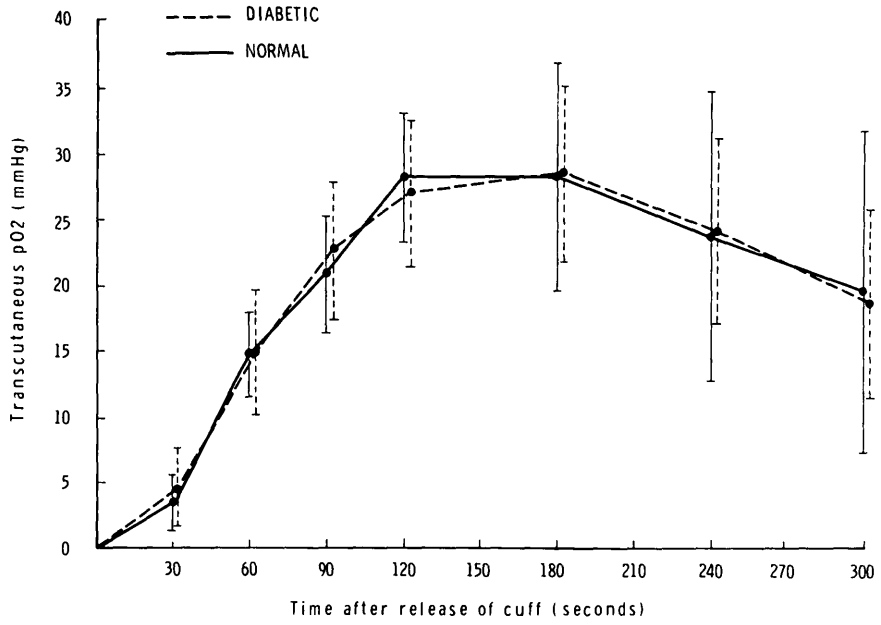

FIG. 3. *Forearm transcutaneous pO_2 after a 10-min period of ischemia in recently diagnosed insulin-dependent diabetic patients and in a matched group of normal subjects. (Median ± SD equivalent.)*

who have developed microangiopathy. A delay in the development of vasodilation was not found in a group of diabetic patients who had minimal retinopathy, despite 27 yr of diabetes mellitus, and their normal response was in accord with their lack of vascular complications.

Recently diagnosed diabetic patients had normal vascular reactivity as compared with matched controls. The lack of correlation between glycosylated hemoglobin and the hyperemic response in any group also indicated that diabetes does not affect this physiologic response. This contradicts a

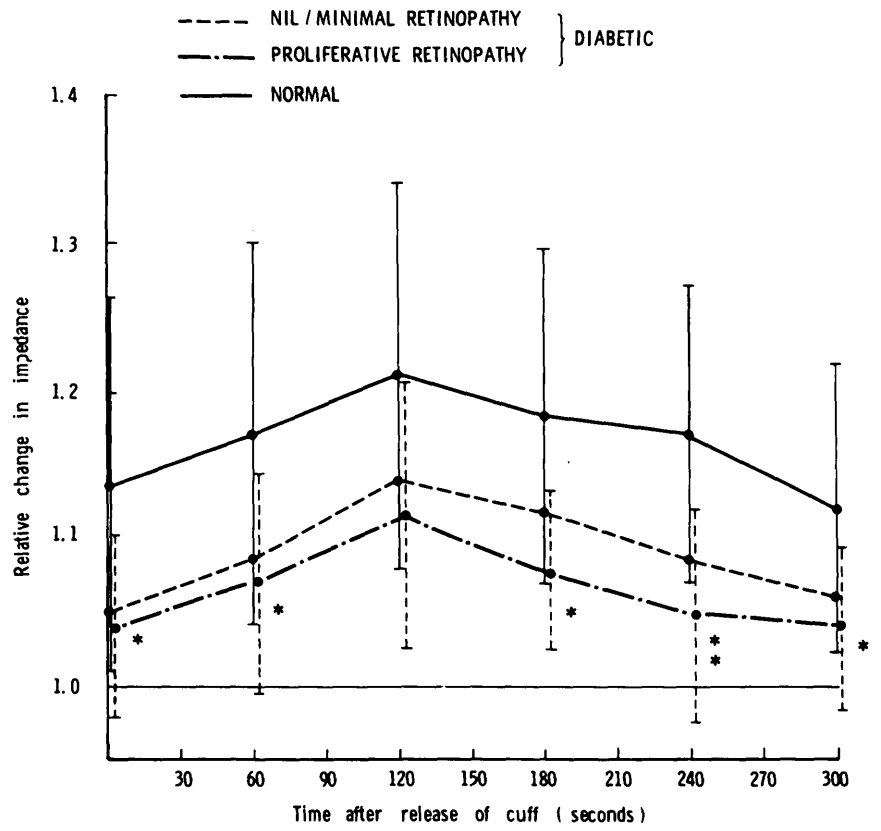

FIG. 4. *Relative increase in forearm electrical impedance after 10 min of forearm ischemia (median ± SD equivalent). Diabetic patients with proliferative retinopathy had an impaired increase in pulsatile total forearm blood flow (*$P < 0.05$, :$P < 0.01$ from normal subjects).*

previous study that examined diabetic patients without microangiopathy.[1] It appears that only a subgroup of diabetic patients exhibits a diminution of response concomitant with the development of microangiopathy. While the abnormal response is probably a manifestation of microangiopathy, we cannot exclude the possibility that patients with an innately sluggish vascular response to ischemia might be prone to develop microangiopathy. An abnormal response to ischemia, with abnormal vascular autoregulation, has been postulated as a factor in other ischemic diseases.[9]

The group with severe microangiopathy also had more severe neuropathy than the nil/minimal retinopathy group, as judged by vibration sense but not by the measurements of autonomic function. Nevertheless, a correlation between the neuropathy and the vasodilatory response suggests abnormal neural control might be a factor. In any case, having developed microangiopathy and neuropathy, the delayed vasodilation in response to ischemia might impair the recovery of an arteriole or capillary that has become blocked for any reason, including red cell rouleaux, a platelet aggregate, or a large white cell. This poor vasodilatory response might convert an otherwise temporary obstruction into pathologic capillary closure and might contribute to the inexorable progression of advanced renal diabetic microangiopathy[10] or retinopathy[11] in spite of excellent diabetes control.

The transcutaneous oxygen electrode rests close to the skin and probably reflects changes in the microcirculation of the dermis. Impedance plethysmography reflects the change in pulsatile total forearm blood flow in response to ischemia. The diabetic patients with proliferative retinopathy had a significantly smaller relative increase in pulsatile blood flow than did the nondiabetic controls, whereas those diabetic patients with nil/minimal retinopathy were not significantly different from normal.

The physiology of postischemic hyperemia is not well understood, although vasoactive endothelial cell products are probably responsible. Prostacyclin has been reported to be released after a short period of ischemia,[12] although improved assay techniques are needed. Abnormal capillary blood flow has been demonstrated in capillaries of diabetic patients,[13] and further investigation is required into the effect of diabetes on the endothelium and on the control of blood flow in the microvascular circulation.

Measurement of postischemic vasodilation offers a possible means of detecting an important subgroup with a high risk of developing microangiopathy, before disastrous diabetic complications appear. Previously, assessment of capillary fragility has been suggested as such a marker,[14] but the test has been difficult to standardize. Postischemic vasodilation is a more simple test to perform, although the effect of different ambient temperatures on the results needs to be noted. Prospective studies would be required to determine whether the test has clinical use.

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