Frequency-response characteristics of vascular resistance vessels

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METHODS

General Considerations

In order to determine the transfer characteristics of a system component it is necessary to know the input and the corresponding output (7). In the case of the resistance vessels there are many inputs (11) and this investigation deals with one of these, the sympathetic vasoconstrictor nerve activity. The output studied was the change in resistance to blood flow, and in our preparation this was determined by the changes in perfusion pressure under conditions of constant flow.

Anesthesia

Sixteen mongrel dogs weighing 10–20 kg were premedicated with intramuscular morphine sulphate 15 mg, and 1 hr later anesthetized with sodium thiopental (20 mg/kg) or sodium pentobarbionate (30 mg/kg). After endotracheal intubation ventilation was maintained with an intermittent positive-pressure respirator delivering pure oxygen. Additional anesthesia was given as required to maintain a light surgical plane of anesthesia.

Vascular Isolation of the Hindlimb

In three experiments on larger dogs, complete vascular isolation of the right hindlimb was achieved by dissection of the aorta, common iliac and external iliac arteries extending from the origin of the inferior mesenteric artery down to the inguinal ligament on both sides, with ligation of all arterial branches, and of both external iliac arteries. Perfusion of the isolated hindlimb was achieved through a cannula placed in the right femoral artery. Venous blood from the limb was collected from the femoral vein and flowed by gravitational drainage to the input of a small disposable bag oxygenator (type U330, Travenol Laboratories Inc., Morton Grove, Ill.) which had been previously primed with 5 % dextrose and blood obtained from the experimental animal. Venous blood from the limb was collected from the femoral vein and flowed by gravitational drainage to the input of a small disposable bag oxygenator (type U330, Travenol Laboratories Inc., Morton Grove, Ill.) which had been previously primed with 5 % dextrose and blood obtained from the experimental animal. The bag was oxygenated at a rate of 10 liters/min. A metal cannula inserted in a retrograde direction into the lower end of the aorta served to prime the oxygenator and as a source of blood to maintain the oxygenator blood level. Outflow from the oxygenator was used to perfuse the
hindlimb via a peristaltic pump (Sigmamotor) and heat exchanger at 37 C, the flow rate being set to maintain the mean hindlimb perfusion pressure at systemic level and this rate was not altered throughout the experiment. The pump had previously been tested for constancy of flow and found to maintain this within ±5% over an input pressure range of 20–200 mm Hg and an output range of 0–300 mm Hg. The time constant of the pump and arterial perfusion line system had been found to be less than 100 msec for a step increase in outflow pressure.

Because of the artificial nature of the perfusion and isolation procedure, and the associated instability in the experimental preparation, in 13 animals a less complex method of perfusion was used. The right carotid artery was dissected in the neck and a polyethylene cannula, as large as possible, inserted through the vessel into the arch of the aorta. This cannula served as the inflow to the pump and outflow was passed via a metal cannula to the femoral artery of the test limb. This femoral artery had previously been dissected for a distance of 2 inches below the inguinal ligament and all exposed branches had been ligated.

Neural Isolation and Stimulation

The sympathetic trunk on the side of the test limb was exposed in the lumbar region by a retroperitoneal approach and all visible grey and white rami were divided. Bipolar platinum electrodes, 3 mm apart, were applied at approximately the level of the fourth lumbar vertebra and the trunk was crushed or divided proximally. In order to ensure complete isolation of the hindlimb, in six experiments the spinal cord was divided in one or two interspaces between the 12th thoracic and 3rd lumbar vertebrae by passing a pointed scalpel blade through the intervertebral disc. In these experiments the opposite sympathetic trunk was ablated at the level of the spinal transection. In three experiments spinal anesthesia was produced immediately after the general anesthetic by the injection of 5 ml of cinchocaine HCl (Nupercaine, Ciba Ltd.) into the lumbar spinal theca followed by the posturing of the animal for 5 min in the head-up and then in the head-down position. In two experiments bilateral adrenalectomy was performed to prevent liberation of adrenal catecholamines during sympathetic stimulation.

The lumbar sympathetic trunk was stimulated with a Tektronix type 160 waveform and pulse generator, the output of which was monitored on an oscilloscope. The pulse duration was 1–5 msec and stimulus voltage was adjusted just to give maximum rise in perfusion pressure, approximately 3 v. The stimulation pattern was of impulses modulated either by a sine or a step function.

Step-function modulation. In 16 dogs step-function stimuli were obtained by delivering a train of impulses at a known rate of between 0.1 and 20 stimuli/sec for a fixed duration sufficient in most cases to produce a maximal steady-state response.

An interval of 4–5 min was allowed between stimulation periods to permit full recovery of the preparation.

Systemic arterial pressure was measured through a polyethylene cannula inserted into the aorta either through the contralateral femoral artery or a carotid artery. Perfusion pressure in the hindlimb was measured through a T connector in the distal end of the perfusion line. Both pressures were recorded by electromanometers (Statham type P 23 Db), the outputs of which were applied directly to the galvanometers of a direct photographic recorder (CEC type 5124). The individual nerve stimuli and the sine-wave modulation envelope were also recorded. All animals received heparin (Evans Medical Ltd.), 2 mg/kg, prior to cannulation, and atropine (Bull Laboratories Ltd.), 0.1–0.2 mg/kg iv, before commencing the experiment in order to block the sympathetic vasodilator fibers (4).

In all experiments adequate vascular and neurological isolation was regarded as having been achieved if the degree of back bleeding when cannulating the femoral artery was small (1–2 ml bleed/min), and if the rise in systemic systolic or diastolic blood pressure with sympa-
Dynamic response of resistance vessels

Effect of sympathetic stimulation at a frequency of 0.1 stimulus/sec on hindlimb perfusion pressure. Sympathetic stimulation (upper), hindlimb perfusion pressure (middle), and systemic arterial pressure (lower) are shown.

sympathetic stimulation was less than 10 mm Hg. The responses of all the experimental preparations studied were similar, and will be considered together in the presentation of results.

Results

In all experiments sympathetic stimulation produced an increase in perfusion pressure in the isolated limb. The maximum pressure increase in each experiment ranged from 60 to 190 mm Hg with an over-all mean of 105 mm Hg. Since the mean control pressure after crushing the sympathetic chain was 75 mm Hg, the limb resistance increased on stimulation by an average factor of 1.4. The maximum response was reached at a stimulus-repetition rate of between 3 and 20 stimuli/sec. For stimulation rates between 0.1 stimulus/sec and that for the maximum response there was a direct relationship between the logarithm of the rate of stimulation and the percent of maximum increase of perfusion pressure over the control level (Fig. 1). These data show that with a stimulation rate as low as 1.0 stimulus/sec the average rise in perfusion pressure achieved was 55% of the maximum response. At stimulus frequencies above that required for maximum response, the response frequently diminished in amplitude. At very low stimulation frequencies the response to individual stimuli could be discerned (Fig. 2).

Form of Response to Step-Function Stimulus

When a stimulus of constant frequency was applied to the sympathetic trunk, after a short interval the perfusion pressure rose to a plateau sometimes with a slight overshoot. On cessation of stimulation there was a short pause before the perfusion pressure fell to the control level sometimes overshooting this to a variable degree. The delay between the onset of stimulation and a detectable rise in perfusion pressure (on delay) varied between 0.2 and 2.0 sec with a modal value of 0.5 sec. The shorter times were associated with higher frequencies of stimulation. The rise of perfusion pressure (rising phase) was approximately exponential. In some experiments at low repetition rates the initial gradient was less, and at rates in excess of 3.0 stimuli/sec was greater than could be seen with a true exponential response (Fig. 3). In three animals overshoot occurred. Assuming these curves to be exponential, the time constants varied between 3 and 15 sec with a mean value of 9 sec. At higher stimulation rates this time constant was shorter than at lower rates (Fig. 3).

After the cessation of stimulation the pressure in the limb remained the same or, if it had been rising at this time, continued to rise. This period (the off delay) lasted for 2–3 sec at stimulation rates faster than 0.5 stimulus/sec and longer at stimulation rates slower than 0.5 stimulus/sec when it reached a maximum of 5 sec. Following this phase the pressure fell to the control level, sometimes with overshoot and further oscillations (Fig. 4). When overshoot did not occur, the fall was approximately exponential and its time constant was slightly longer than that of the rise, varying from 7 to 16 sec at stimulation rates of slower than 10 stimuli/sec and slightly longer at faster rates; whereas at rates in excess of 20 stimuli/sec, the time constant was prolonged up to 30 sec in some experiments.

In those experiments in which overshoot occurred the rate of fall and extent of overshoot varied from experiment to experiment. In addition, the overshoot was greatest when the rate of fall was fastest and both in-
creased with increasing stimulus duration, reaching 30% in some experiments (Fig. 5). The overshoot and oscillation in perfusion pressure resemble the responses seen in a second-order physical system; and, as an approximation ignoring nonlinearity, the form of the response can be defined by a damping ratio and natural frequency. The damping ratio can be obtained from the ratio of overshoot to fall in perfusion pressure by the use of standard tables (7) and is charted on the ordinate of Fig. 5 along with the overshoot. Knowing the value of the damping ratio and the time taken to reach the minimum, the natural frequency of the system can be derived (7). In our experiments this frequency varied from 0.060 to 0.025 cycle/set with a mean of 0.034 cycle/set.

Sine-function modulation. When the stimulation repetition rate was modulated sinusoidally over a narrow fixed range the perfusion pressure closely followed the sinusoidal modulation envelope; however, when the range of modulation was increased above 2 stimuli/sec the pressure change became distorted, usually with a rise slower than the fall. At low modulation frequencies the correspondence between stimulus frequency and perfusion pressure was good but, as the modulation frequency was increased, the response waveform was relatively delayed, and usually smaller in amplitude (Fig. 6). These relationships are expressed in the form of a plot of amplitude and phase angle of the response vs. modulation frequency (Bode plot (7)) and an example is shown in Fig. 7. The amplitude is expressed in decibels relative to the step-function response over the same stimulus range and was calculated from the formula:

$$\text{db} = 20 \log \left( \frac{A}{B} \right)$$

in which db is the decibel value, A is the amplitude of oscillatory response to sine modulation, and B is the amplitude of step-function response over same stimulus-frequency range. The phase angle expressed in degrees is given by the formula:

$$\Phi = 360 \frac{D}{I},$$

in which $\Phi$ is the phase angle, D is the delay of perfusion-pressure waveform relative to modulation envelope, and I is the peak-to-peak interval of modulation envelope. As the modulation frequency was increased the amplitude of the response was maintained until, at a particular frequency, it began to decrease with increasing rapidity towards a rate of 20–40 db/decade. The intersection of the projection of this line and the 0-db line determines the corner frequency, and in our experiments this ranged from 0.011 to 0.020 cycle/sec with a mean of 0.017 cycle/sec. In three experiments a rise in amplitude of the response of 2–3 db occurred in the region of the corner frequency.

The phase lag increased to between 70° and 200° at the maximum modulation frequencies at which individual oscillations could be discerned. This phase lag can be regarded as being the sum of the 'on delay' and 'off delay' of the on and off responses and the times of the rising and falling phases. The phase lag of the on delay and off delay can be subtracted to give the phase lag due to the remaining components of the response (7). When this was done the phase angle...
TABLE 1. *Summary of Bode plots for all experimental data*

<table>
<thead>
<tr>
<th>Replication No.</th>
<th>Stimulus Range, stim/min</th>
<th>Corner Frequency, cycle/sec</th>
<th>Gain at Corner Frequency, dB</th>
<th>Loss at High Frequency, db/decade</th>
<th>Maximum Phase Lag, deg</th>
<th>Approximate Phase Lag Asymptote (Corrected for Dead Time), deg</th>
</tr>
</thead>
<tbody>
<tr>
<td>9</td>
<td>0.5-2.0</td>
<td>0.016</td>
<td>-2.0</td>
<td>40</td>
<td>180</td>
<td>70</td>
</tr>
<tr>
<td>10 L</td>
<td>0.5-2.0</td>
<td>0.020</td>
<td>0</td>
<td>40</td>
<td>180</td>
<td>70</td>
</tr>
<tr>
<td>10 R</td>
<td>0.5-2.0</td>
<td>0.023</td>
<td>+2.0</td>
<td>40</td>
<td>180</td>
<td>70</td>
</tr>
<tr>
<td>11</td>
<td>0.5-2.0</td>
<td>0.016</td>
<td>-2.0</td>
<td>40</td>
<td>180</td>
<td>70</td>
</tr>
<tr>
<td>13</td>
<td>1.0-2.0</td>
<td>0.017</td>
<td>+3.5</td>
<td>40</td>
<td>70</td>
<td>90</td>
</tr>
<tr>
<td>14</td>
<td>2.0-3.0</td>
<td>0.016</td>
<td>+3.5</td>
<td>40</td>
<td>70</td>
<td>90</td>
</tr>
<tr>
<td>15</td>
<td>1.0-2.0</td>
<td>0.013</td>
<td>-2.0</td>
<td>40</td>
<td>180</td>
<td>70</td>
</tr>
</tbody>
</table>

Approached an asymptote of approximately 90° in four experiments and 180° in seven experiments (Table 1). This occurred at frequencies in excess of 0.04 cycle/sec in all experiments.

DISCUSSION

Methodological Considerations

The validity of these results is to some extent dependent on adequate vascular and neurologic isolation of the perfused limb. If adequate vascular isolation is not present the test bed can be shunted by vessels of unknown characteristics and the response altered. In three of the experiments great care was taken to isolate the hindlimb vasculature and in the remaining experiments those with significant backflow were discarded. The results of the two groups are similar and we conclude that vascular isolation was adequate and shunting insignificant. If the nerve supply to the hindlimb had not been satisfactorily isolated, interchange between the stimulated sympathetic trunk and the vasomotor control areas would have distorted the responses, either as a result of direct nervous activity or of the release of adrenal catecholamines. In most experiments this was avoided by dissection and isolation of the sympathetic trunk and in some experiments by additional spinal transection, spinal anesthesia, and adrenalectomy. In addition we eliminated those experimental preparations in which a significant rise in systemic arterial pressure occurred on stimulation.

Experimental Results

These are in keeping with earlier studies on the steady-state response (2, 5, 8), step-function response, (3, 5, 6, 14) and sinusoidal response (14) of the various vascular beds. They indicate that this system is nonlinear, as shown by the steady-state stimulus frequency-resistance plot, the absence of a sinusoidal output on sinusoidal modulated stimulation, and failure of the Bode plots to conform to the patterns seen in linear first- or second-order systems (7). In addition the system is time dependent, as demonstrated by the difference between the on and off response and the effect of stimulus duration on the amplitude of the overshoot. Despite these limitations some quantitative conclusions can be drawn about the vessel response characteristics.

1) The response of the system is slow; apart from an initial on delay of approximately 0.5 sec there is a long rise period, 9.0 sec being required before 63% of the maximum response is obtained (corresponding to the time constant). After cessation of the stimulation there is an off delay of approximately 2–5 sec before the fall in resistance, which is slightly slower than the rise. This slowness of response is associated with limited frequency response of the system, as illustrated by the effect of sinusoidal forcing. Under these conditions, as the modulation frequency rises above the corner frequency of approximately 0.017 cycle/sec, the response of the system falls off rapidly. If nonlinearities are ignored the time constant of the step function response and the corner frequency of the sine-function response can be related by the formula: \( f = \frac{1}{2\pi r} \), in which \( f \) cycle/sec is the corner frequency and \( r \) sec is the time constant of step-function response. For the average rise-time constant of 9 sec the calculated corner frequency is 0.017 cycle/sec which corresponds well with the frequency of 0.017 cycle/sec obtained from the sinusoidal modulation experiments. 2) The presence of overshoot, in responses to a change in state, would indicate that the system has a discernable natural frequency. This is further illustrated by the rise in amplitude of the response at the corner frequency in three of the studies in which the input was sinusoidally modulated. This characterizes the system as at least a second-order system and this is supported by the approach of the phase lag (corrected for the on delay and off delay) towards 180° in most experiments.

These experiments provide some insight into the mechanism of vasoconstriction in the peripheral vascular beds. The on delay, the gradual rise in resistance following the onset of stimulation, the off delay and the slow fall in resistance following cessation of stimulation can be explained by the release of norepinephrine from the nerve ending, its diffusion to the vascular muscle, and its subsequent effect.

However, such considerations do not explain the presence of overshoot. This overshoot following cessation of stimulation has been described as poststimulation hyperemia (6) and has features in common with reactive hyperemia (8). This phenomenon can best be explained by the presence of a control system in the skeletal muscle bed, which acts to maintain peripheral perfusion pressure constant. Such a mechanism when disturbed by the rise in perfusion pressure following sympathetic stimulation would tend to compensate by producing vasodilatation in the resistance vessels of the bed, and this would be unmasked after the cessation of stimulation. This mechanism is similar to the feedback description of autoregulation suggested by Green (8) and our experiments indicate that the mechanism is slow in operation.
the amplitude of the overshoot being determined by the stimulus duration.

Significance of These Findings in Over-all Circulatory Regulation

The presence of the resistance vessels in skeletal muscle with their comparatively slow response rate and natural frequency of about 0.034 cycle/sec elucidate two of the characteristics of blood pressure control.

It has been established that the systemic arterial pressure is regulated by a feedback control system involving the baroreceptors, heart, and peripheral resistance vessels (10, 11). This system is comparatively stable and has a natural frequency of approximately 0.038 cycle/sec (1, 12). The similarity between the natural frequency of the resistance vessels and the overall blood pressure control system suggests that the characteristics of the vessels may in fact be responsible for this frequency, since a peak in the response of one component would tend to control the natural frequency of the whole system (7).

REFERENCES


