

Photoplethysmographic Assessment of Pulse Wave Reflection

Blunted Response to Endothelium-Dependent Beta₂-Adrenergic Vasodilation in Type II Diabetes Mellitus

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- OBJECTIVES** We sought to determine whether a simple index of pressure wave reflection may be derived from the digital volume pulse (DVP) and used to examine endothelium-dependent vasodilation in patients with type II diabetes mellitus.
- BACKGROUND** The DVP exhibits a characteristic notch or inflection point that can be expressed as percent maximal DVP amplitude (IP_{DVP}). Nitrates lower IP_{DVP} , possibly by reducing pressure wave reflection. Response of IP_{DVP} to endothelium-dependent vasodilators may provide a measure of endothelial function.
- METHODS** The DVP was recorded by photoplethysmography. Albuterol (salbutamol) and glyceryl trinitrate (GTN) were administered locally by brachial artery infusion or systemically. Aortic pulse wave transit time from the root of the subclavian artery to aortic bifurcation (T_{Ao}) was measured by simultaneous Doppler velocimetry.
- RESULTS** Brachial artery infusion of drugs producing a greater than threefold increase in forearm blood flow within the infused limb was without effect on IP_{DVP} , whereas systemic administration of albuterol and GTN produced dose-dependent reductions in IP_{DVP} . The time between the first and second peak of the DVP correlated with T_{Ao} ($r = 0.75$, $n = 20$, $p < 0.0001$). The effects of albuterol but not GTN on IP_{DVP} were attenuated by N^G -monomethyl-L-arginine. The IP_{DVP} response to albuterol (400 μ g by inhalation) was blunted in patients with type II diabetes mellitus as compared with control subjects (fall $5.9 \pm 1.8\%$ vs. $11.8 \pm 1.8\%$, $n = 20$, $p < 0.02$), but that to GTN (500 μ g sublingually) was preserved (fall $18.3 \pm 1.2\%$ vs. $18.6 \pm 1.9\%$, $p = 0.88$).
- CONCLUSIONS** The IP_{DVP} is influenced by pressure wave reflection. The effects of albuterol on IP_{DVP} are mediated in part through the nitric oxide pathway and are impaired in patients with type II diabetes. (J Am Coll Cardiol 1999;34:2007-14) © 1999 by the American College of Cardiology

Transmission of infrared light through the finger is proportional to blood volume, and its measurement, "photoplethysmography," gives a digital volume pulse (DVP) (Fig. 1). The DVP exhibits a characteristic "notch" or point of inflection (IP_{DVP}) in its downslope. It has long been recognized that nitrovasodilators such as glyceryl trinitrate (GTN) produce marked changes in the pulse waveform with a reduction in IP_{DVP} (1-3). Accompanying changes in

heart rate and blood pressure are minor in comparison to those in the DVP, and IP_{DVP} has been used as a sensitive index of nitrate bioavailability (4). More recently, acetylcholine, which acts by stimulating release of nitric oxide (NO) from the endothelium, has been shown to lower the height of a similar inflection point in the photoplethysmographic waveform recorded from the rabbit ear (5). This response to acetylcholine in rabbits is antagonized by inhibition of NO synthase and is reduced in cholesterol-fed rabbits, suggesting that it may be used as a measure of endothelial function (5). In Japan the second derivative of the DVP has been widely used by Takazawa et al. (6) to assess effects of vasoactive drugs and characterize vascular aging.

Despite the well-recognized effects of vasodilator drugs and aging on the DVP, the physical properties determining its characteristics remain poorly understood. The fall in

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Abbreviations and Acronyms

DVP	= digital volume pulse
GTN	= glyceryl trinitrate
IP _{DVP}	= inflection point in digital volume pulse expressed as percent DVP amplitude
iv	= intravenous
L _{Ao}	= distance from the root of the subclavian artery to the bifurcation of the aorta
L-NMMA	= N ^G -monomethyl-L-arginine
NO	= nitric oxide
PWV _{Ao}	= aortic pulse wave velocity
sl	= sublingual
T _{Ao}	= aortic pulse wave transit time (root of subclavian artery to aortic bifurcation)
ΔT _{DVP}	= time between first and second peak of digital volume pulse

IP_{DVP} after GTN has been variously attributed to a "Windkessel" effect resulting from increased compliance of large arteries and to decreased venous return to the heart (5,7). The pressure pulse has been studied more extensively than the volume pulse (8), and changes in the pressure pulse caused by GTN are thought to result mainly from decreased pressure wave reflection (9-11). Although infrared light transmission through the finger can, in combination with a servocontrolled finger pressure cuff, be used to derive a pressure pulse (12), the DVP obtained by simple photoplethysmography and pressure pulse waveforms differ and their relation is complex (13). Changes in the DVP produced by GTN do, however, parallel those in the pressure pulse (13). Decreased pressure wave reflection might therefore account for effects of GTN on the DVP. The purpose of the present study was to investigate the factors that

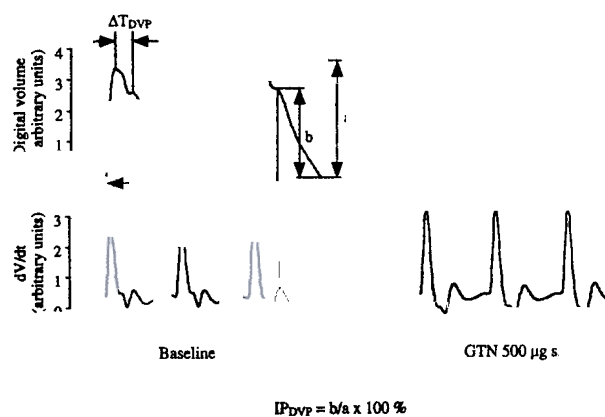


Figure 1. The digital volume pulse (DVP) and first derivative (dV/dt, lower trace) recorded before and after systemic administration of GTN (500 µg sublingually). The notch or point of inflection at height, *b*, is identified by the local maximum in the first derivative. The height of the inflection point (IP_{DVP}) is expressed as percent DVP amplitude, *a*. The IP_{DVP} falls after GTN. The time between the first and second peak of the DVP (ΔT_{DVP}) was measured in some experiments.

influence the DVP in humans and to explore the use of the DVP in detecting abnormalities in vascular reactivity in patients with type II diabetes mellitus, a group known to exhibit marked endothelial dysfunction (14-16). We measured the effects of local and systemic administration of vasodilators on the DVP, and compared the time interval between components of the DVP with aortic pulse wave transit time (T_{Ao}). Aortic pulse wave velocity (PWV_{Ao}) was calculated from T_{Ao} and aortic length. The PWV_{Ao} is an accepted measure of aortic compliance (17,18). We compared the effects of vasodilators on DVP, PWV_{Ao} and peripheral vascular resistance, and we examined the effects of altering wave reflection from the lower body by supra-systolic leg cuff inflation.

Our results suggested that the DVP comprises a direct component arising from pressure waves propagating from the heart to the finger and a delayed component arising from pressure waves reflected backward from peripheral arteries mainly in the lower body, which then propagate to the finger. In further studies IP_{DVP} was used to assess the effects of the beta₂-adrenergic agonist albuterol (salbutamol). We have previously shown that the vasodilator effects of albuterol on forearm resistance arteries are mediated, in part, through the L-arginine-NO pathway (19). To determine whether the effects of albuterol on the DVP are similarly dependent on this pathway, we performed studies in the presence and absence of an NO synthase inhibitor, N^G-monomethyl-L-arginine (L-NMMA). Finally we compared the effects of albuterol and GTN on the DVP in patients with uncomplicated type II diabetes and healthy control subjects.

METHODS

Subjects. Healthy volunteers, recruited from the local community by advertisement, were screened by physical examination and routine biochemistry. All were normotensive (office blood pressure <140/90 mm Hg) and none had total serum cholesterol values >230 mg/dl. Patients with type II diabetes were recruited from the Diabetic Clinic at St. Thomas' Hospital; they were managed by diet or by diet plus oral hypoglycemic therapy. No patients had complications other than background diabetic retinopathy, and none were receiving vasoactive drug therapy. The characteristics of the subjects participating in the sub-study of the comparison of the DVP between patients with type II diabetes and control subjects are shown in Table 1. Control subjects in this sub-study were recruited concurrently with patients with diabetes and were similar in terms of age and gender distribution. Subjects in all other studies were male volunteers. The study was approved by St. Thomas' Hospital Research Ethics Committee, and all subjects gave written, informed consent.

Photoplethysmography. A photoplethysmograph (Micro Medical, Gillingham, Kent, United Kingdom) transmitting infrared light at 940 nm was placed on the index finger of

Table 1. Characteristics of Patients With Type II Diabetes Mellitus and Control Subjects

	Control Subjects (n = 20)	Diabetics (n = 20)
Gender (M/F)	15/5	13/7
Smokers/nonsmokers	4/16	5/15
Age (yrs)	44 ± 6.9	48 ± 10
Systolic BP (mm Hg)	123 ± 17	131 ± 21
Diastolic BP (mm Hg)	71 ± 13	78 ± 12
BMI (kg/m ²)	24 ± 2.5	28 ± 4.7*
Glucose (mmol/liter)	4.8 ± 0.6	10.3 ± 5.3*
HbA _{1c} (%)	4.9 ± 0.6	7.7 ± 2.0*
Total cholesterol (mg/dl)	189 ± 31	201 ± 46
Triglycerides (mg/dl)	115 ± 71	230 ± 186
HDL cholesterol (mg/dl)	54 ± 12	46 ± 12

*p < 0.05 compared with control subjects. Data are presented as mean value ± SD.
 BP = blood pressure; BMI = body mass index; HbA_{1c} = glycosylated hemoglobin; HDL = high density lipoprotein

the right hand (except in experiments involving brachial artery infusion when bilateral measurements were made). Frequency response of the photoplethysmograph was flat to 10 Hz. Digital output from the photoplethysmograph was recorded through an analogue-to-digital converter (12 bit, sampling frequency 100 Hz). The first derivative with respect to time of the DVP signal was used to identify the notch or inflection point as the point, after the first peak of the waveform, at which the first derivative was at a local maximum. The IP_{DVP} was taken as the height of this point (b) expressed as percent amplitude of the waveform (a): IP_{DVP} = b/a × 100% (Fig. 1). The IP_{DVP} was calculated from the mean of three or more consecutive cycles of the DVP. In some experiments the time between the first and second peak of the DVP (ΔT_{DVP}) (Fig. 1) was measured. All measurements were made with the subject supine in a temperature-controlled laboratory at 26 ± 1°C. All subjects were allowed to acclimatize to this temperature for at least 30 min before recordings commenced.

Photoplethysmographic measurements during brachial artery infusion of vasodilators. Bilateral DVP and forearm blood flow measurements were made simultaneously during brachial artery administration of albuterol and GTN. The brachial artery was cannulated using a 27-gauge steel needle (Coopers Needleworks, Birmingham, United Kingdom) using <0.25 ml of 1% lidocaine as local anesthetic. Drugs diluted in 0.9% saline and saline alone were infused at 1 ml/min. Forearm blood flow was measured in both arms by venous occlusion strain gauge plethysmography (20) electrically calibrated (21). Wrist cuffs were not used so as to include the contribution (~50%) from the hand to total forearm blood flow (22). After baseline measurements during infusion of saline alone, the DVP and blood flow were measured during infusions of four cumulative doses (0.1, 0.3, 1.0 and 3.0 µg/min) of albuterol (Allen and Hanburys, United Kingdom) or four cumulative doses of GTN (0.1, 0.3, 1.0 and 3.0 µg/min) (David Bull Laboratories, Australia). Each dose was infused for 5 min with

blood flow (mean of five venous occlusions) measured during the last 2 min of each infusion period. The DVP recordings were obtained immediately after forearm blood flow measurements.

Photoplethysmographic measurements during systemic administration of vasodilators and leg cuff inflation. The DVP recordings were obtained during administration of GTN, 500 µg sublingual (sl) for 5 min and 10 to 100 µg/min intravenous (iv) and albuterol 100 to 400 µg by inhalation through a spacer or 2 to 20 µg/min iv. During sl administration of GTN, changes in IP_{DVP} were maximal between 3 and 5 min. The mean of measurements over this period was used to quantify the response to sl GTN. The response to inhaled albuterol was taken as the mean of measurements at 10 and 15 min after inhalation. This avoided artifactual responses relating to the inhalational maneuver (assessed using a placebo inhaler), which resolved within 5 min. Responses to iv administration of GTN and albuterol were measured after the attainment of steady state or when responses were maximal. In some experiments simultaneous measurements of PWV_{Ao}, derived from aortic transit times as described subsequently, were made before, during and after administration of GTN and albuterol. Changes in IP_{DVP} were also measured before and after bilateral suprasystolic leg cuff inflation.

Measurement of T_{Ao}: Comparison with ΔT_{DVP} . The T_{Ao} was measured from the "foot to foot" delay time between Doppler velocity sonograms obtained using 4-MHz continuous wave transducers (Sonicaid, BV 380, Oxford, United Kingdom). One transducer was placed in the left anterior triangle of the neck to insonate the root of the left subclavian artery and the other at the midpoint between the anterior superior iliac spines to insonate the abdominal aorta just above the aortic bifurcation. Real-time spectral analysis was used to obtain the maximal frequency envelopes of the Doppler signals and the "foot to foot" transit time between these obtained as previously described (23). The distance

from the root of the subclavian artery to the bifurcation of the aorta (L_{Ao}) was measured from surface markings (24). The PWV_{Ao} was calculated from L_{Ao}/T_{Ao} (23). This method is similar to that described by Avolio et al. (17,18). The T_{Ao} was compared with the ΔT_{DVP} . In two subjects the second peak of the DVP was not clearly defined, and in these subjects the first derivative was used to identify the time of the second peak.

Effects of L-NMMA on the DVP response to albuterol and GTN. The IP_{DVP} response to albuterol (400 μ g by inhalation) was assessed 15 min after administration of L-NMMA (3 mg/kg IV over 5 min), and on another occasion, separated by at least one week, after saline placebo in a two-phase randomized crossover study. We and other investigators have previously established that the response to this dose of L-NMMA is maximal at ~ 15 min (25,26). The response to GTN (500 μ g sl) was measured after the same dose of L-NMMA and saline placebo in the same study design. In addition to IP_{DVP} , mean arterial blood pressure (Dinamap model 1846 SX, Critikon, Florida) and cardiac output (bioimpedance cardiac output monitor: BoMed NCCOM3, BoMed Medical Manufacturing Ltd, California) were measured noninvasively using previously validated methods (27,28). Total systemic vascular resistance was estimated by dividing mean arterial pressure by cardiac output. All measurements were made with subjects supine.

DVP responses in patients with type II diabetes and control subjects. After 30 min rest supine basal measurements of IP_{DVP} , pulse rate and blood pressure were obtained at 5 min intervals for 15 min. Glyceryl trinitrate (500 μ g sl) was then administered and measurements obtained at 1-min intervals for 5 min and at 5-min intervals for 30 min, by which time all hemodynamic values had returned to baseline. Albuterol (400 μ g by inhalation through spacer) was given and further measurements were made at 5-min intervals for 20 min. Responses to GTN and albuterol were assessed as described earlier.

Statistics. Subject characteristics are presented as the mean value \pm SD. Results are presented as the mean value \pm SE. Analysis of variance for repeated measures was used to test for differences in IP_{DVP} and other hemodynamic measurements. The Mann-Whitney *U* test was used to test for differences in IP_{DVP} response between patients with diabetes and control subjects. Correlation between T_{Ao} and ΔT_{DVP} was sought using least squares regression analysis. Differences were considered significant at $p < 0.05$ (two-tailed).

RESULTS

Brachial artery infusion of vasodilators. Brachial artery infusion of albuterol (0.1 to 3 μ g/min) increased total blood flow in the infused arm from 4.9 ± 0.7 ml/min per 100 ml forearm to 24.5 ± 3.3 ml/min per 100 ml ($p < 0.001$), but had no significant effect on forearm blood flow in the

noninfused arm. Brachial artery infusion of albuterol increased the amplitude of the DVP waveform but produced no significant change in IP_{DVP} in the infused or noninfused arm. The ratio of forearm blood flow in the infused to noninfused arm increased more than threefold, while the ratio of the IP_{DVP} in the infused to noninfused arm remained constant to within 10% ($p < 0.001$ for comparison of blood flow ratio and IP_{DVP} ratio) (Fig. 2). Brachial artery infusion of GTN (0.1 to 3 μ g/min) increased forearm blood flow in the infused arm from 9.8 ± 2.0 to 22.4 ± 2.4 ml/min per 100 ml ($p < 0.01$) but had no significant effect on forearm blood flow in the noninfused arm. Brachial artery infusion of GTN was associated with a small but significant fall in IP_{DVP} of the infused arm ($60 \pm 5.5\%$ to $53 \pm 5.8\%$, $p < 0.05$), but IP_{DVP} of the noninfused arm fell to a similar degree ($63 \pm 5.1\%$ to $49 \pm 6.3\%$) so that the ratio of the IP_{DVP} in the infused to noninfused arm remained constant ($p < 0.001$ for comparison of blood flow ratio and IP_{DVP} ratio) (Fig. 2).

Systemic administration of vasodilators and leg cuff inflation. Albuterol (100 to 400 μ g by inhalation and 25 to 100 μ g iv bolus and 2 to 20 μ g/min iv) and GTN (500 μ g sl and 10 to 100 μ g/min iv) reduced IP_{DVP} (Fig. 3). Changes from baseline in IP_{DVP} and in systemic hemodynamic values after albuterol (400 μ g by inhalation) and GTN (500 μ g sl) in 10 healthy volunteers are shown in Figure 4. Reductions in IP_{DVP} of $16.8 \pm 3.2\%$ ($26.2 \pm 5.7\%$ change from baseline) and $24.0 \pm 1.9\%$ ($40.4 \pm 4.2\%$ change from baseline) after albuterol and GTN, respectively, were accompanied by relatively minor changes in heart rate (increase of 12.9 ± 2.9 and 6.4 ± 2.3 beats/min for albuterol and GTN, respectively, each $p < 0.05$) and blood pressure (changes in systolic blood pressure of 5.1 ± 2.7 and -4.0 ± 3.0 mm Hg, each $p = NS$, and decreases in diastolic blood pressure of 3.2 ± 1.8 mm Hg [$p = NS$] and 4.9 ± 1.6 mm Hg [$p < 0.05$] for albuterol and GTN, respectively). Systemic vascular resistance fell by $26 \pm 3.6\%$ and $16 \pm 3.4\%$ and cardiac output increased by $30 \pm 5.5\%$ and $8.2 \pm 3.1\%$ (each $p < 0.05$) after albuterol and GTN, respectively. In experiments where PWV_{Ao} and IP_{DVP} were

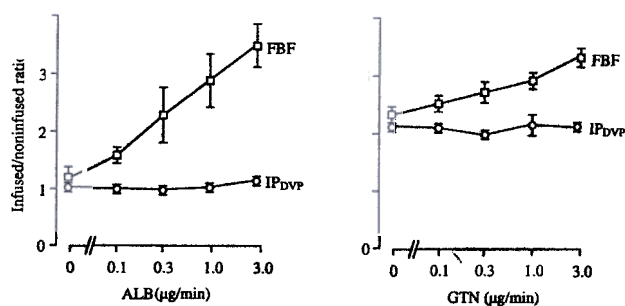


Figure 2. Ratios of forearm blood flow (FBF, open squares) and inflection point of the digital volume pulse (IP_{DVP} , circles) in the infused-noninfused arms during brachial artery infusion of albuterol (ALB, n = 5) and glyceryl trinitrate (GTN, n = 5).

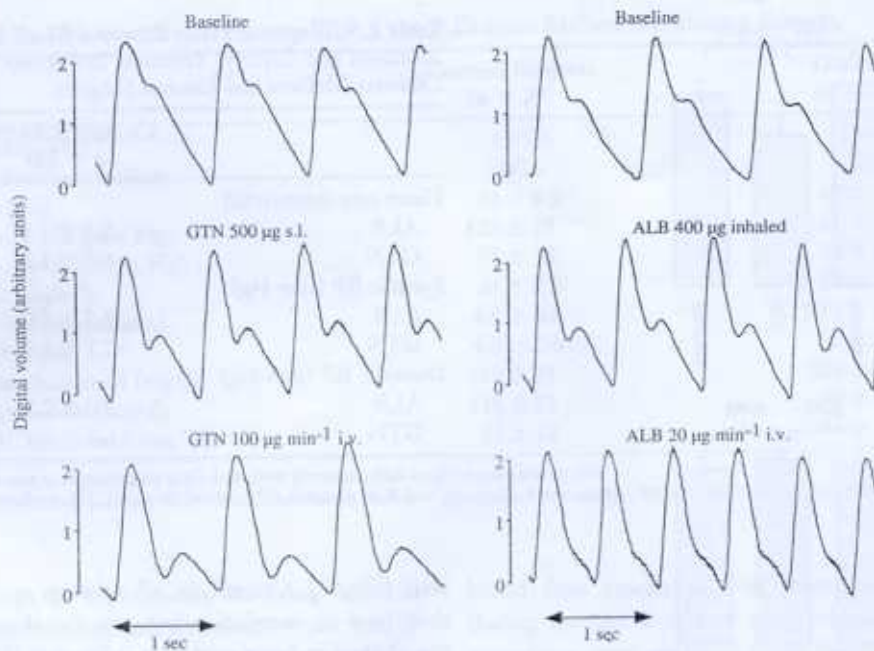


Figure 3. Typical DVP traces showing responses to sublingual (s.l.) and intravenous (i.v.) and to inhaled and i.v. albuterol (ALB).

measured simultaneously, changes in IP_{DVP} after administration of albuterol or GTN were not accompanied by significant changes in PWV_{Ao} (Fig. 5). Bilateral suprasystolic leg cuff inflation increased IP_{DVP} by $13.3 \pm 2.5\%$ ($n = 9$, $p < 0.01$).

Comparison of ΔT_{DVP} with T_{Ao} . The ΔT_{DVP} was correlated with T_{Ao} —the delay between the foot of the velocity sonogram at the subclavian artery and that at the aortic bifurcation ($r = 0.75$, $n = 20$, $p < 0.0001$) (Fig. 6). The slope of the regression line was 3.9 ± 0.81 .

Effect of L-NMMA on DVP response to albuterol and GTN. Changes in IP_{DVP} in response to albuterol were less when albuterol was administered after L-NMMA as compared with saline placebo ($5.4 \pm 7.5\%$ change from baseline for albuterol after L-NMMA vs. $26 \pm 5.7\%$ change from baseline for albuterol after saline, $n = 10$, $p < 0.01$) (Fig. 4). In contrast, changes in IP_{DVP} after GTN were similar after L-NMMA and saline placebo ($37.2 \pm 6.4\%$ change from baseline for GTN after L-NMMA vs. $40.4 \pm 4.2\%$ change from baseline for GTN after saline, $n = 10$, $p = 0.68$) (Fig. 4). The increase in cardiac output after both GTN and albuterol was significantly less after L-NMMA than after saline placebo (each $p < 0.05$). Changes in other hemodynamic values after albuterol and GTN did not differ significantly according to whether L-NMMA or saline had been administered, although there was a tendency for the fall in systemic vascular resistance after albuterol to be less after L-NMMA than after saline placebo ($p = 0.09$).

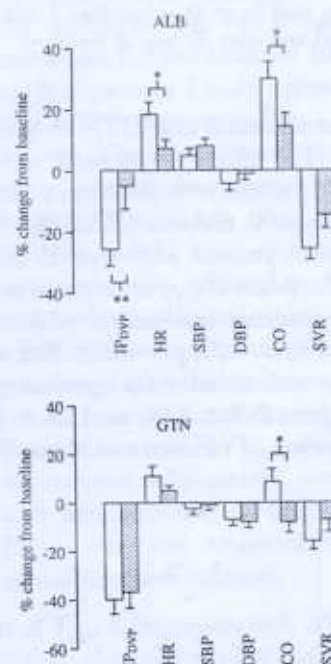


Figure 4. Changes from baseline in hemodynamic measurements after GTN ($n = 10$) and albuterol (ALB, $n = 10$) after saline placebo or L-NMMA administered 15 min before GTN/ALB. CO = cardiac output; DBP = diastolic blood pressure; HR = heart rate; IP_{DVP} = height of inflection point of DVP measured as percent amplitude; SBP = systolic blood pressure; SVR = systemic vascular resistance. Percent change from baseline of the IP_{DVP} refers to change in percent units (e.g., fall from 80% to 60% = $[80-60]/80 = 25\%$). * $p < 0.05$ for L-NMMA vs. saline. ** $p < 0.01$ for L-NMMA vs. saline. Open box = saline; dotted box = L-NMMA.

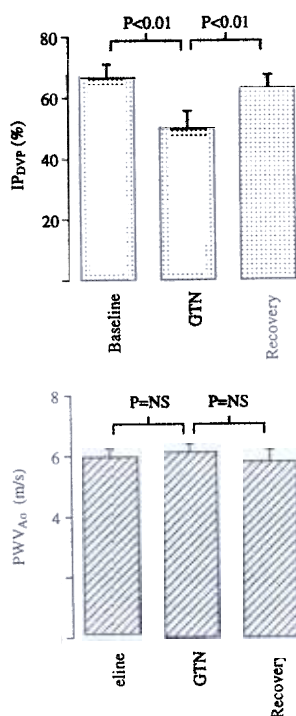


Figure 5. Height of the inflection point of the digital volume pulse relative to the amplitude (IP_{DVP}) and aortic pulse wave velocity (PWV_{Ao}) in healthy men ($n = 5$) at baseline, 5 min after GTN ($500 \mu\text{g}$ sublingually) and after 20 min of recovery.

DVP responses to albuterol and GTN in patients with type II diabetes. The effects of albuterol and GTN on heart rate and blood pressure were similar in control subjects and patients with type II diabetes (Table 2). At baseline IP_{DVP} was similar in patients with type II diabetes and control subjects. The fall in IP_{DVP} in response to GTN was similar in diabetic patients and control subjects ($18.3 \pm 1.2\%$ vs. $18.6 \pm 1.9\%$, $n = 20$, $p = 0.88$). The response to albuterol in patients with diabetes was significantly less than that in control subjects ($5.9 \pm 1.8\%$ vs. $11.8 \pm 1.8\%$, $n = 20$, $p < 0.02$ by analysis of variance and Mann-Whitney U

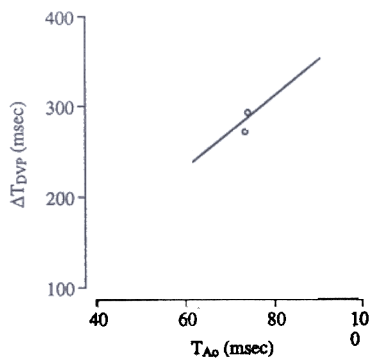


Figure 6. Correlation between the time from the first to second peak of the digital volume pulse (ΔT_{DVP}) and pressure wave transit time from the root of the subclavian artery to the aortic bifurcation (T_{Ao}) in 20 healthy men ($r = 0.75$, $p < 0.0001$).

Table 2. Changes in Heart Rate and Blood Pressure After Albuterol and Glyceryl Trinitrate in Patients With Type II Diabetes Mellitus and Control Subjects

	Control Subjects (n = 20)	Diabetics (n = 20)
Heart rate (beats/min)		
ALB	$9.8 \pm 1.6^*$	$7.1 \pm 1.2^*$
GTN	$6.5 \pm 1.2^*$	$5.6 \pm 1.2^*$
Systolic BP (mm Hg)		
ALB	0.3 ± 3.0	1.6 ± 3.1
GTN	-2.7 ± 1.9	-5.4 ± 2.1
Diastolic BP (mm Hg)		
ALB	-2.0 ± 2.3	-6.4 ± 2.6
GTN	$-6.1 \pm 1.4^*$	$-6.7 \pm 1.4^*$

* $p < 0.05$ compared with zero. Data are presented as mean value \pm SD.
ALB = albuterol; BP = blood pressure; GTN = glyceryl trinitrate.

test) (Fig. 7). Overall, in all subjects and for both drugs, there was no correlation between the change in IP_{DVP} and the change in heart rate ($r = 0.26$, $p = 0.24$). At the doses used, GTN produced a greater fall in IP_{DVP} than did albuterol ($p < 0.0001$), despite a less marked increase in heart rate.

DISCUSSION

Photoplethysmography. Photoplethysmography provides a simple means for deriving the DVP. Although widely used as a means for displaying the pulse (for example, in pulse oximetry), its use to study the detail of the shape of the waveform itself has received surprisingly little attention. This may be because the peripheral site of recording and the difference between the DVP and arterial pressure waveform give the misleading impression that the DVP is influenced mainly by local factors. However, Takazawa et al. (6) have shown that the second derivative of the DVP may be used to

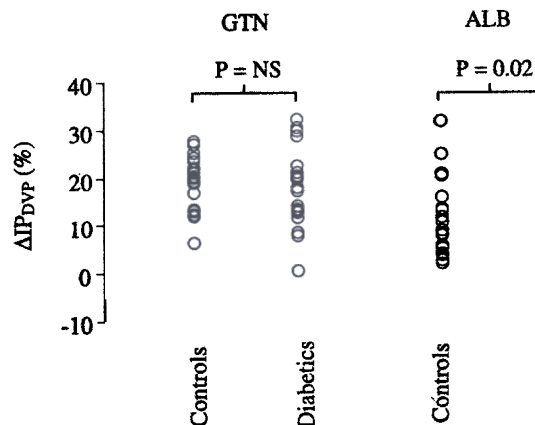


Figure 7. Decrease from baseline in the height of the inflection point of the digital volume pulse (IP_{DVP}) after GTN ($500 \mu\text{g}$ sublingually) and albuterol (ALB, $400 \mu\text{g}$ by inhalation through spacer) in patients with type II diabetes ($n = 20$) and control subjects ($n = 20$).

infer changes in the systemic circulation relating to the effects of drugs and aging. In the present study, we have focused on IP_{DVP} —the relative height of the inflection point separating the systolic and diastolic components of the DVP.

Lack of effect of local vasodilation on IP_{DVP} . We found that direct infusion of vasodilators into the brachial artery sufficient to increase total forearm blood flow more than threefold had little or no effect on IP_{DVP} . The highest dose of GTN was associated with a small but significant effect on the IP_{DVP} , but this was similar in the infused and noninfused arms, suggesting that it was due to a systemic rather than local effect. In contrast, systemic administration of GTN produced a profound change in IP_{DVP} while having no significant effect on forearm blood flow. These observations effectively exclude local circulatory changes in the arm or hand (which contributes ~50% to forearm blood flow [22]) as responsible for effects of systemic administration of albuterol or GTN on IP_{DVP} in normal subjects at an ambient temperature of 26°C.

Influence of wave reflection. Of the various explanations that have been suggested to account for the IP_{DVP} that which best fits our observations is that the DVP is determined by direct and reflected pressure waves. Reflected waves arising mainly from the lower body are delayed relative to the direct wave, and therefore produce an inflection point or second peak in the DVP. O'Rourke et al. (8,9) have previously suggested that systemic administration of GTN reduces pressure wave reflection, and this is consistent with the reduction of IP_{DVP} seen after systemic but not local administration of GTN in the present study. Suprasystolic pressure cuff inflation around the thighs would be expected to increase pressure wave reflection from the legs, and indeed this was accompanied by an increase in IP_{DVP} , supporting the concept of the IP_{DVP} being influenced by wave reflection. Further evidence of wave reflection determining the characteristics of the DVP arises from the correlation between ΔT_{DVP} and T_{Ao} : if the second peak of the DVP is caused by pressure waves reflected from peripheral arteries, then the ΔT_{DVP} of the DVP waveform would be expected to be related to the time taken for pressure waves to pass from the heart to the "site of reflection" and back to the heart (the transit times for pressure waves to pass from the heart to the subclavian artery and from the subclavian artery along the arm to the finger being common for both the direct and reflected waves). We observed a strong correlation between ΔT_{DVP} and the propagation time of pressure waves along the aorta from the T_{Ao} . This correlation again supports the concept of wave reflection as a major determinant of IP_{DVP} . Reflections from many sites within the vascular tree are likely to contribute to the reflected wave seen at the periphery, resulting in temporal spread of the reflected wave. The time between the peak of the direct wave and the peak of the reflected wave cannot, therefore, be used to define precisely the time taken for pressure waves to pass from sites of reflection to the upper limb, because there are multiple such sites and because such timing information would need to be inferred

from the foot to foot delay between direct and reflected waves. Our observation that ΔT_{DVP} is approximately four times the T_{Ao} is nevertheless compatible with the suggestion by Yaginuma et al. (10), Latson et al. (29) and others (8) that wave reflection occurs predominantly from small arteries in the trunk and lower limbs. Because large changes in DVP pulse are observed in response to GTN in the absence of large changes in systemic vascular resistance, such arteries must be proximal to resistance vessels.

Influence on IP_{DVP} of systolic ejection time, heart rate and pulse wave velocity. Systolic ejection time and heart rate are likely to influence IP_{DVP} (8). However, in the present study we found no correlation between the change in IP_{DVP} and the change in heart rate. Furthermore, compared with albuterol, GTN produced less increase in heart rate and cardiac output but a greater fall in IP_{DVP} . This suggests that the effects of these drugs on IP_{DVP} occur independently of any effect on heart rate. A change in IP_{DVP} could occur as a result of vasodilation of the arteries, which contributes most to wave reflection, thus reducing the reflected waves as well as IP_{DVP} . Alternatively, decreased aortic pulse wave velocity, resulting from increased aortic and large artery compliance (17,18), could delay arrival of the reflected wave relative to the direct wave, increasing ΔT_{DVP} and hence reducing IP_{DVP} . To distinguish between these possibilities we made simultaneous measurements of PWV_{Ao} and IP_{DVP} during administration of vasodilators. We found that changes in IP_{DVP} were not accompanied by changes in PWV_{Ao} . This is consistent with the observations of Yaginuma et al. (10), who found GTN to have no effect on the timing of vascular reflections. This suggests that during vasodilator therapy, a reduction in IP_{DVP} is due mainly to dilation of small arteries reducing wave reflection from the lower body.

Effect of albuterol on IP_{DVP} as a test of endothelial function. Vasodilator effects of GTN are mediated through its metabolism in vascular smooth muscle to NO or a nitrosothiol (30). Vasodilators that stimulate NO release from the endothelium through the L-arginine-NO pathway might therefore be expected to have a similar effect on IP_{DVP} to GTN. In the present study we found that albuterol produced a marked change in IP_{DVP} . We and other investigators have previously shown that beta-adrenergic agonists—albuterol, in particular—produce vasodilation in resistance arteries, which is dependent on the L-arginine-NO pathway (19,31). To determine whether the effects of albuterol on the IP_{DVP} are mediated through the L-arginine-NO pathway, we examined responses to albuterol in the presence and absence of L-NMMA. We also examined the effects of L-NMMA on GTN as a control study. L-NMMA blunted the effect of albuterol on IP_{DVP} but did not influence the effect of GTN. This suggests that the effect of albuterol on IP_{DVP} is mediated at least in part through the L-arginine-NO pathway.

The finding that the IP_{DVP} response to albuterol depends

on the endothelial L-arginine-NO pathway raises the possibility that this response may be used to examine the integrity of this pathway in conditions associated with endothelial dysfunction. To investigate this, we examined the response to albuterol (and GTN as an endothelium-independent control) in patients with type II diabetes. We chose this group because there is evidence of marked impairment of endothelium-dependent vasodilation in such patients (14-16). We found that responses to albuterol in type II diabetic patients are indeed blunted relative to nondiabetic control subjects, whereas responses to GTN are preserved, consistent with a defect in the endothelial L-arginine-NO pathway in type II diabetes. These findings therefore suggest that the IP_{DVP} response to albuterol may be used as a simple test of endothelial function. Its application for this will require further validation, however.

Conclusions. Our results suggest that IP_{DVP} is influenced by wave reflection from the lower body. Glyceryl trinitrate and albuterol lower IP_{DVP} through vasodilation of arteries in the lower body. The effects of albuterol are mediated in part through the L-arginine-NO pathway and are impaired in patients with type II diabetes. Photoplethysmographic assessment of the DVP may provide a useful method for examining vascular reactivity.

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