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INDICATOR-DILUTION MEASUREMENT OF BLOOD FLOW IN THE UPPER EXTREMITY OF MAN: DEVELOPMENT AND USE OF A METHOD IMPROVING MIXING OF INDICATOR AND VASOACTIVE AGENTS WITH BRACHIAL ARTERIAL BLOOD

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CHAPTER I

INTRODUCTION

The investigator may calculate resistance to blood flow through the vascular bed of a limb, if he simultaneously measures blood flow rate through the bed and blood pressure gradient across the bed. On this basis, he may make inferences with a fair degree of certainty about the net state of activity of the vascular smooth muscle in the bed, blood viscosity and transmural pressure remaining constant. Vasoactive substances may be introduced into the blood stream of the arteries supplying the vascular bed, and responding changes in the activity of the vascular smooth muscle downstream may be inferred from changes in local vascular resistance. By these means the investigator may study normal and abnormal peripheral vascular physiology in animals and in man.

Measurement of blood flow in the vascular beds of man must of necessity be indirect and subject to a high degree of error. Because it is difficult or impossible to verify these indirect measurements by

direct measurements of blood flow, several techniques of measurement have been developed, each of which has its own advantages and disadvantages. Perhaps the best known and currently most widely used indirect technique for determination of blood flow in the limbs of man is venous occlusion plethysmography, but no one technique is universally accepted.

The mode of administration of vasoactive agents is a second problem perplexing the student of human vascular physiology. J. T. Shepherd (39) points out in his book, Physiology of the Circulation in Human Limbs in Health and Disease, that

> with intravenous infusion of drugs, complex effects on the general circulation complicate the picture. With intra arterial infusions, which do not have this disadvantage, it is doubtful whether uniform distribution [of the drug] throughout the limb is ever achieved.

Poor mixing may be mainly attributed to the strong tendency for laminar flow of blood.

Andres, Zierler, Anderson, Stainsby, Cader, Ghrayyib, and Lilienthal (2) in 1954, applied the constant infusion indicator-dilution technique of Stewart and Hamilton to measure blood flow in the human forearm. They concluded that the technique provided accurate measurement in most subjects. There are two major advantages to this approach to the study of human vascular physiology. First, the human upper extremity vascular bed is readily accessible for study. Second, indicator and vasoactive agent may be administered simultaneously intra-arterially, and, if indicator is well mixed with arterial blood, so also is the vasoactive agent. Therefore, if there is evidence of mixing of indicator, the investigator may both calculate

blood flow and also infer that the vasoactive agent is uniformly distributed throughout the vascular bed of the limb. However, Andres et al. reported that they achieved relatively uniform distribution of indicator in effluent venous blood in only eighty per cent of cases. Since mixing occurs on both the arterial and venous sides of the capillary bed, the distribution of infused substances, notably vasoactive agents, was probably even less satisfactory at the arteriolar level than in the venous blood. In addition, Andres et al. did not measure indicator concentration in arterial blood downstream to point of injection. This latter measurement is necessary to the argument that vasoactive agent is uniformly distributed at the level of the arteriole.

An investigator validates an indirect technique for determination of regional blood flow by comparing calculated flows with actual flows. In addition, values calculated by the new technique will be compared to values calculated using older, more established indirect techniques. Therefore, it is in order to present a brief discussion of these other indirect techniques which have been applied to the determination of limb blood flow in man. Techniques requiring surgical intervention, such as the use of electro-magnetic flow meters and rotameters will not be considered, because they are inapplicable to measurements in human limbs under ordinary conditions.

Indirect Methods for Determining Regional Blood Flow

Indirect methods for determining regional blood flow fall into two general categories, mechanical methods and blood tissue exchange

methods. The former category includes venous occlusion plethysmography.

Venous Occlusion Plethysmography

Since Hewlett and van Zwaluwenburg (27) modified the original method of Brodie(11) introduced in 1905, there has been a wealth of literature reporting measurement of blood flow in human digits and extremities by the technique of venous occlusion plethysmography. The underlying principle of plethysmography is that the rate of increase in limb volume during brief arrest of the venous return from a limb represents the rate of venous collection, which equals the rate of arterial inflow. Conrad and Green (14) have summarized the several conditions that are essential to the inference that plethysmographically recorded resting blood flow is equal to arterial flow before occlusion: (a) the veins must be totally occluded; (b) the arterial inflow must not be altered initially by the occlusion pressure or the rising venous pressure (in a study of vascular responses, this assumption should be independently validated for each vasoactive agent which alters venous compliance); (c) the occlusion artifact should be insignificant or easily evaluated; (d) the preceding factors should hold true for a time sufficient for measurement. These four assumptions have been tested to some degree. Barnett (7) inferred that all collapsible veins were occluded after noting that the same apparent flow was recorded within a wide range of occlusion pressures. Landowne and Katz (33) concluded, after measuring tissue pressures under an occlusion cuff, that fifty mm. Hg. of cuff pressure is

sufficient to occlude all collapsible veins. Formel and Doyle (18) noted no escape of intravascular radioisotope under an occlusion cuff until venous pressure distal to the cuff approximated occlusion pressure and concluded that venous leakage did not occur during a time sufficient for measurement. Conrad and Green (14), in probably the most satisfactory and direct study of these assumptions, using amputated forepaws of dogs, noted that "venous tamponade was complete for a period of time dependent on the height of occlusion pressure and the magnitude of arterial flow. Paw venous pressure rose rapidly to effective occlusion pressure, at which time blood began to escape beneath the occlusion cuff." However "the duration of tamponade was adequate for the measurement of inflow within a wide range of occlusion pressures."

Landowne and Katz (33) stopped flow and measured the occlusion artifact, finding an initial abrupt rise followed by a slower secondary rise. Conrad and Green (14) confirmed this finding, noting that the secondary artifact represented a positive error of about 4.4 per cent of the plethysmographic flow. They also found that occlusion did reduce arterial flow significantly, representing a negative error of about 7 per cent. These two errors tended to cancel each other, so that Conrad and Green reported good agreement between plethysmographically measured flow and true arterial inflow. Although these authors made recordings at several spontaneously different magnitudes of flow, they did not investigate the effects of locally infused vasoactive agents upon plethysmographically recorded flows.

Other objections to plethysmographic technique would include

the point that venous outflow through bone cannot be occluded by the cuff and may represent a significant flow. In addition, the technique requires high, non-physiologic venous pressures, and therefore elevated capillary hydrostatic pressure, and doubtless transcapillary movement of fluid is produced. One also wonders whether this technique might be erroneous in cases of systemic arterial hypotension, or venous hypertension. In addition, many errors in plethysmograph technique are possible and can easily result in recording of erroneous blood flows. These errors are outlined by Greenfield (22) and include positioning of cuffs, plethysmograph, and subject. Correct water bath temperature is also of great importance, according to Barcroft and Edholm (5).

In summary, although venous occlusion plethysmography, if correctly done, results fortuitously in apparently correct values for resting blood flow, the validity of the technique for measurement of flow responses to vasoactive agents has not been tested and is particularly questionable in the cases of those agents which decrease venous compliance.

Table 1 is a summary of representative values for forearm and hand blood flow as determined by several authors using venous occlusion plethysmography. The prominent effect of water bath temperature is apparent. Shepherd (39) states that "water temperatures of 30 to 32°C. for the hands and feet and 34 to 35°C. for the forearm and calf have been found most suitable for the study of vascular reactivity." From Table 1 mean hand plus forearm resting blood flow at these "optimal" temperatures would be 8.8 ml. per 100 cc. hand plus forearm volume per

TABLE 1

RESTING FOREARM OR HAND BLOOD FLOW DETERMINED BY PLETHYSMOGRAPHY

Investigator Technique	Forearm or Hand	Water Bath Temperature °C	Ambient Temperature °C	Blood Flow ml/100cc/min	
				Mean	Range
Barcroft and Edholm (5) Air Plethysmography	Forearm	..	18.5	3.1	2.6-3.6
Slaughter et al. (40) Air Plethysmography	Forearm	..	27-29	4.9	1.7-7.3
Barcroft and Edholm (5), (6) Water Plethysmography	Forearm	33°C	15-20	2.7	1.9-3.8
Barcroft and Edholm (5), (6) Water Plethysmography	Forearm	35°C	15-20	4.2	1.5-7.0
Catchpole and Jepson (12) Water Plethysmography	Hand	25°C	21	2.5	1.7-3.7
Catchpole and Jepson (12) Water Plethysmography	Hand	30°C	21	4.6	1.7-9.3

minute.

Blood-Tissue Exchange Methods

The basis of blood-tissue exchange methods, a more extensive category, is the Fick principle. These methods utilize exchange of several different substances such as heat energy (skin temperature, calorimetry, thermal-dilution techniques), O_2, inert diffusible substances such as Na^{22}, Kr^{79}, Xe^{133}, and N_2O, and inert non-diffusible substances such as the various indicator dyes which combine with plasma protein, and isotope labelled serum albumin or erythrocytes. The Fick principle derives from the principle of material conservation. The Fick principle states that for any substance carried by the flow of blood to a region within a stipulated time interval (Δ t), the quantity brought in must be equal to the quantity removed. Disposal may be by accumulation in the region or by removal (Qm) from the region by transport out of the region through all the routes of egress. In a steady state where there is no accumulation, where blood flow represents the only significant patn of supply and removal, where the rates of flow of arterial and venous blood are equal (F) and constant, and where the concentrations of the substance in arterial (C_a) and mixed venous blood ($C_{\bar{v}}$) from the region are constant, the principle reduces to the familiar Fick equation:

$$F \quad (C_a - C_{\bar{v}}) = Qm/\Delta t$$

The Fick principle may also be applied to a steady state where there is accumulation in tissue of substance introduced abruptly into the arterial blood. If the arterial and venous blood of a region represent the only significant pathways of entrance or exit of the substance, and if the substance is non-metabolizable, Qm = 0 and the

equation can be written as $dQi/_{dt} = F(C_a - C_{\bar{v}})$ where Qi is the total quantity of the foreign substance in the region, including its contained blood.

Blood flow in small areas, the venous drainage of which is not accessible, may be determined by a variation of the Fick equation. A diffusible substance is injected directly into the tissue of that area and the rate of disappearance is measured:

$$C_i \quad (T) = C_{io} e^{-kiT},$$

where $C_i(T)$ represents the concentration of substance in the tissue under study at time, T; C_{io} represents concentration of the substance in the tissue at time, 0, and $ki = m_i F_i / \lambda_i V_i$, where m_i denotes the extent to which diffusion equilibrium for a particular inert substance is achieved between blood and tissue during passage from the arterial to the venous end of the capillary, λ_i represents the tissue-blood partition coefficient for the substance and tissue in question, V_i represents the volume of the region in question and F_i represents the flow through the region in question. Assumptions necessary for the above equation are: (a) M_i is close to unity, that is, the exchange of the substance between capillary blood and tissue is not limited by the process of diffusion; and (b) C_a, the arterial concentration of the substance, is negligible.

It is apparent that techniques dependent on sampling of venous blood to measure $C_{\bar{v}}$ yield results that are at best valid only for the regions represented in the sample. Therefore, in this case, consideration must be given to the adequacy of mixing and the purity of the venous blood from the region in question. On the other hand, in those

techniques in which venous blood is not available and where the assumption of rapid equilibration between capillary blood and tissue is made, diffusion is an important consideration. It should also be pointed out that those equations depending on venous sampling measure total blood flow, whereas those techniques which do not measure concentration in venous blood neglect the flow through arterio-venous shunts and estimate only capillary blood flow. Total extremity blood flow can be accurately measured by these techniques only if the substance is introduced into all tissues (or vascular beds) of the extremity and measured in venous blood which is truly representative of all extremity venous blood.

Diffusible agents injected directly into tissue. To consider first techniques employing diffusible substances injected into the tissues, an index of peripheral muscle blood flow has been derived by Kety (31) using radioactive sodium Na^{24}, by Human (30) using Na^{22}, Bauer et al. (8) using K^{40}, and Rapaport et al. (35) using I^{131}. More recently Lassen et al. (34) employed the radioactive inert gas $Xenon^{133}$ as indicator to calculate absolute muscle blood flows in the legs of subjects. This substance does not recirculate and readily crosses cell membranes, advantages not true of the sodium isotopes. Lassen et al. noted that "at higher rates of blood flow the Na^{24} clearance-rate was much slower than that of Xe^{133}, plainly showing an impeded transfer of Na^{24} to the total capillary venous efflux from the injected area." Calculated resting calf muscle blood flow averaged 2.0 ml. and 2.2 ml. per 100cc. volume per minute in healthy subjects over and under fifty years of age respectively.

Heat, introduced into the muscle tissues from a heated element, may be used as indicator. The heat clearance rate is measured continuously by thermocouple. Heat at a reference junction one centimeter from the heated element is simultaneously sensed and subtracted to eliminate changes in absolute temperature of the muscle. Change in the heat conductivity of the tissue, thus measured, is an index of change in blood flow, Gibbs (21), Hensel and Ruef (25), Stow and Schieve (42). Hensel and Bender (26) have also devised a heat conductivity meter to measure skin blood flow by the same principle. Stow and Schieve (42) report that skin blood flow in the calf of the leg of man measured by this technique ranges from 0.9 to 56 ml. per 100 cc. volume per minute, with magnitude directly proportional to skin temperature. The lower measurement was made at skin temperature of 33.5°C. and the higher at 35.2°C. Hensel and Bender (26) report values between 0.3 and 50 ml. per 100 cc. volume per minute for blood flow in the skin of the finger.

Diffusible agents injected intra-arterially. Diffusible substances injected intra-arterially to measure blood flow by blood-tissue exchange include Na^{24} (Dobson and Warner [16]). In this case complex clearance curves are obtained suggesting washout of sodium through three parallel pools. From analysis of these components, the relative pool sizes can be calculated. The authors state that

> if the ratio of sodium concentration in tissue to that in blood is known, the relative pool sizes can be converted into relative tissue volumes and the turnover rates can be converted into the blood-tissue perfusion factors with units of blood flow per unit volume of tissue per unit time.

However, as is pointed out by Katy (32), in the physiological system

"the mathematical theory based on simplified models becomes increasingly complicated" and it is difficult to determine exactly how many components are included in the actual curves obtained. Recently, using intra-arterial injections of Krypton^{85}, a radioactive gas emitting mainly beta particles, Bell and Harper (9) have reported measurements of regional blood flow through the skin. Similar to Xenon, this element is nearly completely cleared by the lungs and there is the advantage of no effective recirculation. Since beta particles have short range in tissue, skin blood flow alone is measured. The authors did not report quantitative values, because the partition coefficient of krypton between skin and blood had not yet been determined.

Shepherd and Warren(38) used the nitrous oxide method of Kety to measure lower limb blood flow in man, finding the average flow to be 4 ml. per 100 cc. per minute in normal patients. They concluded that due to the extremely slow saturation and desaturation of tissues with nitrous oxide, the method was applicable only in a steady state and even then should be considered semi-quantitative due to complex clearance curves.

Holling (29) and Roddie et al. (36) deduced qualitative changes in blood flow from changes in the oxygen saturation of blood from veins draining mainly skin and muscle. They assumed that under their experimental conditions, the oxygen consumption of tissue was constant.

Skin temperature measurements. Shepherd (38) points out that "in the proximal portion of the limb, the temperature of the skin is influenced by the rate of blood flow, the metabolism of the underlying

muscles, and the temperature of the blood returning in the superficial veins from the distal parts of the limb." Even in the digits, "at higher rates of flow, skin temperature is an insensitive and uncertain index of flow."

Calorimetry. Provided the metabolic heat is very small in comparison with the heat conveyed by the circulating blood, calorimetry gives a semiquantitative measure of blood flow. Thus, this technique is especially applicable to the digits, where the amount of metabolic heat is small. However, in limbs or digits measurement of true $C_{\bar{v}}$ is impossible because the venous blood does not become mixed in one vessel before leaving the extremity. Furthermore it is a known erroneous assumption that blood arrives in the extremity at central body temperature and leaves at calorimeter temperature. Thus, quantitative limb or digit blood flow cannot be calculated by calorimetry (Greenfield [23]).

Indicator-dilution techniques. Indicator-dilution measurements of blood flow are based upon the familiar Fick equation:

$$F(C_a - C_{\bar{v}}) = Qm/\Delta t$$

Ideally, for accuracy these measurements require the following conditions: (a) a steady state, in which the quantity of indicator leaving the bed equals the quantity introduced during the same interval of time; (b) a situation in which there is no accumulation of indicator, i.e., nondiffusible indicator; (c) a situation in which blood flow represents the only significant path of supply and removal of indicator; (d) a situation in which rates of flow of arterial and venous blood are equal and constant; (e) a situation in which the concentration of indicator

in arterial and mixed venous blood from the region are constant; (f) uniform distribution of indicator over some cross-sectional area of the vascular bed upstream to the sampling site.

In the special case of bolus injections, the Fick equation takes the form:

$$F = q/\int_0^\infty c(t)\,dt$$

where q equals the quantity of indicator injected and $\int_0^\infty c(t)\,dt$ represents the area under the concentration curve described downstream. It may be perceived immediately that in use of bolus injections of indicator for measurements of pulsatile blood flow, two of the conditions necessary for application of the Fick equation are violated: (a) the rate of flow of arterial blood is not constant, but pulsatile; (b) the concentration of indicator in arterial blood is not constant. Visscher and Johnson (43) have pointed out, however, that blood flow may still be measured if either blood flow or indicator concentration remains constant during the period of measurement. Recently Cropp and Burton (15) have reviewed this problem. They state that in the case of bolus injections into variable flow the conventional theory is invalid if the blood flow varies significantly during the inscription of the downstream time-concentration curve. However they point out that the wash-out curve recorded distal to an effective "mixing region" does allow accurate calculation of flow, if no changes in stroke volume and rate occur during the period of inscription of the dilution curve. In this case the mixing region acts to attenuate the beat-to-beat oscillations of pulsatile flow. In flow models they found that lack of such a mixing region caused calculated flows derived from

bolus injections of indicator to be grossly inaccurate.

In view of Cropp and Burton's findings, it is surprising that several authors have used bolus injections of indicator into the arteries of peripheral vascular beds to measure flow and reported accurate flows, although no apparent mixing region existed. Froněk and Ganz (19) injected boli of thermal indicator into single blood vessels, detecting time concentration curves with thermistors located immediately downstream, within five to ten mm. of the injection orifice. They attempted to validate their technique in model experiments, where flow was constant, not pulsatile, and found no systematic deviation of calculated from actual flows. They also reported that calculated carotid artery flow in dogs did not differ systematically from flow determined by rotameter. In addition calculated pulmonary artery flow in dogs agreed to within $\pm$ 6.7 per cent of flow determined by the Fick method. Although they reported that the kinetic energy of their injections lay between 10,000 and 13,000 gm. cm.^{2} sec.$^{-2}$, they did not determine if the injections caused hemolysis. In a later paper, the same authors (20) reported using their technique to measure blood flow in the femoral artery in man at rest and during exercise. They found average resting flows $\pm$ S.D. of 6.0 $\pm$ 2.3 ml. per 100cc. leg volume per minute. The variability seen in 48 measurements was $\pm$ 13.7 per cent, explained by the authors as probably due to "phasic alterations of flow during the cardiac cycle." The authors point out that their values were 1.2 - 4.9 ml. per 100cc. volume per minute higher than those reported in the literature from occlusion plethysmographic studies.

Radioiodinated (I^{131}) human serum albumin was injected as bolus indicator into the femoral arteries of men by Agrifoglio et al. (1). Downstream concentration was monitored in the femoral vein and lower extremity flow calculated on the basis of the described curve. Injections were made upstream "as rapidly as possible," through an eighteen gauge indwelling Cournand needle. Calculated flows averaged 6.88 ml. per 100cc. volume per minute, with range of 6.30 to 7.78, in normal subjects. The authors demonstrated a significant rise in calculated flow following lumbar sympathectomy, administration of reserpine for ten days, or following muscular exercise. No calculations were made of the kinetic energy of the injections and no investigation was made of production of hemolysis by the injections. Folse (17) made a similar study in the femoral arteries of man, using indocyanine dye as indicator, and found mean resting femoral arterial blood flow to be 301 ml. per minute with range 196 to 484.

Hobbs, Agrifoglio, and Edwards (28), using the same technique as Agrifoglio et al., compared calculated femoral arterial blood flows in twenty one dogs with simultaneous flows recorded by a flow meter included in the artery. Over the flow range 30 to 120 ml. per minute the agreement of values obtained by the two methods was close. However indicator-dilution values were considerably higher than those from the flow-meter over flow range 10 to 30 ml. per minute.

The work of Baker and O'Brien (3) indicates that the type of bolus technique used by Agrifoglio et al. and Hobbs et al. apparently can yield accurate calculated flows even over low flow

ranges if the time-concentration curve is observed in pooled venous blood. These authors perfused the isolated forelimbs of dogs with a pulsatile blood pump. The muscles above the elbow were ligated and cut, leaving the forelimb connected to the body only by the humerus, brachial artery, brachial and cephalic veins and the brachial nerve trunk. Total venous outflow was measured by collection and compared to flow calculated by an indicator-dilution technique. The indicator red cells-Cr^{51}, albumin-I^{131}, or indocyanine green was injected as a bolus of 0.5 ml. in 1 second through a 20 gauge needle into the brachial artery downstream to the blood pump, and the time-concentration curve was measured in the pooled venous blood downstream. One hundred and eighteen simultaneous measurements of actual and calculated flow were made in eighteen dogs over a flow range of twenty to two hundred ml. per minute. (In some cases 2 μg. per kilogram body weight per minute acetylcholine was infused with indicator to dilate the bed.) All but 17 calculated flows were within $\pm$ 10 per cent of the actual flows.

In several experiments flow was also calculated from the concentration of indicator (either red blood cells-Cr^{51} or albumin-I^{131}) in collected total venous effluent to determine if all the injected indicator was being returned from the limb. It was found that there was no trend for appreciable loss of either indicator. These authors also placed a mixer at the point of injection and point of sampling and found no differences in the shape of the curves or the pattern of the data.

The findings of Baker and O'Brien suggest that limb flow may

be accurately calculated using the bolus indicator-dilution technique and that complete arterial mixing of indicator is not essential for accuracy. In addition it seems probable that the assumption that indicator albumin-I^{131} is not lost from the circulation during transit through the limb is true.

Although basically similar, theory of the constant infusion indicator-dilution technique is simpler than theory of the bolus injection technique. In addition the constant infusion technique better satisfies the Fick principle requirement of constant indicator concentration. Stewart (41) first proposed that constant injection of indicator could be used for measurement of flow. Hamilton and Remington (24) and later Andres et al. (2) formally considered this application. The basic principle of this technique is as stated by Andres et al.:

> When indicator is injected at constant rate into a vascular bed ultimately all blood free of indicator will be displaced from the system by indicator-laden blood and the concentration at exits from the system will become equal to the rate of injection (mass per unit time) divided by the constant rate of volume flow (volume per unit time) through the system, provided that there is no recirculation.

The expression proposed by Andres et al. (2) for calculation of forearm flow during constant intrabrachial arterial infusion of indicator is

$$F = \frac{I}{C_{(r_o,t)} - \frac{a(t - t_o)}{F}}$$

where F equals total forearm flow, I represents rate of constant injection of indicator, $C_{(r_o,t)}$ represents observed concentration at exit, and $\frac{a(t - t_o)}{F}$ is the concentration of recirculating indicator, adjusted for its time intercept.

Cropp and Burton (15) also reviewed the validity of constant infusion indicator-dilution methods for measurements of variable flow. They found that with adequate mixing close to the injection site, valid calculations for non-steady (pulsatile) flow are possible if the mean of the reciprocal of the instantaneous sampled concentration ($\overline{1/c}$) is used in calculations, rather than the reciprocal of the mean concentration ($1/\bar{c}$). Cropp and Burton showed in flow models, using pulsatile water flow and thermal indicator, that use of the reciprocal of the mean concentration always leads to underestimates of true flow, of increasing magnitude as the degree of fluctuation of flow increases. On the other hand, their experiments in flow models indicate that use of the reciprocal of the instantaneous sampled concentration gives valid calculated flows. They point out that when they added a mixing region between the injection and sampling sites, oscillation of the indicator time curves due to pulsatility of flow were attenuated. In these cases, use of either $\overline{1/c}$ or $1/\bar{c}$ in calculations gave valid calculated flows. The "mixing region" of their model was a chamber in series having five times the cross-sectional area of the tube and a volume approximately twice the stroke volume of the pump. They state that the mean transit time of the mixing chamber should optimally be short compared to the period of fluctuation of flow. They felt that "the lung, a cardiac chamber or an aneurysmal dilatation" would have effects similar to their "mixing region." They did not mention a peripheral capillary venous bed as a possible "mixing region."

Cropp and Burton (15) also discuss the theory of the distance-distortion error. At a sampling site some distance downstream from

the injection site, the fluctuation of sampled concentration will not correspond accurately to changes in flow. Not only is there a phase difference, but more important, the contour of the concentration-time curve will be distorted. At this sampling site the detector will usually be exposed to low concentration, which is generated in systole, for a longer time than the actual duration of systole. This is due to the fact that the low concentration segment may move past the sampling site, not at high systolic velocity, but at low diastolic velocity. This "distortion" always results in an over estimate of the total flow. In their experimental model, Cropp and Burton, confirmed these theoretical predictions. They also determined that at certain points downstream to the injection site, the contour of the concentration time curve is not distorted, and sampled concentration corresponds accurately to flow. These points are multiples of a "wave length," defined by them as:

$$\text{wave length in cm.} = \frac{\text{pump (heart) stroke volume } (cm.^3)}{\text{cross-sectional area of artery } (cm.^2)}$$

Distance-distortion error reaches a maximum at 0.75 wave length (or multiples thereof) at which points it is as much as a + 22.6 per cent error. However if a "mixing region" is interposed between injection and sampling sites, distance-distortion error disappears.

These same authors investigated adequacy of mixing of indicator in the pulsatile stream by sampling indicator-time curves at several points across the stream during constant infusion of indicator. When these curves were similar, this was considered proof of adequate mixing. They found that in 1 cm. I.D. tubing it was necessary to

infuse indicator at a kinetic energy of at least 30,000 g. cm.^{2} sec.$^{-2}$ in order to achieve good mixing within 3 cm. Cropp and Burton point out that these energies would cause hemolysis *in vivo*. In 2 cm. I.D. tubing, they were not able to achieve good mixing within 5 cm. of the injection site even with high energy infusions. However, when a "mixing region" was added between the injection and sampling sites, mixing was good, even if indicator had been infused with a low kinetic energy. In this situation, the correlation between calculated and collected flows was 0.998 $\pm$ 0.01 S.E. Cropp and Burton summarize by stating that "the most favorable conditions for the measurement of mean flow rates in the model were provided by continuous infusion of indicator and by a mixing chamber between injection and sampling sites." They go on to state that "under the right conditions, indicator-dilution methods, by steady infusion, can be remarkably accurate."

The classical work on constant infusion indicator-dilution measurement of blood flow in the upper extremity of man, is that of Andres *et al.* (2). These authors desired to develop an approach to measurement of human forearm blood flow which would yield continuous recording of blood flow as well as serial samples of arterial and venous blood for metabolic studies. In addition, they pointed out that no verification of the plethysmographic method by an independent technique had been made in man up to that date (1954). They felt that the continuous constant rate injection technique was preferable to the single, nearly instantaneous injection (bolus) technique for their work, because it answered their need for prolonged measurements of blood flow. Evans Blue dye, T-1824, was chosen as indicator, to be infused

into the brachial artery at the elbow and sampled continuously from two ipsilateral forearm veins in the antecubital fossa. They attempted to use veins which appeared to drain the deep and superficial circulations respectively. They found that if indicator was injected through a twenty gauge needle at a low volume rate, the concentration of indicator in one sampled vein differed from that in the other vein. They concluded from this finding that indicator had not been distributed uniformly throughout the forearm vascular bed. Possible explanations suggested by these authors for this uneven distribution of indicator included: (a) collateral arteries about the elbow diluting labeled with unlabeled blood downstream to the injection site. They dismissed this possibility on the grounds that collateral arteries about the elbow probably contribute only a small volume relative to brachial arterial blood flow, and this small amount of dilution could not cause a significant lack of uniformity in indicator concentrations downstream; (b) Uneven escape of dye from vascular channels. The authors dismissed this possibility on the grounds that the amount of albumin leaving the extremity via the lymphatics is negligible compared to that leaving via the veins; (c) Anomalous bifurcation of the brachial artery (to be discussed below); (d) Incomplete mixing of dye and blood in the brachial artery. This latter factor seemed to the authors to be the most likely explanation, and the problem appeared to be one of achieving complete mixing of indicator and arterial blood as near the site of injection as possible. In order to accomplish this, according to these authors, the laminar flow of brachial arterial blood must be transformed momentarily into turbulent flow. However, it can be calculated that the

Reynolds number of brachial arterial blood is equal to or less than one-tenth of the critical Reynolds number. Therefore, laminar flow in the brachial artery is so stable that ordinary methods of intra-arterial infusion do not produce turbulence and mixing. Andres et al. developed a jet injector to achieve turbulence and mixing of injectate with brachial arterial blood.

Their flow model experiments, using glass tubing of 6 mm. I.D. through which water flowed at a constant, not pulsatile, rate of 50 ml. per minute, indicated that mixing was achieved within 2 cm. of the jet-injector if indicator was infused upstream at a rate of 1 ml. per minute through a needle tip jet orifice of 0.001 inch diameter.

Andres et al. also isolated the femoral arteries of three dogs, ligating all but two branches, which were cannulated for sampling of indicator concentration. Indicator was infused constantly against the direction of flowing blood two to three centimeters upstream from the proximal collecting site. They demonstrated that concentrations of indicator differed greatly at the two sites when the infusion was made through a twenty-six gauge needle. When infusion at the same volume rate was made through the jet injector the ratios of dye concentration at the two sites were nearly unity, demonstrating that the indicator had been distributed uniformly within a distance of several centimeters of the injector. The authors made no attempt to calculate blood flow on the basis of indicator concentration and compare it to actual flow in these dogs. Despite these favorable data, when the authors tested the jet injector in the forearms of twenty-seven subjects, comparing indicator concentrations in a deep and superficial vein, they were not

able to demonstrate that the jet injector achieved a reduction in the differences in indicator concentration between veins over that produced by injection through a standard eighteen or twenty gauge arterial needle.

Because the authors made multiple paired measurements of venous indicator concentrations in each subject under steady state resting conditions, they were able to compare statistically the paired indicator concentrations in each subject to determine the probability that the samples from the two veins had arisen from the same population, despite the fact that they contained different amounts of indicator. In their analysis, they suggested the following symbols, which will also be used in the study presented in this thesis:

C_D, a concentration of indicator in the deep vein;

C_S, a concentration of indicator in the superficial vein, occurring simultaneously with C_D;

n, the number of pairs of samples in a single experiment;

$C_M = \frac{C_D + C_S}{2}$, the mean concentration of a pair of samples;

$\bar{C}_D = \frac{\sum C_D}{n}$, the mean concentration of indicator observed in the deep vein during a single experiment under constant conditions;

$\bar{C}_S = \frac{\sum C_S}{n}$;

$\bar{C}_M = \frac{\sum (C_D + C_S)}{2n} = \frac{\sum C_M}{n}$;

$r.d. = \frac{C_D - C_S}{C_D + C_S} 100$, the relative difference, the per cent by

which a concentration of indicator in either vein differs from their mean concentration, C_M;

$$m.r.d. = \frac{\Sigma r.d.}{n}$$, the mean relative difference in a single experiment.

Coefficient of variation about mean flow $= \frac{\sigma}{\bar{C}_M}$ 100, where σ is the standard error of $\bar{C}_M$.

They found that with either the jet or regular injection system, the differences between the means exceeded chance at the one per cent level in fifty per cent of the subjects and exceeded chance at the five per cent level in sixty per cent. They also found that the mean concentrations from either vein differed from the overall mean by no more than twenty per cent in eighty per cent of subjects and differed from the overall mean by no more than ten per cent in two-thirds of the subjects.

The authors suggested that relative constancy of rate of blood flow is a requisite for the statistical analysis above. In their groups of subjects forearm blood flow was "remarkably constant." In ninety per cent of subjects the coefficient of variation was less than 20 per cent; that is, during about two-thirds of the period of observation in each of the subjects, flow did not vary from its mean by more than 20 per cent.

The authors also analyzed the mean relative difference in indicator concentration, which should not differ significantly from zero if indicator were distributed uniformly. In about sixty per cent of subjects, with either type of injector, the m.r.d. indicated that

there was a real difference in indicator concentrations in the two sampled veins (probability of chance occurrence less than one per cent). The authors interpreted these results as indicating that "in approximately half the subjects, the concentrations of indicator in the two veins sampled were in the same statistical population and that indicator was, therefore, distributed uniformly over venous blood draining the forearm."

The authors go on to a discussion of the error involved in calculating flow from the mean indicator concentration obtained by sampling only two of the veins draining the forearm. They point out that,

> if the blood in the two large veins sampled represented a very large portion of total forearm blood, it is probable that departures in indicator concentration elsewhere in the forearm would not cause the true mean concentration of dye in all blood flowing from the forearm to lie outside the range established by the paired samples.

Therefore, if this assumption is true, flow calculated from the mean concentration of indicator provides a measure of blood flow with known error. On this basis, the authors pointed out that in 80 per cent of their subjects the flow calculated from the mean indicator concentration did not differ from the true flow by more than $\pm$ 20 per cent. The authors concluded that in 80 per cent of their subjects there appeared to be sufficient intermingling of blood upstream to sampling site to produce a relatively uniform distribution of indicator among the veins draining the forearm, thus permitting measurement of resting blood flow.

It is seen that the essential conclusions made by Andres *et al.*

are dependent on their statistical analysis of the observations. To review these analyses, this group makes the following comparisons between two sample means: (a) A Student t analysis in each subject of the null hypothesis that over time the mean indicator concentration in one sampled vein, $\bar{C}_D$, is identical to the mean indicator concentration in the other sampled vein, $\bar{C}_S$. The authors do not state whether the paired or unpaired test was used. (b) A Student t analysis in each subject of the null hypothesis that the mean relative difference in indicator concentration, m.r.d., does not differ significantly from zero.

The authors state that the former analysis assumes that the rate of blood flow is relatively constant. However it is difficult to see the importance of constancy of blood flow in this analysis, if $s_{\bar{d}}$, the sample standard deviation, remains unchanged. In addition, the authors did not mention that conclusions from the analysis are dependent on the magnitude of $s_{\bar{d}}$. No data is given to enable the reader to calculate $s_{\bar{d}}$. If $s_{\bar{d}}$ is large, the beta type error may be quite large and the test will have little power to distinguish a real difference between the two means. Andres et al. state that the difference between the means exceeded chance at the one per cent level in fifty per cent of the subjects and exceeded chance at the five per cent level in sixty per cent.

Their second analysis compared m.r.d. against zero by the t test. The same criticism may be made of the authors' use of this test. Again, the power of the test is dependent upon the magnitude of the standard deviation, and the authors give us no data on this point. They

conclude that in sixty per cent of subjects there was a real difference between m.r.d. and zero at less than the one per cent level of probability. In both tests it would seem that the findings would be best interpreted as indicating that in at least sixty per cent of their subjects the indicator concentrations in the two veins did not arise from the same population. Neither analysis as presented allows the reader to conclude in what percentage of their subjects indicator concentrations in the two sampled veins derived from the same population. Certainly it must be less than forty per cent, rather than "approximately half" as they conclude. It seems, therefore, that in less than forty per cent of their subjects did uniform distribution of indicator over venous blood draining the forearm occur.

Their argument that the true mean concentration of indicator in all blood flowing from the forearm would probably not lie outside the range established by the paired samples seems reasonable. If true, it follows that the r.d. between paired samples provides a good measure of the error of blood flow calculated from the mean concentration. However, here the assumption would be, as pointed out by Andres et al., that the blood in the two large veins sampled represents a very large portion of total forearm blood. Another assumption would be that there is no loss of indicator due to, among other things, short-circuiting vascular channels at the elbow. Given these assumptions, it would seem that the r.d. is of value as an index both of the degree of mixing and of the confidence limits of calculated blood flow. In this regard it is interesting that m.r.d. was less than twenty per cent in eighty per cent of the subjects observed by Andres et al. It seems reasonable to

conclude that Andres et al. were able to measure resting forearm blood flow to within ± 20 per cent of its actual value in 80 per cent of their subjects. This interpretation of their data might be preferable to their conclusion that they were able to measure accurately resting blood flow in 80 per cent of their subjects.

These authors, using a standard needle for infusion, found mean resting forearm blood flow in 7 subjects to be 4.67 ml. per 100 cc. forearm volume per minute with range 2.70 to 7.05. The mean m.r.d. was 6.0 per cent with range 0.9 to 13.3 per cent. Three patients were excluded, two because of m.r.d.'s greater than 20 per cent, and one because of CV of 52 per cent. The authors state that in these three subjects there was reason to suspect that the mean concentration of dye was an improper measure of resting flow.

The authors soon discovered that intra-arterial infusions through their jet injector at rates calculated to be necessary to provoke turbulence and mixing also caused hemolysis of erythrocytes and resulting vasodilatation. This vasodilatation is known to be dependent on adenosine triphosphate and related substances released from erythrocytes. Andres et al. found that this mechanical destruction of erythrocytes was related to the kinetic energy per unit time of the injection, and determined, by in vitro jet injections into a pool of citrated human blood, that hemolysis became detectable when the kinetic energy per second of injection reached 10,000 to 20,000 g. $cm.^{2}$ $sec.^{-2}$. They also found that these figures correlated with the kinetic energies of infusions which produced hemolysis and vasodilatation in vivo in man. In addition, the authors found in experiments on the hind limb of the dog

that hemolysis of as little as 0.09 ml. of blood per minute increased limb total blood flow seven-fold over the resting value, so that hemolyzed blood has quite powerful vasodilating properties.

These authors concluded that use of the jet injectors with kinetic energy of infusion theoretically sufficient to effect mixing did not improve the agreement between the indicator concentrations in the two veins sampled over that produced by a non-jet injector. In addition, they concluded that the jet injections produced hemolysis and vasodilatation. In view of these results, the authors concluded that "it does not appear profitable to continue to use the jet injector for measurement of resting blood flow." The values for resting forearm blood flow and blood flow during infusions of vasoactive agents reported in this and later papers by this group were all during intra-arterial infusions of less than one ml. per minute of indicator solution through a standard gauge arterial needle.

As a result of their observations of the distribution of indicator in man, two questions arose for Andres et al. : "Why did the jet injector fail to produce better mixing than the non-jet injector? Why was there mixing when the non-jet injector was used?"

Regarding the former question, the authors had concluded from statistical analysis of their data that indicator concentrations in the two veins, although having differing values, derived from the same population in approximately eighty per cent of subjects. They concluded that in these subjects complete mixing did occur. In the twenty per cent of cases where the n.r.d. exceeded twenty per cent, the authors concluded that mixing did not occur and suggested that this was due

either to a rate of injection too low to provoke turbulence, a rate of brachial arterial blood flow too fast to allow mixing of indicator before the bifurcation, or to anomalous bifurcation of the brachial artery above the elbow. Such anomalous bifurcation is known to occur in about twenty per cent of the population. In these cases, indicator was injected into the ulnar or radial artery rather than the brachial artery. The authors point out that the percentage of failures to achieve adequate mixing of indicator and blood did not differ from that to be anticipated from the known rate of occurence of anomalous bifurcation.

The authors suggest that when the non-jet injector was used, sufficient mixing occurred on the venous side of the forearm circulation in eighty per cent of subjects to allow conclusion that the indicator-concentrations in the two veins sampled derived from the same population.

Andres et al. (2) attempted to measure blood flow with venous occlusion plethysmography simultaneously with their indicator-dilution procedure in a few of their subjects. In general they found that flow per 100 cc. forearm volume measured by indicator-dilution was 50 per cent greater than that measured by plethysmography. They point out that the difference may reflect real differences in the methods or may be owing to the fact that the plethysmograph had to be placed more distally than usual in order to provide room for the arterial injector, so that the plethysmograph may have enclosed a volume of forearm with relatively less vascular supply. In addition, in these cases high energy jet injections of indicator were used, which caused hemolysis, vasodilatation, and high blood flows. The authors suggest that in

this situation the plethysmograph may tend to yield lower than actual values, but they do not indicate their reasons for this suggestion.

More recently Wahren (44) attempted to use the constant infusion indicator-dilution technique to measure brachial arterial blood flow during exercise. He infused indicator in 5 per cent dextran solution into the brachial artery through a standard needle at a high volume rate (about 34 ml. per minute). At this rate of infusion he found that indicator concentrations in the radial artery, a deep, and a superficial vein were similar, and he concluded that he had achieved satisfactory mixing. He made a comparison between blood flows determined for the upper extremity simultaneously with venous occlusion plethysmography and with the indicator-dilution method. Varying amounts of bradykinin were infused with the indicator solution in order to increase the blood flow. Over a range of 180 to 680 ml. per minute total flow there was no significant difference between flows calculated by the two techniques. Wahren points out that an infusion rate of 34 ml. per minute may well be a significant fraction of the total flow through the brachial artery at rest and during light exercise. However, he found that oxygen saturation values for venous blood did not change during infusion of indicator, and that decrease in venous hemoglobin concentration indicated a simple addition of the infused solution to the initial blood flow. From these data he concluded that there was "no evidence of active vasodilatation," and that "the added infusion may have been accommodated by reduction of blood viscosity due to hemodilution." Wahren does not state the gauge of his arterial needle, so it is impossible to

calculate the kinetic energy of his infusion. Furthermore, he does not rule out the possibility that his infusions cause hemolysis of erythrocytes. However the significant point in this paper is the correlation found between indicator-dilution and plethysmographic flow measurements.

More recently Andres' group (4) have reported using the indicator-dilution technique with standard needle injector to measure forearm blood flow and metabolism during local infusion of the vasoactive agent, epinephrine. They found that infusion of 0.002 μ g. per kilogram body weight per minute causes an immediate and sustained increase in forearm blood flow. Infusion of larger amounts of epinephrine 0.0057 to 0.025 μ g. per kilogram per minute, resulted in a transient increase in blood flow followed by either prolonged reduction or no net change in blood flow. The authors followed blood flow from twenty to forty minutes in eight subjects after stepping epinephrine at the lower dosage and found flow was still significantly increased in five of them at the end of the period. In five subjects blood flow was measured for forty to sixty minutes following the end of the epinephrine infusion; in only one of these was flow above basal levels at the end of the time period.

The authors point out that with constant infusion of vasoactive substance, arterial concentration of the substance varies inversely with forearm blood flow. This relationship should lead to oscillations in blood flow which dampen with time. They observed that venous concentration of indicator oscillated up until the tenth minute of infusion, but the mean flow, and therefore concentration of epinephrine, was reasonably constant from the tenth to the twenty-fifth minute.

The authors do not give any data regarding the effect of the vasoactive agent on the r.d. However they do mention the case of one subject in whom concentration of dye in the deep venous plasma was large, but almost no dye appeared in the superficial venous plasma. The deep forearm bed, in this subject, which received almost all the epinephrine, showed all the characteristic epinephrine metabolic effects. On the other hand the superficial bed, which received essentially no epinephrine, failed to show the typical metabolic effects of epinephrine. The authors point out that in order to avoid misinterpretation of results due to maldistribution, dispersion of arterially administered vasoactive agents must be monitored by simultaneous injection of an indicator. However the authors do not point out that monitoring of arterial concentrations of indicator upstream gives much more satisfactory evidence of uniform mixing of the vasoactive agent at the arteriolar level than does monitoring the venous concentrations downstream.

Finally the authors state that response of venous indicator concentration to altered blood flow is slow and is a function of the mean transit time through the system. They state that a time equal to about three mean transit times is required to establish a new steady state. Since the magnitude of mean transit time is directly related to volume rate of blood flow, it follows that establishment of the new steady state of venous indicator concentration is slower with vasoconstrictor agents than with vasodilators. The authors state that during resting forearm blood flow, mean transit time is about 2.3 minutes. If flow doubles, mean transit time is reduced to a little more than a

minute, and the new steady state is achieved in about three minutes. It follows that this indicator-dilution technique tends to record an average blood flow, damping out oscillations.

In the technique developed by Andres et al. (2) tha assumption is made that deep and superficial venous blood is sampled in the two forearm veins. Coles et al. (13) investigated the source of blood samples withdrawn from deep forearm veins via catheters passed upstream from the median cubital vein. They found that, if the catheter is passed distal to the first valve encountered, the samples are a mixture of hand and deep forearm venous blood but include no superficial forearm venous blood. If, however, venous pressure is elevated by venous occlusion above the elbow, these samples also contain superficial forearm (cephalic) blood. This data does not totally support the assumption that the antecubital vein drains the deep muscular circulation alone. However analysis of the data of Coles et al. reveals that on the average the hand venous system contributes the smaller proportion of blood to the deep veins of the forearm.

Vascular Distribution of Intra-Arterially Administered Substances

The problem of vascular distribution of substances during intra-arterial infusions has been recently discussed by Rush et al. (37), who were interested in intra-carotid arterial chemotherapy in man. These authors point out that skin areas of maldistribution are seen occasionally during such chemotherapy. From Tygan they constructed a model arterial system similar to the common, external and internal carotid system and followed the course of dye injected at 1 ml. per minute through a 23 gauge needle into water perfusing the system at 500 ml.

per minute. They also injected dye into the lower aorta of dogs and sampled downstream simultaneously from points in the common iliac and sacral branches. In their model they demonstrated that branches of the vessel closer than 1 to 3 cm. to the injection site received little of the dye, if the injecting needle lay in the center of the stream. On the other hand injection adjacent to the vessel walls caused most or all of the material to enter the first branch encountered. These authors suggest the use of a jet needle similar to that of Andres et al. (2) to achieve arterial mixing. In the dog aorta, similar to the finding of Andres et al. in the dog femoral artery, Rush et al., using the jet injector, demonstrated good mixing in the arteries downstream. They also did not attempt to calculate flow and compare it to actual measured flow.

The present work explores the concept that a new type of injection system for brachial arterial infusions in man may be developed, which satisfactorily mixes both indicator and vasoactive substances with brachial arterial blood and does not create hemolysis. Such an injection system would improve indicator-dilution measurement of blood flow and vascular distribution of vasoactive agents in the human upper extremity and would contribute to the study of vascular physiology and vascular responses in normal and diseased man.

CHAPTER II

METHODS

Description of Special Equipment Used

Jet-injector Needle

Intra-arterial infusions were made through a modified twenty-six gauge stainless steel hypodermic needle, manufactured by Kimray, Incorporated, Oklahoma City. Two side holes each of .006 inch diameter were drilled into the shank of the needle about 1 mm. behind the tip and 180 degrees apart. The needle tip opening was sealed and cut so that the tip was blunt. A Clay-Adams plastic tubing to male luer-lock adapter was soldered to the hub of the needle. Figure 1 is a photograph of this needle and Figure 2 is a magnified view of the needle tip, showing how jets of infusate are directed laterally from the side holes. A B-D Swinney Filter Adapter is connected in series with the needle to eliminate any particulate matter in the infusate, which might occlude the holes of the needle. Plugging of the holes is rare, although there is often a reflux of blood into the needle. The jet needle is introduced into the brachial artery through a twenty gauge Riley arterial needle, the adapter on the needle hub locking into the hub of the Riley needle.

Infusion Pumps

Because the jet-injector requires a high volume rate of

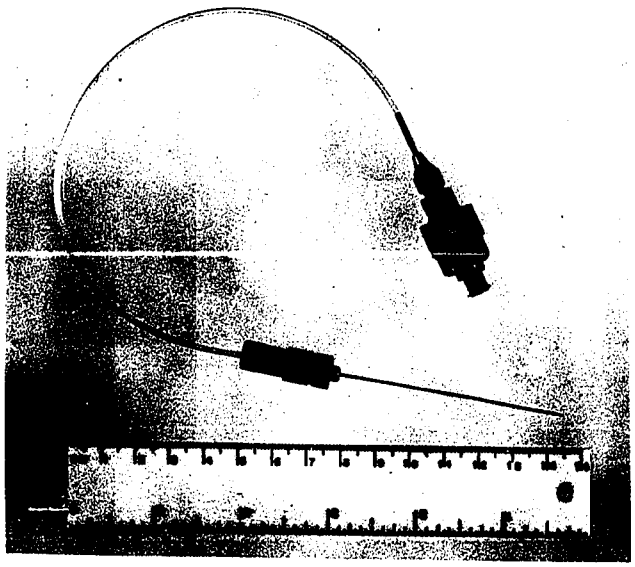

Fig. 1.-- Jet injector needle with Swinney filter adapter.

Fig. 2.-- Tip of jet injector, showing jets of Evans Blue Dye infusate.

infusion (8 ml. per minute) at high infusion pressure (up to 1500 mm. Hg.), the standard Harvard Infusion Pump proved inadequate. Kimray, Incorporated, Oklahoma City, modified the standard Harvard Infusion Pump with a more powerful motor, so that the pump was able to deliver infusions at the required volume rate and pressure. However it was not found possible to lubricate the glass syringes well enough to prevent some sticking, usually minor. In addition, at infusion rates of eight ml. per minute, these fifty ml. glass syringes contained infusate enough for only six minutes. For these reasons it was necessary to construct a special infusion pump for this project.

Kimray constructed a hydraulically operated infusion pump, pressure independent to 45 pounds per square inch and driving two 125 ml. stainless steel syringes. Power is provided by an electric motor, and delivery of infusate is held constant despite level of resistance, internal or external to the pump, by a series of pressure "bleed off" valves. Driving pressure within the pump is monitored by an external meter and pressures within a certain range provide a check of proper pump infusate delivery. Viscosity of the silicone hydraulic fluid is held constant by a thermostatically controlled heating element in the reservoir. This pump delivers at a rate of 8 ml. per minute with variation less than $\pm$ 1 per cent for periods of at least 5 hours. Figure 3 shows this infusion pump.

Figure 4 gives the detail of the syringes, barrels, and plungers. The plungers of the syringes are screwed into drive shafts projecting from the body of the pump. The barrels of the syringes are screwed into supports on the pump frame. The barrels and plungers were

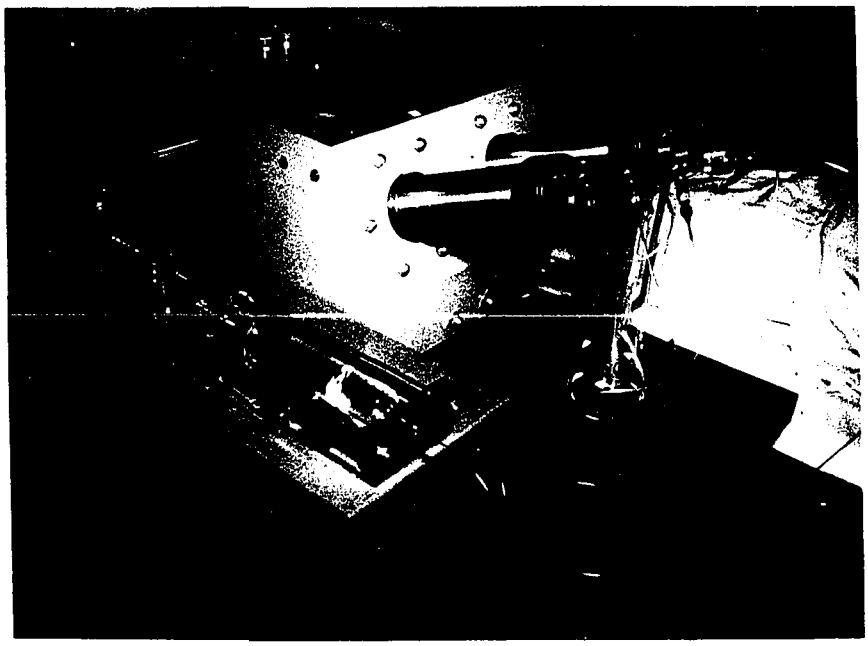

Fig. 3.-- Special high pressure, high volume infusion pump.

Fig. 4.-- Syringe barrel and plunger, showing teflon ring and lucite disc.

machined to a loose fit, and the plunger is fitted with a ring consisting of an inner "o ring" of silicone rubber and outer band of teflon, providing a lubricated, expansile, leakproof seal. A clear lucite disc is fixed onto the delivery end of the syringe, so that infusate within the syringe can be monitored for air. The infusion orifice was drilled through this lucite disc and ends in a luer-lok adapter. Syringe barrels and metal plungers are sterilized by autoclave and the teflon-rubber ring and lucite disc are sterilized in a solution of benzalkonium chloride, which is thoroughly rinsed with sterile isotonic sodium chloride solution prior to use. Syringes are filled while fixed to the pump, by reversing the direction of movement of the plunger.

Pressure between the syringe and the jet-injector is monitored and provides a check on the patency of the orifices in the jet injector.

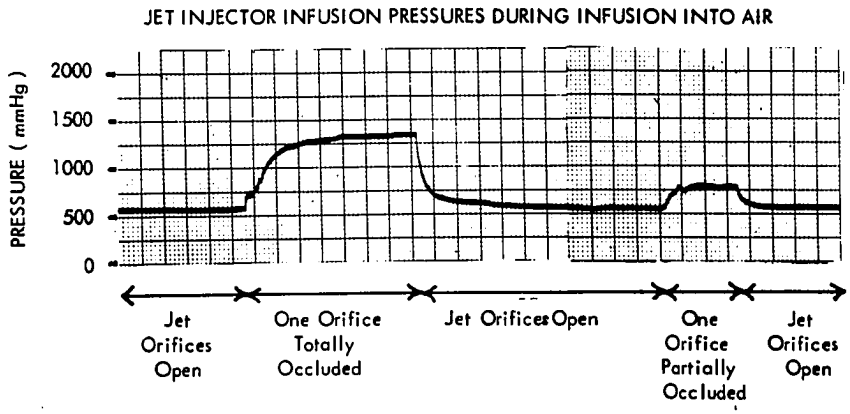

Fig. 5.-- Pressures during infusion through jet-injector

Figure 5 is a recording of such pressures obtained with both holes open and with one hole totally or partially closed. Kinetic energy of infusion with both holes open is approximately 4500 g. $cm.^2$ $sec.^{-2}$, whereas, if one hole is completely occluded so that total infusion passes through

one hole, kinetic energy of infusion becomes four times as great or 18,000 g. $cm.^2$ $sec.^{-2}$. The magnitude of the former kinetic energy is less than that found by Andres et al. (2) to produce hemolysis. The magnitude of the latter kinetic energy, however, may produce hemolysis in some instances. However, the infusion pressure will double if one orifice becomes occluded, so monitoring of this pressure will help to ensure that no hemolysis occurs. The calculated Reynolds number of the infusate from this system is approximately seventy-one, considerably less than that calculated as necessary to produce turbulent flow in the brachial artery of man.

In Vitro Experiments

Evans Blue dye was infused through the jet injector and also through a standard twenty gauge hypodermic needle against a stream of water flowing through glass tubing. The water was pumpad by a Sigmamotor pump, which provided pulsatile flows of between 25 and 200 ml. per minute. Pump frequencies were 28 per minute and 216 per minute, respectively. Internal diameter of the glass tubing used was five mm., approximately that of the human brachial artery. Flows usad were within the range to be found in the brachial artery of man. Degree of mixing of dye and water with each needle at various flow rates was observed visually and recorded by high-speed flash photography.

In Vivo Experiments

Dogs

Hemolysis. A pressure independent Sigmamotor blood pump was interposed between the femoral artery and brachial artery of mongrel dogs

anaesthetized with sodium pentothal and heparinized. Blood flow in the brachial artery was thereby held constant, and monitored perfusion pressure measured limb vascular resistance. A standard twenty gauge hypodermic needle and the jet injector were in turn introduced into the tubing downstream from the pump. Isotonic sodium chloride solution was infused constantly through the indwelling needle at eight ml. per minute, and resulting changes in perfusion pressure were recorded on a Sanborn oscillographic recording machine.

Comparison of actual with calculated flows. The forelimbs of mongrel dogs heparinized (10,000 U.S.P. units) and anaesthetized with sodium pentothal (30 mgm. per kilogram) were totally severed from the bodies and tourniquets applied to tissue of both stumps. In some cases the humerus was not cut. A pressure independent Sigmamotor pulsatile blood pump was interposed between the femoral artery and brachial artery, so that limb blood flow was supplied solely by pump. Total venous outflow from the limb drained from the cephalic and brachial veins into a reservoir and was measured from each vein by collecting blood in graduated cylinders. Pressure was monitored in each vein in order to detect any obstruction to venous outflow. Pressures remained less than ten mm. Hg. Blood was pumped from the venous reservoir back into the femoral vein of the animal. Indicator (I^{131} serum albumin in isotonic sodium chloride solution) was infused through the jet injector or through a standard twenty gauge hypodermic needle introduced against the direction of blood flow into the pump tubing immediately upstream from the junction between tubing and brachial artery. Rate of infusion of indicator was eight ml. per minute through the jet injector, and

approximately one ml. per minute through the standard needle. Samples of blood for determination of indicator concentration were simultaneously collected from the two veins and from the tubing upstream to the blood pump (recirculation concentration) at four minutes after beginning each infusion. Venous outflow was measured before and after sampling. Calculated and actual flow measurements were made at several pump flow settings in each dog. In some dogs pump flows were randomly changed, whereas in others flows were sequentially adjusted from lower to higher values or vise versa. Figure 6 diagrams the experimental situation in dogs with severed humerus.

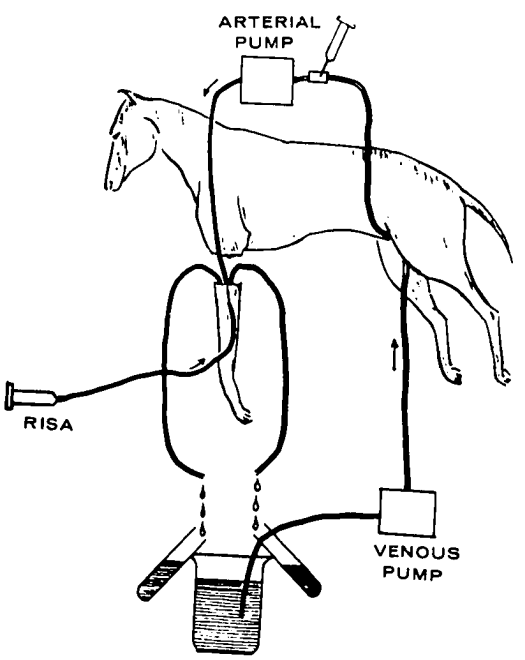

Fig. 6.-- Comparison of actual with calculated blood flow; experimental situation.

Man

General procedures. All subjects participating in this study were fully informed by the author of the purposes, hazards, and proce-

dures of the experiment. Written consent was obtained from all subjects. These male normotensive or essential hypertensive volunteers in the resting, post-absorptive state were studied in an air conditioned laboratory, with ambient temperature maintained at approximately 78°F. Prior to study, the volume of the upper extremity to the level of the intercondylar line at the elbow was measured by water displacement. With the subject comfortable in the supine position and his arms supported at a 45 degree angle from the long axis of the body, 20 gauge hypodermic needles were inserted in an upstream direction into the basilic and cephalic veins of one upper extremity (designated "ipsilateral extremity") distal to the elbow. A twenty-three gauge needle was also inserted upstream into a dorsal metacarpal vein of the ipsilateral extremity. Under local xylocaine anaesthesia the ipsilateral brachial and often the radial arteries were also cannulated in an upstream direction with twenty gauge Riley arterial needles. In addition, the contralateral brachial artery was cannulated with a twenty gauge Riley needle. Figure 7 is a drawing of upper extremity vascular anatomy with catheters in place. Cannulas in the contralateral brachial artery and in the ipsilateral radial artery, cephalic vein, basilic vein, and dorsal metacarpal vein were connected by sterile polyethylene tubing to a stopcock manifold and from there to a Statham wire resistance pressure transducer and Sanborn four channel direct writing oscillograph. The arms and transducer were adjusted to the level of the right atrium. The venous catheters and cannulae were maintained patent by intermittent flushing with heparinized isotonic sodium chloride solution.

The jet injector described above was inserted into the ipsi-

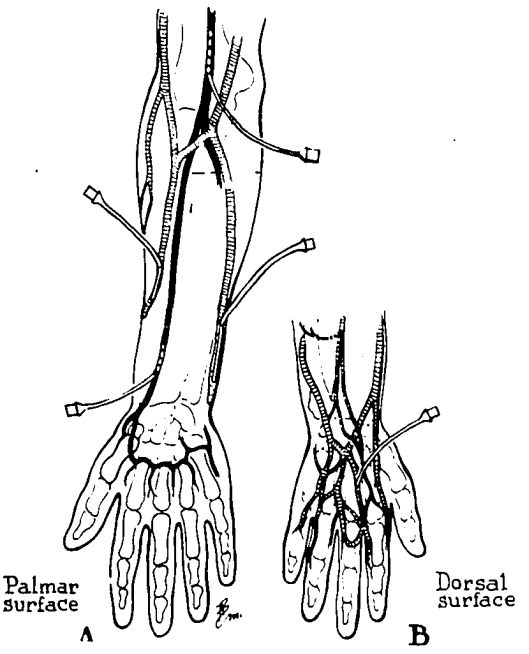

Fig. 7.-- Diagram of upper extremity vascular anatomy with catheters in place.

lateral brachial artery through the Riley needle and locked in place. The tip of the jet needle protruded up to but not proximal to the intercondylar line at the elbow. In some cases the jet injector struck the vessel wall and there was slight difficulty in introducing it to its full length, but usually manipulation of the position of the Riley needle afforded clear passage. Blood was allowed momentarily to reflux through the jet injector into the plastic tubing to show that the injector tip lay within the lumen of the artery. Infusate passed from the infusion syringe and filter through sterile polyethylene tubing to the jet injector. Infusion pressure in this tubing was monitored in some subjects to ensure that the injection orifices remained patent. In addition the patency was checked at the end of each experiment. During waiting periods, between jet infusions, heparinized isotonic sodium chloride solution

was infused at a slow rate through the jet injector to maintain patency. Everything possible was done to keep the subjects comfortable and reassured during the procedure, and they were carefully instructed not to move the ipsilateral extremity. Most subjects remained reasonably comfortable and calm in this position for the two and one-half to three hours required by the experiment. Figures 8 and 9 are photographs of a typical procedure.

Vasoactive infusions. The general protocol called for paired infusions, first an isotonic sodium chloride control solution, then the vasoactive solution in equal isotonic volume. During procedures in which the special Kimray infusion pump was used, pressures in the ipsilateral cephalic, basilic, and dorsal metacarpal veins, radial artery, and contralateral brachial artery were recorded in turn usually at three, eight, thirteen, and sixteen minutes during each infusion. Ipsilateral cephalic and basilic venous, radial arterial, and contralateral brachial arterial blood was sampled simultaneously, usually at five, ten, and fifteen minutes during each infusion. In cases where the Harvard Infusion Pump was used, pressures were recorded usually at three and five minutes, and blood samples taken at two and four minutes during each infusion. Resistance was calculated for each flow measurement, using the pressures recorded after blood samples were taken. Between pairs of infusions there was a pause of from five to thirty minutes to allow vascular resistance to return toward initial levels.

All infusions contained the same concentration of I^{131} labeled human serum albumin (Squibb - Albuminotope). Total isotope dosage per patient was less than fifty microcuries.

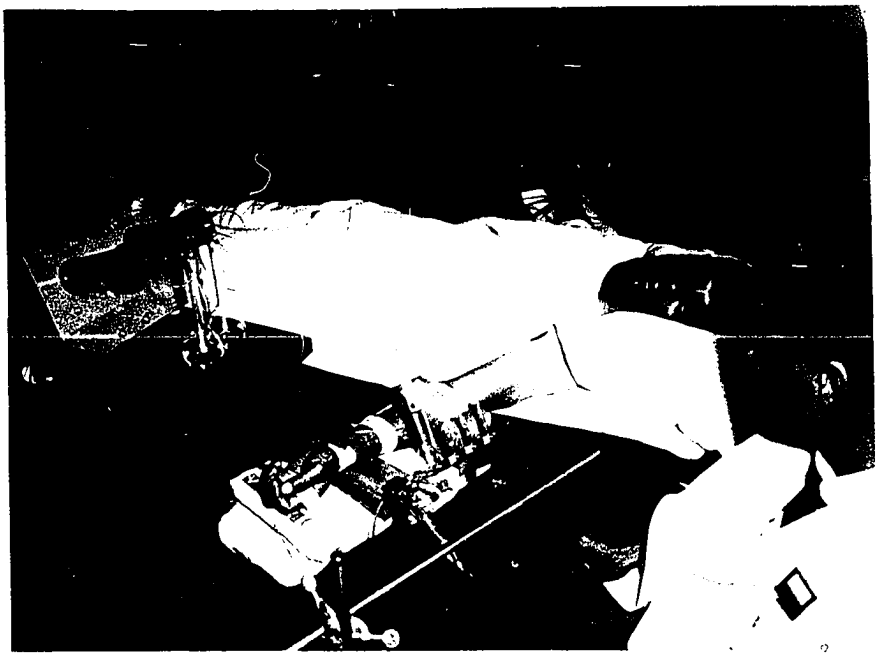

Fig. 8.-- Photograph of typical procedure.

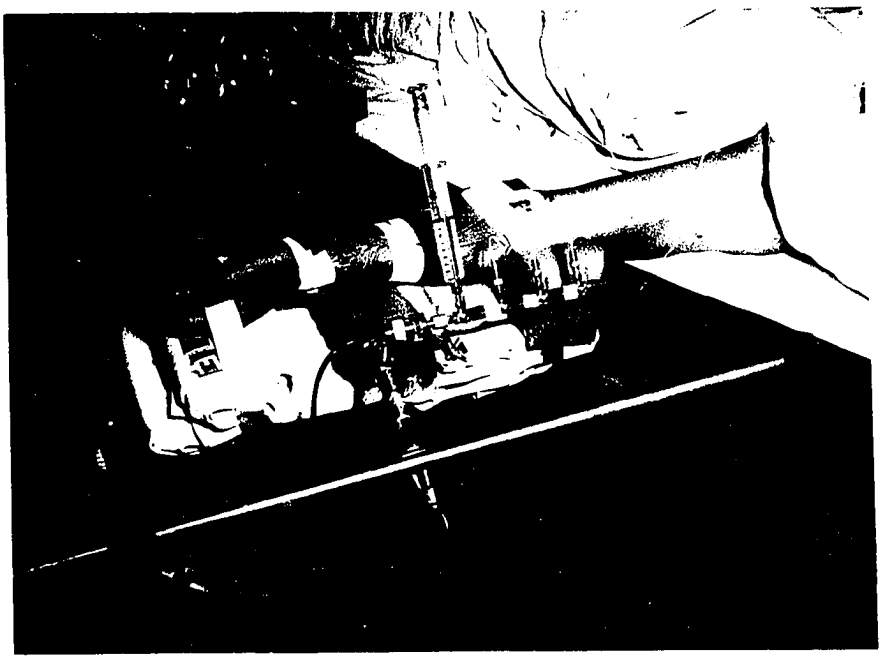

Fig. 9.-- Close-up of forearm and hand during typical procedure.

Control infusions were isotonic sodium chloride solution, eight ml. per minute, and vasoactive infusions contained the vasoactive agent, usually in isotonic solution, plus isotonic sodium chloride solution to make up the same volume. Vasoactive agents, freshly prepared for each procedure, included: (a) angiotensin, (Ciba - Hypertensin), the octapeptide, 0.025, 0.05, and 0.10 μg. per minute; (b) epinephrine, (Park, Davis and Company - Adrenalin Chloride Solution), 0.025, 0.05, and 0.10 μg. per minute; (c) ten per cent magnesium sulfate solution, (E. Lilly and Company, Magnesium Sulfate, N.F. 10 per cent), 0.2, 0.28, and 0.4 ml. per minute. Diluted with isotonic sodium chloride, the infused magnesium sulfate solution had an osmolarity of 305 mOs. per liter; (d) 150 mOs. per liter sodium chloride solution, 8 ml. per minute. During blood sampling, the cannulae and catheters were first flushed by drawing and discarding 1.5 to 2 ml. of blood, a volume at least three times the volume of the tubing. Actual drawing of samples immediately followed, each two ml. in volume, and these samples were placed in glass test tubes containing dried sodium oxylate. The tubes containing samples were rotated for at least three minutes and then one ml. aliquots were pipetted into plastic tubes for isotope counting. These specimens were counted on a Tracerlab crystal scintillation counter.

Subjects participating. All subjects participating in this study were male inpatients at the Veterans Administration Hospital, Oklahoma City, Oklahoma. Subjects had either definite arterial hypertension or normotension, and no subjects were accepted who were receiving drugs with vasoactive properties or who had clinically discernable left or right ventricular failure. No subjects were acutely ill although sev-

eral were in the afebrile convalescent phase of acute febrile illnesses. None had other peripheral vascular disease except mild degrees of asymptomatic peripheral arteriosclerosis commensurate with their ages. An attempt was made to exclude subjects having anomalous bifurcation of the brachial artery by palpating the antecubital fossae, but this was unsuccessful in a few instances.

Calculation of Blood Flow and Vascular Resistance

Figure 10, after Andres et al. (2) represents the time concentration curve of indicator in a downstream vein during constant indicator infusion into the supplying artery. The upper drawing represents the relationship between concentration and time if there is no recirculation of indicator, and it may be seen that in this case, after the plateau in

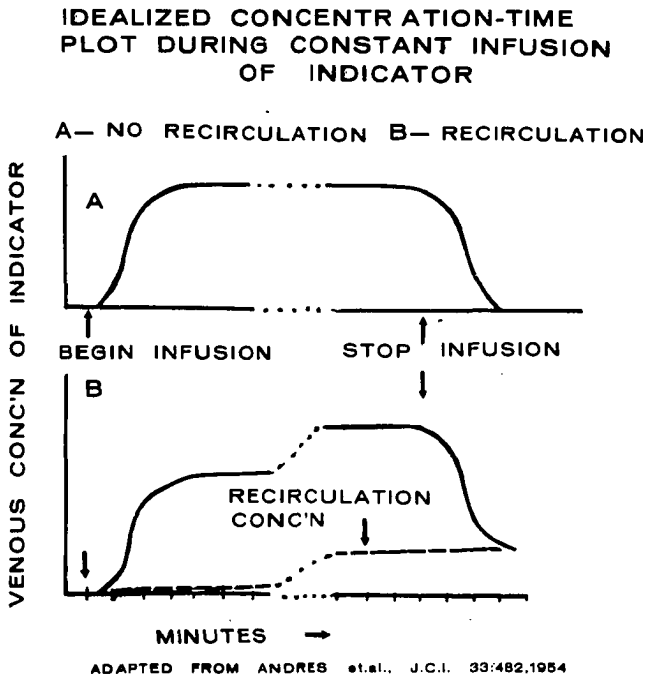

Fig. 10.--Idealized concentration-time plot during constant infusion of indicator.

concentration is reached, flow is inversely proportional to the concentration of indicator in the vein. The lower drawing represents the relationship if there is recirculation of indicator. It may be seen that recirculating indicator adds a time dependent increment to the venous concentration. To calculate flow, this increment must be subtracted from the venous concentration. The calculating equation for upper extremity blood flow is as suggested by Andres et al.:

$$\text{Flow (volume per minute)} = \frac{\text{Quantity of indicator infused (weight per minute)}}{\text{Mean venous indicator concentration (weight per volume)} - \text{Mean arterial indicator concentration (weight per volume)}}$$

which, in case of this present study, using I^{131} labeled human serum albumin (RIHSA) as indicator becomes:

$$\text{Upper Extremity Blood Flow (F)} = \frac{\text{CPM RIHSA infused per minute into brachial artery}}{\text{Mean CPM per ml. venous blood} - \text{CPM per ml. contralateral brachial arterial blood}}$$

It should be noted that the mean indicator concentration of the paired venous samples is used in calculation of blood flow.

Calculated upper extremity blood flow is expressed as ml. per 100 cc. upper extremity volume per minute. Total upper extremity vascular resistance is calculated as follows:

$$\text{Upper Extremity Vascular Resistance} = \frac{\bar{P}_{BA} - \bar{P}_{LV}}{F}$$, and is

expressed as mm. Hg. per ml. blood flow per 100 cc. upper extremity volume per minute. Upper extremity segmental vascular resistances may also be calculated as follows:

Venous Resistance $= \frac{\bar{P}_{SV} - \bar{P}_{LV}}{F}$;

Arterial Resistance $= \frac{\bar{P}_{BA} - \bar{P}_{RA}}{F}$, where $\bar{P}_{BA}$, $\bar{P}_{RA}$, $\bar{P}_{LV}$, $\bar{P}_{SV}$, and F represent mean brachial arterial pressure, mean radial arterial pressure, mean cephalic or basilic venous pressure, mean dorsal metacarpal venous pressure, and total upper extremity blood flow per 100 cc. extremity volume per minute, respectively.

The Student's t test, paired or unpaired depending on the particular experimental design, was used for statistical analyses of the data.

CHAPTER III

RESULTS

In Vitro Experiments

Figures 11a through 14b are high speed flash photographs of Evans Blue dye being infused at eight ml. per minute through either the jet injector or a standard twenty gauge hypodermic needle against the flow of water in glass tubing from left to right at water flow rates of twenty five, fifty, one hundred, and two hundred ml. per minute. It may be seen that, at all flows, mixing of indicator with water is improved by the jet injector. This is especially true at the high flow rates. It may also be seen that most mixing occurs as a result of "rebounding" of the jet stream from the walls of the tube into the main stream. This "rebounding" would not be present in the end-orifice type of jet injector used by Andres *et al.* (2). Figures 15 (a and b) illustrate that mixing is improved at flows of either one hundred or two hundred ml. per minute even if the jet injector lies against a wall of the tubing, a situation likely to occur *in vivo*. Figures 16 (a, b, and c) were taken at different phases of the pump cycle and illustrate the effect of pulsatile flow upon indicator dispersion. In this case pump flow and stroke frequency were 100 ml. per minute and 112 per minute, respectively. It will be seen that there are regions of greater concentration of indicator (pump diastole) and of lesser concentration (pump systole). These regions move downstream with

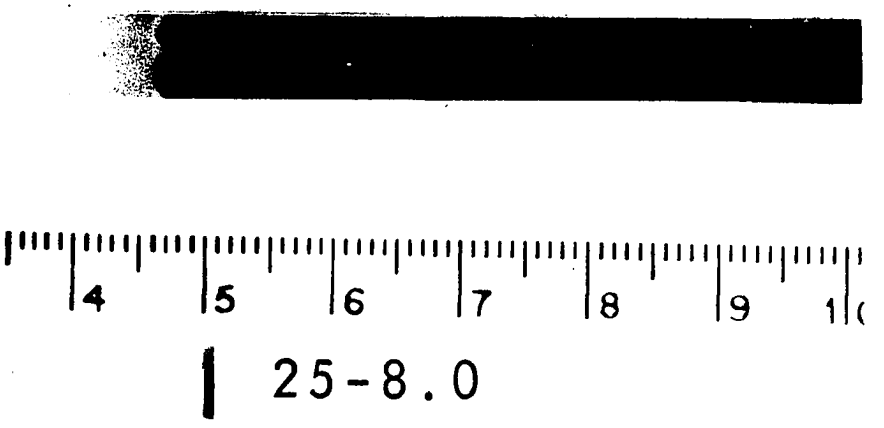

Fig. 11. a-- Infusion of Evans Blue Dye through jet injector into water flowing from left to right at 25 ml. per minute.

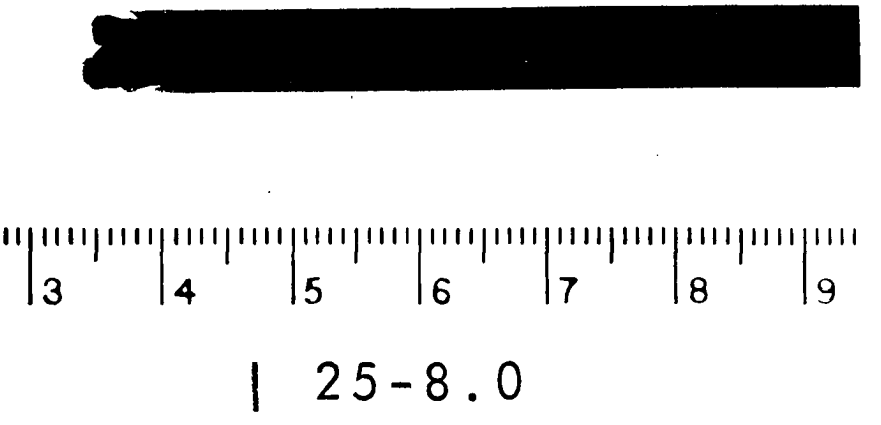

Fig. 11. b-- Infusion of Evans Blue Dye through standard 20 gauge needle into water flowing from left to right at 25 ml. per minute.

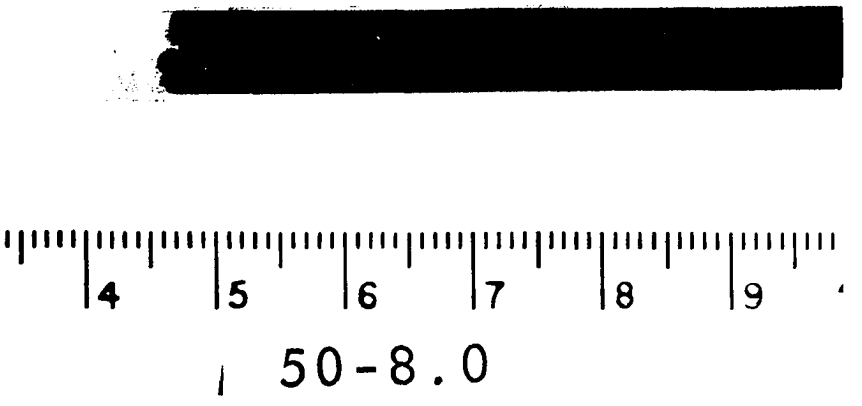

Fig. 12. a-- Infusion of Evans Blue Dye through jet injector into water flowing from left to right at 50 ml. per minute.

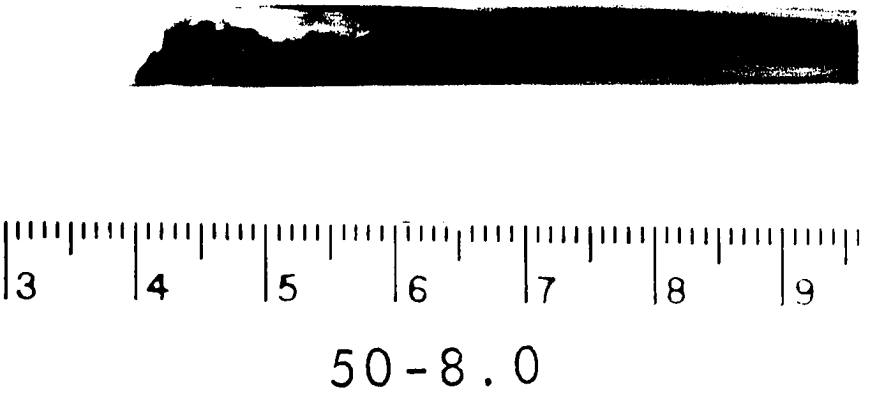

Fig. 12. b-- Infusion of Evans Blue Dye through standard 20 gauge needle into water flowing from left to right at 50 ml. per minute.

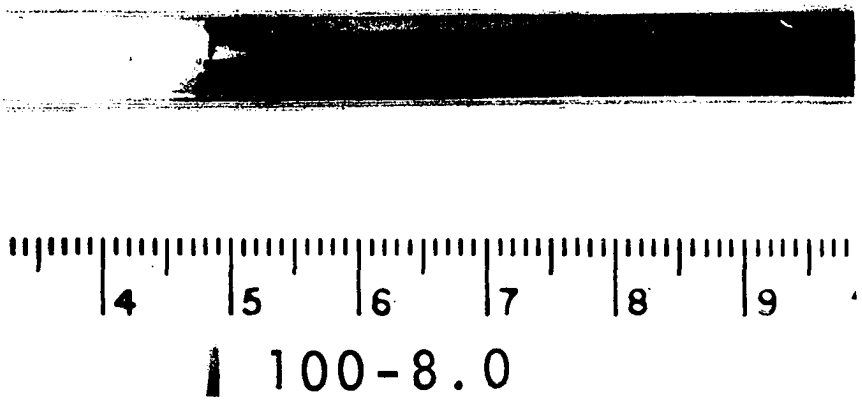

Fig. 13. a-- Infusion of Evans Blue Dye through jet injector into water flowing from left to right at 100 ml. per minute.

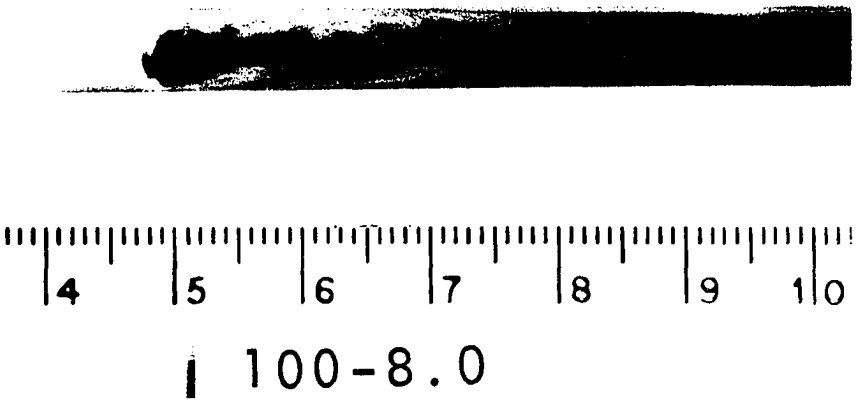

Fig. 13. b-- Infusion of Evans Blue Dye through standard 20 gauge needle into water flowing from left to right at 100 ml. per minute.

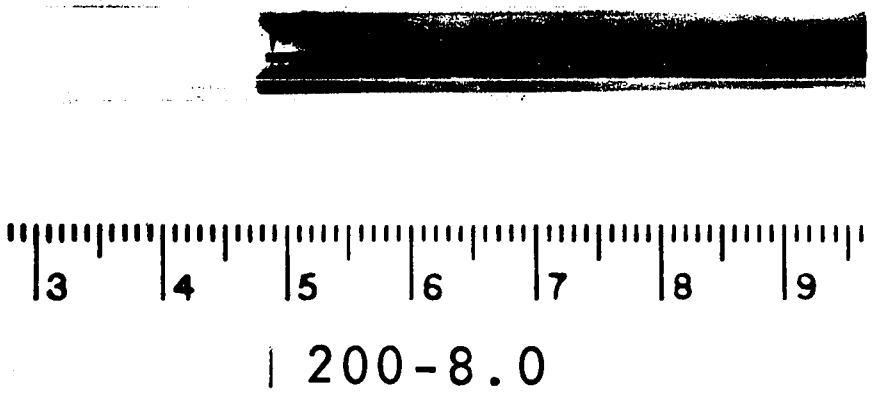

Fig. 14. a-- Infusion of Evans Blue Dye through jet injector into water flowing from left to right at 200 ml. per minute.

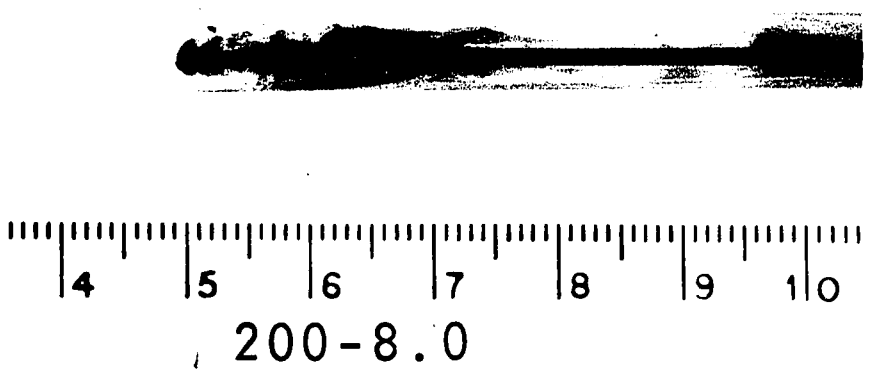

Fig. 14. b-- Infusion of Evans Blue Dye through standard 20 gauge needle into water flowing from left to right at 200 ml. per minute.

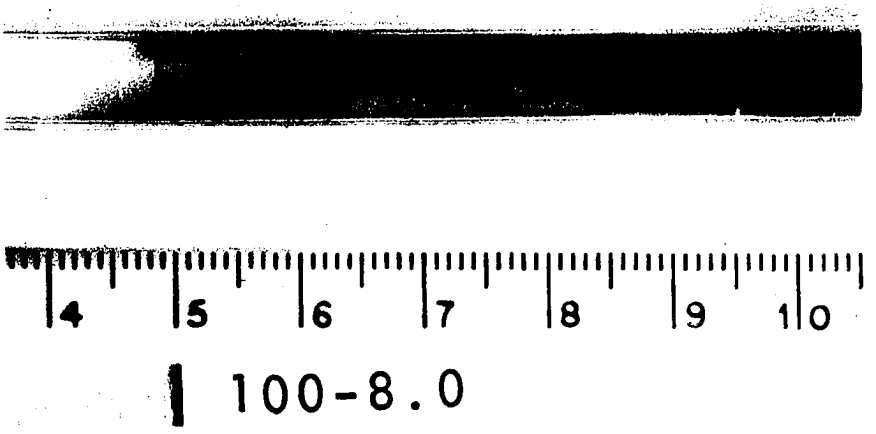

Fig. 15. a-- Infusion of Evans Blue Dye through jet injector into water flowing from left to right at 100 ml. per minute.

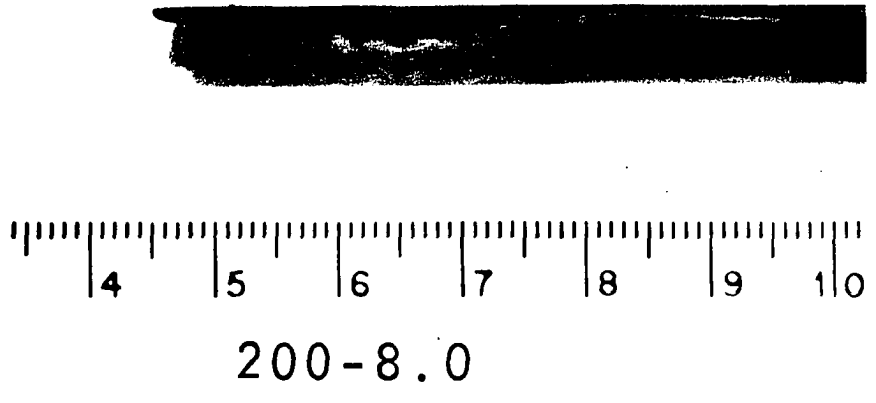

Fig. 15. b-- Infusion of Evans Blue Dye through jet injector into water flowing from left to right at 200 ml. per minute.

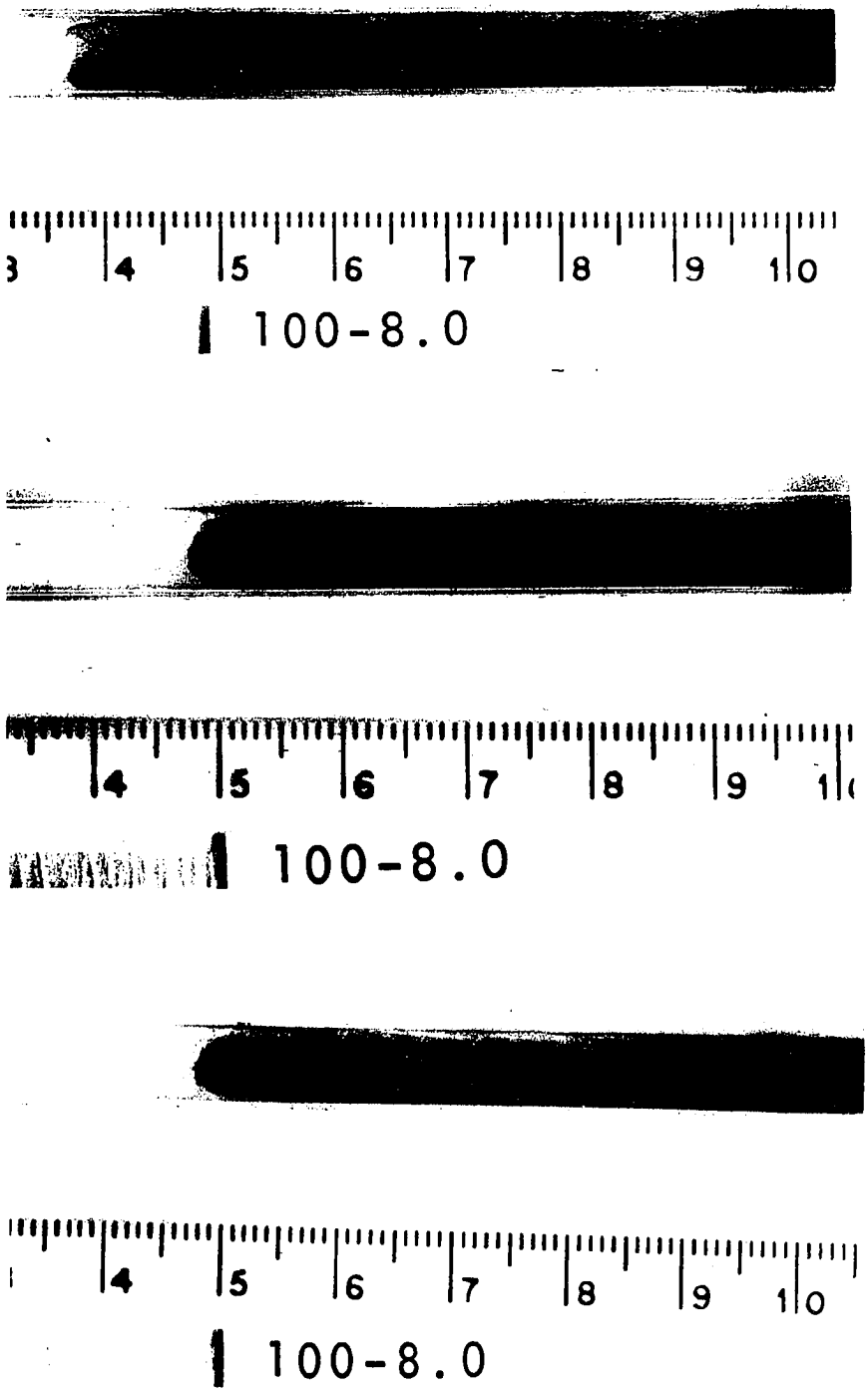

Fig. 16. a,b,c--Infusion of Evans Blue Dye through jet injector into water flowing from left to right at 100 ml. per minute, at sequential phases of the pump cycle.

time and change in configuration.

In Vivo Experiments

Dogs

Hemolysis. Figure 17 is representative of the pump pressure tracings obtained in the pump-perfused forelimbs of seven dogs during intrabrachial arterial infusions. The top panel is the pump pressure tracing obtained during intra-brachial arterial infusion of isotonic sodium chloride solution at eight ml. per minute through a standard twenty gauge hypodermic needle. The center panel is the pressure tracing obtained during an identical infusion through the jet injector. It may be seen that neither infusion significantly altered perfusion pressure, or, therefore, limb vascular resistance, either immediately upon starting or

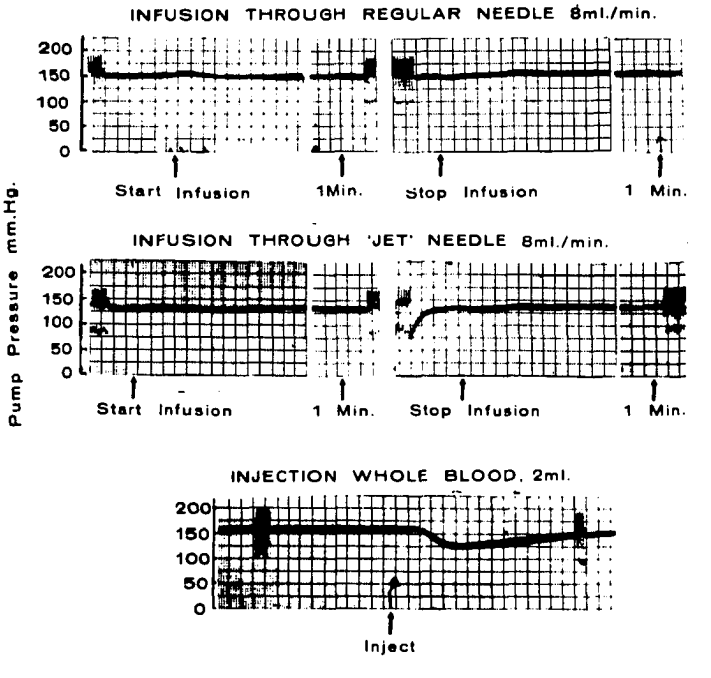

Fig. 17. --Pump pressure tracings during infusion through regular and jet needles into pump perfused dog forelimb.

stopping the infusion, or after one minute. However the limb vascular bed was responsive to products of hemolysis, as indicated in the bottom panel by vasodilation following intra-brachial arterial injection of two ml. of whole blood through a twenty seven gauge hypodermic needle at a velocity which hemolyses the injected blood.

Comparison of actual with calculated flows. Standard Hypodermic needle. Indicator was infused through a twenty gauge standard hypodermic needle at approximately one ml. per minute into the pump perfused brachial arteries of seven dogs. Table 2 itemizes and Table 3 summarizes these experiments. In five of the seven dogs only the pump inflow was measured. It was then realized that collateral circulation through the bone may contribute a significant proportion of the limb blood flow, especially at low pump flows. At these low pump flows perfusion pressures may be lower than systemic blood pressures, favoring bone flow which is supplied at the higher systemic perfusion pressures. The venous outflow was also measured in the two final dogs and only in these two dogs may a valid comparison be made between actual and calculated flow. However in all seven dogs measurements of m.r.d. are valid. It may be seen that in two dogs the calculated flows averaged 44.4 per cent above the actual flows, and that in seven dogs the m.r.d.'s averaged 18.0 per cent. In all of these experiments the standard Harvard Infusion Pump was used to infuse indicator, and in none of these experiments was the humerus cut.

Jet injector needle. These results should be compared to the results in ten other dogs itemized in Table 4 and summarized in Table 3. In these experiments indicator was infused through the jet injector at eight ml. per minute into the pump perfused brachial arteries. It may be

TABLE 2

COMPARISON OF ACTUAL AND CALCULATED FLOWS IN PUMP PERFUSED DOG FORELIMBS USING STANDARD NEEDLE INJECTOR

Dog No.	Bone Cut	"Actual Flow" ml.per minute		Mean Calc. Flow ml.per minute		$\frac{\text{Act.-Calc.}}{\text{Act.}} \times 100\%$
		Pump	Venous	Total	r.d.%	
1NJ	No	136.0	..	136.4	19.9	..
		96.0	..	104.0	7.2	..
		60.0	..	150.6	56.0	..
2NJ	No	35.5	..	64.3	39.0	..
		82.0	..	92.5	12.4	..
		136.0	..	163.1	9.0	..
3NJ	No	93.0	..	94.7	12.4	..
		74.0	..	77.7	9.9	..
		93.0	..	90.6	3.0	..
		74.0	..	70.7	13.0	..
4NJ	No	74.0	..	114.2	30.1	..
		98.0	..	114.2	10.3	..
5NJ	No	51.0	..	205.3	9.5	..
6NJ	No	88.0	96.5	91.7	1.9	- 3.8
7NJ	No	66.0	87.0	139.8	41.3	+ 60.7
		110.0	121.0	229.3	28.9	+ 89.5
		165.0	160.0	210.1	2.2	+ 31.3
$\bar{x}$ ($\pm$ S.E.)					18.0 $\pm$ 3.8	46.3
Signed $\bar{x}$						+ 44.4

TABLE 3

SUMMARY OF COMPARISON OF ACTUAL AND CALCULATED FLOWS IN PUMP PERFUSED DOG FORELIMBS

Standard Needle

N			Mean		
Dogs	Observations	Comment	m.r.d.	% Difference	Signed % Difference
5	13	Venous flow not measured	17.8	..	..
2	4	Venous flow measured	18.6	46.3	+ 44.4
7	17	Not Edited	18.0	..	..

Jet Needle

2	4	Bone Not Cut	9.0	9.1	+ 4.4
8	56	Bone Cut	3.5	7.7	+ 2.9
5[a]	30	Technically Satisfactory	3.6	4.1	+ 1.3
5[b]	30	Technically questionable	4.2	11.4	+ 4.7
10	60	Not Edited	3.9	7.8	+ 3.0

[a]See Figure (19)

[b]See Figure (18)

seen that for all animals calculated flows averaged 3 per cent above the actual flows, and that the m.r.d.'s averaged 7.8 per cent. In dogs 1J and 2J the standard Harvard Infusion Pump was used. In the remainder, the modified Harvard pump was used. In dogs 1J and 2J the humerus was not cut and in the remainder the humerus was cut.

Table 4 cites some technical difficulties encountered in five of these experiments. In four cases, dogs 1J, 4J, 8J, and 9J, the errors would tend to make the calculated flows erroneously high. These errors include loss of indicator from the tubing or syringe, faulty operation of the pump, or sticking of the syringe. In one case, dog 10J, a fast stopwatch caused faulty calibration and therefore the calculated flows were erroneously low. Figure 18 represents a comparison of actual and calculated flows in these five dogs with recognised technical problems. It will be seen that even in these dogs all but one of the thirty observations lay within $\pm$ 20 per cent of actual flow, and that four observations lay within $\pm$ 5 per cent of the actual flow. Figure 19 represents a similar comparison in the case of the remaining five dogs, where no technical error was detected. It will be seen that all but two of these thirty observations lay within $\pm$ 10 per cent of the actual flow and 77 per cent lay within $\pm$ 5 per cent of the actual flow. Table 3 summarizes the overall figures for these ten experiments with the jet injector. It may be seen that in all subgroups there is a clear decrease in m.r.d. and per cent difference between actual and calculated flows as compared to similar values in the dogs in which the standard needle was used. In addition the data suggest that cutting the humerus may have contributed to improved accuracy of calculation and decreased m.r.d.

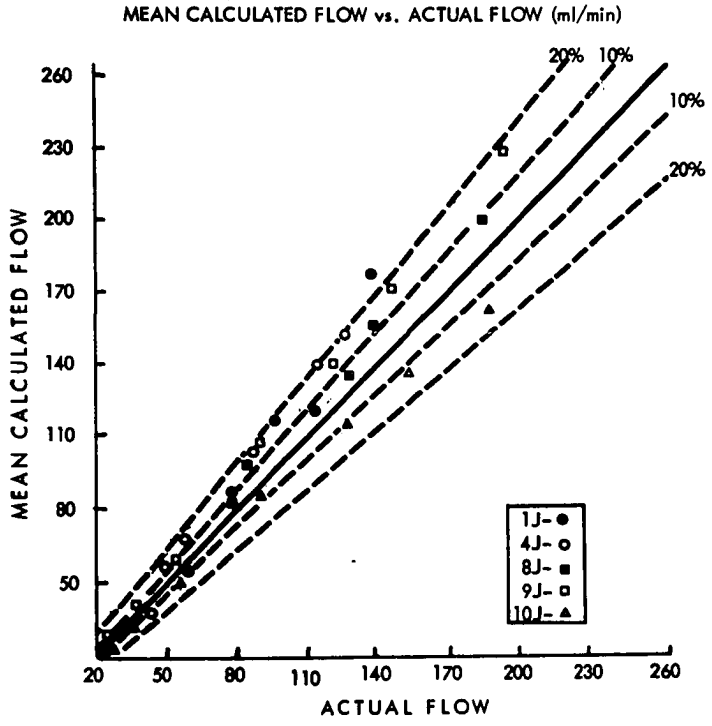

Fig. 18.-- Mean calculated flow vs. actual flow (technical errors present).

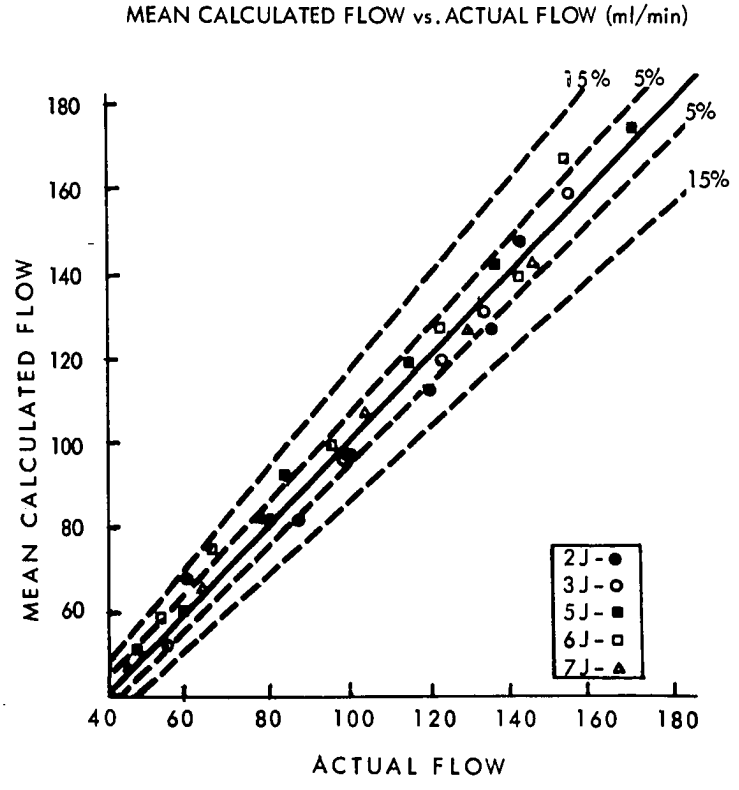

Fig. 19.-- Mean calculated flow vs. actual flow

TABLE 4

COMPARISON OF ACTUAL AND CALCULATED FLOWS IN PUMP PERFUSED DOG FORELIMBS USING JET INJECTORS

Dog No.	Bone Cut	Actual Flow			Calc. Flow		$\frac{\text{Act.} - \text{Calc.}}{\text{Act.}} \times 100\%$	Remarks
		Basilic Vein	Cephalic Vein	Total	Total	r.d.%		
1J	No	..	..	58.0	55.0	9.1	- 5.2	Standard Harvard Infusion Pump Used. Faulty Operation
		..	..	78.0	84.2	2.6	+ 7.9	
		..	..	98.0	115.0	5.2	+ 17.3	
		..	..	114.0	118.8	4.5	+ 4.2	
		..	..	140.0	179.8	10.5	+ 28.4	
2J	No	..	..	59.0	66.6	26.4	+ 12.9	Standard Harvard Infusion Pump Used.
		..	..	88.5	83.4	13.9	- 5.8	
		..	..	100.5	98.0	12.1	- 2.5	
		..	..	119.5	111.8	8.3	- 6.4	
		..	..	135.0	127.6	4.2	- 5.5	
		..	..	144.0	149.1	2.2	+ 3.5	
3J	Yes	..	..	54.0	51.8	11.0	- 4.1	
		..	..	80.5	81.4	5.2	+ 1.1	
		..	..	100.0	97.9	5.4	- 2.1	
		..	..	122.5	118.1	3.6	- 3.6	
		..	..	133.0	129.9	0.2	- 2.3	
		..	..	155.5	158.8	1.7	+ 2.1	

TABLE 4 - Continued

4J	Yes	4.0	46.0	50.0	57.2	9.8	+ 14.4	Stopcock Leakage. Syringe Sticking. BV Flow Poor Initially.
		7.0	53.0	60.0	64.8	6.0	+ 8.0	
		15.5	72.0	87.5	101.6	5.9	+ 16.1	
		35.5	79.5	115.0	137.9	1.7	+ 19.9	
		59.0	68.5	127.5	149.6	1.7	+ 17.3	
5J	Yes	119.0	52.0	171.0	173.5	0.6	+ 1.5	
		94.0	42.0	136.0	142.4	1.3	+ 4.7	
		80.0	35.0	115.0	118.4	0.9	+ 3.0	
		51.0	34.0	85.0	90.2	0.4	+ 6.1	
		33.5	25.5	59.0	59.6	0.5	+ 1.0	
		25.0	22.0	47.0	49.1	0.0	+ 4.5	
6J	Yes	72.0	82.0	154.0	164.6	1.7	+ 6.9	
		66.0	76.0	142.0	138.7	0.4	- 2.3	
		54.0	68.0	122.0	126.2	2.2	+ 3.4	
		38.5	57.0	95.5	98.4	1.0	+ 3.0	
		23.0	42.0	65.0	74.7	0.0	+ 14.9	
		16.0	38.0	54.0	56.6	0.4	+ 4.8	
7J	Yes	91.0	56.0	147.0	141.6	0.4	- 3.7	
		82.0	48.5	130.5	126.0	1.7	- 3.4	
		67.0	36.5	103.5	105.8	0.7	+ 2.2	
		40.0	23.0	63.0	64.0	0.2	+ 1.6	
		50.5	29.5	80.0	81.4	0.7	+ 1.8	
		22.5	21.5	44.0	44.4	0.6	+ 0.9	

TABLE 4 - Continued

8J	Yes	16.0	24.0	40.0	39.2	14.5	- 2.0	Syringe Leaking.
		82.0	45.5	127.5	135.0	2.4	+ 5.9	
		46.8	32.0	78.8	81.0	1.9	+ 2.8	
		124.0	65.0	189.0	202.0	1.6	+ 6.9	
		50.0	34.8	84.8	95.8	0.6	+ 13.0	
		90.0	49.0	139.0	155.8	2.8	+ 12.1	
9J	Yes	60.0	63.0	123.0	139.3	6.5	+ 7.2	Syringe Sticking.
		4.0	18.0	22.0	25.8	10.1	+ 17.3	
		96.2	100.8	197.0	232.8	0.3	+ 18.2	
		17.9	36.5	54.4	58.0	3.3	+ 6.6	
		84.0	66.0	150.0	170.4	2.0	+ 13.2	
		44.3	44.5	88.8	104.8	3.2	+ 18.0	
		13.5	22.3	35.8	40.6	4.2	+ 13.4	
10J	Yes	58.8	72.0	130.8	117.2	1.8	- 10.4	Stopwatch Faulty. Erroneous Calibration of Indicator
		8.8	14.7	23.5	20.8	0.0	- 11.5	
		97.0	95.5	192.5	167.8	1.2	- 12.8	
		29.0	27.0	56.0	50.5	1.4	- 9.8	
		74.3	77.5	151.8	136.0	6.1	- 10.4	
		44.8	46.0	90.8	86.6	4.7	- 4.6	
		13.4	21.7	35.1	32.6	1.5	- 7.1	

Man

Subjects participating. Table 5 presents clinical data on the fifty subjects participating in this study. In the case of hypertensive subjects, the clinically estimated degree of severity is noted. Three subjects participated twice in the procedure: A.L., J.E.C., and G.E.

For several reasons data on some subjects were excluded from some or all of the tables presented in this section. For example, there was occasionally considerable difficulty with sticking of the glass syringes. This sticking caused inconstant infusion of indicator, and for this reason most data on subjects R.L.A. and L.T. were not used. In several subjects the orifices of the jet injector lay outside the lumen of the brachial artery and indicator was infused subcutaneously. In these cases blood would not reflux through the needle and there was subcutaneous swelling during the infusion. For this reason most data on subjects G.E.-2, E.C.M., and B.G.L. were not used. In several subjects it was not possible to draw samples from both veins during all or a part of the procedure. For this reason all or a portion of the data on subjects W.O.D., N.P.L., and F.M.H. was not used.

All or a portion of the data accumulated during study of response to epinephrine and angiotensin in subjects E.L.F., N.P.L., L.S., F.F., G.E.-1, and L.P. was discarded because a steady state of limb blood flow had not been achieved prior to some vasoactive infusions. In most cases this was due to persistance of changes caused by a previous infusion.

In a group of subjects there was evidence that some or all of the infused indicator was lost from the limb bed due to vascular shunting around the elbow. A striking case of this was subject D.H.R., in whom

TABLE 5

SUBJECT LIST

Subject	Date	Age	Race/ Sex	Wt. Kg.	Limb Volume cc.	Diagnosis	Drugs	Laboratory	
								Hct.	BUN
R.L.A.	26 Mar.64	49	N/M	80.5	1850	Essential Hypertension (moderate)	Digitoxin, Phenobarbital	50	12
J.D.H.	26 Mar.64	42	N/M	72.7	1700	Essential Hypertension (mild)	Digitoxin	47	10
R.T.	27 Mar.64	23	W/M	96.8	1750	Essential Hypertension (moderate)	None	50	17
F.G.D.	9 Apr.64	51	W/M	98.6	1850	Essential Hypertension (moderate)	None	50	13
J.E.C.	9 Apr.64 19 May 65	36 37	W/M	81.1	1700 1525	Essential Hypertension (mild)	None	47	8
I.S.	23 Apr.64	40	N/M	68.2	1550	Essential Hypertension (moderate)	None	46	13
W.W.Z.	21 May 64	47	W/M	82.3	1450	Essential Hypertension (mild)	None	50	14
A.L.	4 Jun.64 7 Jan.66	41 42	N/M	85.1	1625 1650	Essential Hypertension (moderate)	None	53	10
I.L.	18 Jun.64	48	W/M	66.4	1300	Essential Hypertension (moderate), Pulmonary emphysema, Inactive duodenal ulcer	Digitoxin	49	14
O.H.P.	18 Jun.64	57	N/M	71.5	1575	Essential Hypertension (mild), Diabetes Mellitus	None	37	32

TABLE 5 - Continued

W.M.T.	19 Jun.64	72	W/M	68	1250	Essential Hypertension (moderate), Old Subarachnoid hemorrhage	None	44	22
M.C.	16 Jul.64	42	W/M	85	1875	Essential Hypertension (mild), Chronic Pyelonephritis	None	51	10
E.A.K.	16 Jul.64	68	W/M	60.5	1300	Essential Hypertension (mild)	Digitoxin	44	19
W.P.	17 Jul.64	66	M/M	88.4	1950	Essential Hypertension (mild) Old cerebral vascular accident	None	39	21
L.T.	30 Jul.64	47	M/M	66.4	2075	Essential Hypertension (mild), Chronic bronchitis	None	42	16
H.B.	24 Sep.64	53	W/M	75	1500	Essential Hypertension (mild)	None	47	13
F.F.	24 Sep.64	40	M/M	115.9	2350	Essential Hypertension (mild)	None	45	13
L.P.	8 Oct.64	69	W/M	72.7	1500	Essential Hypertension (mild), Diabetes Mellitus	Chlorpropamide, Insulin	45	21
W.J.S.	9 Oct.64	57	M/M	88.6	1800	Essential Hypertension (moderate), Diabetes Mellitus, Chronic pyelonephritis	None	47	10
G.E.	30 Oct.64 8 Dec.65	51 52	W/M	67.7	1325 1325	Essential Hypertension (mild), Chronic pyelonephritis	None	41	13
L.C.B.	11 Dec.64	34	M/M	70	1525	Essential Hypertension (mild)	None	45	20
W.E.S.	8 Jan.65	32	W/M	94.5	2025	Essential Hypertension (moderate to severe)	None	47	14

TABLE 5 - Continued

R.A.	15 Jan.65	43	W/M	67	1550	Diabetes Mellitus, Duodenal ulcer	Chlordiazepoxide, Insulin	35	10
W.A.F.	22 Jan.65	71	W/M	51.6	1125	Cholelithiasis Pyelonephritis	None	42	15
W.D.	22 Jan.65	49	W/M	68.2	1500	Silicosis	Penicillin, Streptomycin, Ferrous Sulfate, SS-KI	44	18
S.W.P.	5 Feb.65	45	W/M	91.8	2150	Essential Hypertension (mild)	None	47	16
E.L.F.	12 Feb.65	31	W/M	69	1500	Gastritis, Urethritis	Meprobamate, Phenylbutazone, Urecholine	46	11
J.J.	12 Feb.65	50	N/M	73.6	1850	Subsiding Pneumonitis, Bronchial Asthma	Streptomycin, Penicillin, Ephedrin, SS-KI	39	11
E.C.M.	26 Feb.65	46	W/M	84.5	1725	Chronic Bronchitis, Arteriosclerotic heart disease	Digitoxin, Tetracycline	46	19
F.L.Z.	26 Feb.65	61	W/M	62.7	1500	Lymphoma	Chloral hydrate	42	18
R.C.F.C.	5 Mar.65	50	W/M	86.4	1600	Ulcerative Colitis, Schizophrenia	Chlorpromazine	38	21
N.P.L.	10 Mar.65	56	W/M	70.2	1500	Chronic Pyelonephritis	Sulfisoxazole	47	12
E.P.	17 Mar.65	59	N/M	54.5	1100	Adenocarcinoma	None	42	10
L.S.	19 Mar.65	31	W/M	65	1225	Psychoneurosis	None	44	9
M.P.J.	31 Mar.65	60	W/M	53.9	1250	Diabetes Mellitus, Chronic Bronchitis	Tetracycline, Tolbutamide	42	12
B.G.L.	2 Apr.65	51	W/M	67.7	1325	Broncho-pneumonia	Erythromycin	44	21
R.H.T.	16 Apr.65	47	W/M	77.8	1300	Diabetes Mellitus	None	44	15

TABLE 5 - Continued

W.C.W.	21 Apr.65	52	W/M	87.0	1700	Diabetes Mellitus	None	47	11
L.M.	12 May 65	51	W/M	68	1325	Gastric Ulcer	Phenobarbital	53	..
S.G.	8 Oct.65	73	W/M	91.1	1900	Metastatic Adeno-carcinoma	None	48	20
C.T.L.	2 Nov.65	48	N/M	70	1900	Femoral Neuritis	Diazepan, Hydroxyzine	44	20
L.C.H.	5 Nov.65	30	N/M	76	1800	Myalgia, probably viral	None	45	10
W.O.D.	17 Nov.65	36	W/M	59	1300	No disease	Tetracycline	40	..
F.M.H.	19 Nov.65	55	W/M	65.6	1400	Essential Hypertension (mild)	None	50	25
T.K.	24 Nov.65	52	W/M	87.5	1525	Essential Hypertension (mild)	Digitoxin	46	18
C.G.R.	15 Dec.65	44	W/M	64	1425	Cervical spondylosis	Methocarbamol	47	20
J.L.G.	17 Dec.65	38	N/M	56.4	1400	Subsiding Acute Gastroenteritis	None	42	12
W.H.	12 Jan.66	48	W/M	77.7	1850	Gastric Ulcer	Gelusil, Phenobarbital	42	..
H.W.M.	14 Jan.66	49	W/M	86.8	1900	Depression	None	44	24
D.H.R.	19 Jan.66	37	I/M	70.9	1425	Essential Hypertension (severe)	None	47	20

blanching of a five by six centimeter area of skin proximal to the infusion site was noted during vasoactive infusions. Later it was found that concentrations of isotope in the veins of the limb were only slightly greater than recirculation concentrations. In this case the tip of the jet injector probably entered a small branch of the brachial artery at the elbow, blood from which did not enter the forearm. In other cases evidence of shunting was less clear-cut, but, because the calculated flows were unrealistically high in these cases, it was felt that a significant proportion of indicator had not reached the forearm vessels. Such cases were S.G., and S.W.P. Data from these three cases are presented in Table 6 and excluded from most other tables.

TABLE 6

SUSPECTED SHUNTING OF INDICATOR AT ELBOW. MEAN CALCULATED UPPER EXTREMITY BLOOD FLOW AND MEAN RELATIVE DIFFERENCE IN PER CENT IN CONCENTRATION OF INDICATOR IN PAIRED VENOUS SAMPLES

Subject	Observations	Mean Calculated		m.r.d.,%
		Total Flow ml/min	ml/100cc/min	
D.H.R.	6	∞	∞	11.4
S.W.P.	5	380.6	17.7	7.6
S.G.	7	407.9	21.5	5.1

Finally, in subjects W.D., O.H.P., F.L.Z., J.E.C., and R.H.T. there was good evidence of anomalous bifurcation of the brachial artery. In these cases there was a considerable difference between the concentrations of indicator in the paired venous samples. Table 7 gives data

TABLE 7

SUSPECTED ANOMALOUS BIFURCATION OF THE BRACHIAL ARTERY

Subject	No Observations	Mean Indicator Concentration (CPM) During Resting Flow				m.r.d. % BV vs. CV
		Contralateral Brachial Artery	Radial Artery	Cephalic Vein	Basilic Vein	
W.D.	5	2766	..	13227	4235	75.4
O.H.P.	5	1028	..	1114	4728	95.4
F.L.Z.	5	2092	..	5561	9429	35.8
J.E.C.-2	11	3861	3823	10710	15878	27.4
R.H.T.	7	2936	2944	3112	7151	91.9

on these five subjects. It is interesting to note that these five subjects comprise about ten per cent of all subjects studied, whereas it is known that twenty per cent of humans have this anomaly. Data from these subjects were excluded from most tables.

Use of the technique to study upper extremity resting blood flow. Comparison of mixing obtained with standard and jet injectors. Indicator was infused through a standard twenty gauge arterial needle into the brachial arteries of ten subjects and through the jet injector into the brachial arteries of fourteen subjects, in whom more than one measurement of resting blood flow was made. The resulting calculated mean relative differences and mean resting blood flows are presented in Tables 8 and 9. In the cases of J.E.C.-1 and O.H.P. with the standard needle and of R.H.T. with the jet injector, results were questionable for reasons outlined above. From Table 8 it may be seen that the m.r.d. is significantly de-

TABLE 8

COMPARISON OF MIXING EFFECTIVENESS
OF JET AND STANDARD INJECTORS

Mean Relative Difference, Per Cent, Between
Indicator Concentrations in Paired Venous Samples

Standard Needle		Jet Needle	
Subject	m.r.d. %	Subject	m.r.d. %
I.S.	6.5	C.T.L.	5.2
J.D.H.	28.9	W.H.	1.5
F.G.D.	7.6	J.L.G.	3.6
R.T.	14.3	L.C.H.	9.0
W.W.Z.	25.2	H.W.M.	11.2
I.L.	17.6	C.G.R.	5.5
W.M.T.	3.4	A.L.-2	1.7
A.L.-1	21.6	T.K.	5.9
J.E.C.-1[a]	48.0	W.C.W.	2.4
O.H.P.[a]	100.0	L.M.	3.2
		M.J.	1.6
		S.G.	4.5
		G.E.-1	22.4
		R.H.T.[a]	100.0
$\bar{x} \pm$ S.E.	27.31 ± 9.07		12.69 ± 6.88
P	.15 - .10[b]		
$\bar{x} \pm$ S.E.[a]	15.64 ± 3.29		5.98 ± 1.59
P	.05 - .02[b]		

[a]Subjects with proved or highly suspected anomalies, excluded from edited totals.

[b]Alternate hypothesis $u_1 \neq u_2$.

TABLE 9

COMPARISON OF CALCULATED BLOOD FLOW USING JET AND STANDARD INJECTORS

Calculated Mean Resting Blood Flow
ml/100cc/min

Standard Needle		Jet Needle	
Subject	Flow	Subject	Flow
I.S.	5.3	C.T.L.	6.7
J.D.H.	3.3	W.H.	6.7
F.G.D.	3.0	J.L.G.	5.7
R.T.	6.0	L.C.H.	5.2
W.W.Z.	4.5	H.W.M.	7.3
I.L.	6.6	C.G.R.	5.6
W.M.T.	3.9	A.L.-2	3.0
A.L.-1	7.2	T.K.	10.4
J.E.C.-1[a]	15.8	W.C.W.	5.0
O.H.P.[a]	..	C.M.	9.7
		M.J.	9.4
		S.G.	26.7
		G.E.-1	13.1
		R.H.T.[a]	..
$\bar{x} \pm$ S.E.	6.18 $\pm$ 1.30		8.81 $\pm$ 1.67
P	.20 - .30[b]		
$\bar{x} \pm$ S.E.[a]	4.98 $\pm$ 0.55		8.81 $\pm$ 1.67
P	$>$.05[b]		

[a]Subjects with proved or highly suspected anomalies, excluded from edited totals.

[b]Alternate hypothesis $u_1 \neq u_2$.

creased by the jet injector in the edited subjects. Note especially subject A.L. in whom both needles were used and in whom m.r.d. decreased from 21.6 per cent to 1.7 per cent. It may also be seen in Table 9 that calculated mean resting upper extremity blood flows using the two injection systems are not significantly different, either edited or non-edited. Mean resting blood flow determined using the jet injector in this group was 8.8 ml. per 100 cc. extremity volume per minute. Excluding subject S.G., in whom there was evidence for vascular shunting of indicator at the elbow, mean resting blood flow for this group was reduced to 7.3 ml. per 100 cc. per minute. These results may be compared to those of Andres et al. (2) in a table of cummulative per cent of subjects, Table 10. It will be seen that m.r.d. is lower in the present study, jet or standard needle, even in non-edited data, than in the study of Andres et al.

Constancy of resting blood flow. Data regarding constancy of resting blood flow in the present study are presented in Table 11. It is possible to compare these data against the study of Andres et al. (Table 12). It will be seen that there was greater constancy of flow in the present study. Constancy of flow in two typical subjects, in whom intermittent infusions of indicator were made, is illustrated in Figure 20. Here the coefficient of variation about mean flow is 1.9 per cent in the case of subject M.J. and 2.1 per cent in the case of subject W.C.W. There is no grossly apparent correlation in Table 11 between magnitude of blood flow and constancy of blood flow or between constancy of blood flow and mean relative difference.

Measurement of resting blood flow. A comparison is made in Table 13 between mean resting blood flows obtained in the present study

TABLE 10

MEAN RELATIVE DIFFERENCE BETWEEN INDICATOR CONCENTRATIONS IN TWO VEINS SIMULTANEOUSLY SAMPLED[a]

Mean Relative Difference %				<5	<10	<15	<20	<25
Cumulative Per Cent of Subjects	Standard Needle	Present Study	n = 10	10	33	40	50	60
		Andres et al.	n = 10	40	70	80	80	80
	Jet Needle	Present Study	n = 14	50	79	86	86	93
		Present Study[c]	n = 13	54	85	92	92	100
		Andres et al.[b]	n = 9	44	74	78	78	100

[a]After Andres et al. (2)

[b]Forearm only

[c]Excluding R.H.T.

TABLE 11

CONSTANCY OF RESTING BLOOD FLOW AND MEAN RELATIVE DIFFERENCES IN INDICATOR CONCENTRATION BETWEEN PAIRED SPECIMENS FROM BASILIC VEIN AND CEPHALIC VEIN DURING RESTING FLOW

Subject	Mean Calculated Blood Flow ± S.E. ml/100cc/min	Coeficient of variation, %	m.r.d.%, BV vs. CV
C.T.L.	6.7 ± 0.4	6.2	5.2
W.H.	6.7 ± 0.7	10.2	1.5
J.L.G.	5.7 ± 0.2	3.8	3.6
L.C.H.	5.2 ± 0.2	4.4	9.0
H.W.M.	7.3 ± 0.3	4.2	11.2
C.G.R.	5.6 ± 1.2	20.8	5.5
A.L.-2	3.0 ± 0.1	2.3	1.7
T.K.	10.4 ± 0.8	7.5	5.9
W.C.W.	5.0 ± 0.1	2.1	2.4
L.M.	9.7 ± 0.2	2.0	3.2
M.J.	9.4 ± 0.2	1.9	1.6
S.G.	26.7 ± 0.6	2.3	4.5
G.E.-2	13.1 ± 0.1	1.0	22.4
$\bar{x}$	8.8 ± 0.4	5.3	4.6

TABLE 12

CONSTANCY OF FLOW[a]

	Coefficient of Variation about Mean Flow			< 5	< 10	< 15	< 20	< 25
Cumulative Per Cent of Subjects	Jet Needle	Present Study	n = 13	69	85	92	92	100
		Andres et al.	n = 8	6	46	71	71	83
	Standard Needle	Andres et al.[b]	n = 9	11	78	78	89	89

[a]After Andres et al. (2)

[b]Forearm only.

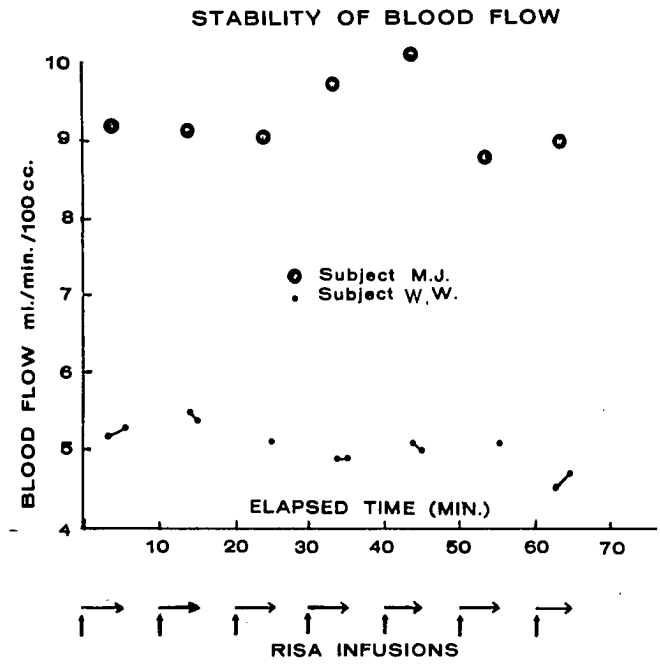

Fig. 20.-- Stability of blood flow

and those obtained by Andres *et al.* (2). Here cases in which calculated flows were of questionable accuracy were excluded from both studies. Higher values in the present study are probably due to the fact that the hand was included. The values in Table 13 may be compared to values de-

TABLE 13

COMPARISON OF CALCULATED BLOOD FLOW VALUES

Investigator	N	Ambient temperature, °C	Mean	Range
Andres *et al.* (2)[a]	7	25 - 27	4.7	2.7 - 7.0
Present Study[b]	12	25 - 27	7.3	3.0 -13.1

[a]Hand excluded

[b]Hand included

rived from plethysmography in Table 1. It is of interest that although ambient temperature was higher in the present study (26°C. as compared to 21°C.), the mean of resting blood flows is slightly lower than that obtained by plethysmography (forearm plus hand) using water bath temperatures believed to be optimal.

Infusion of indicator into the radial artery. In four subjects indicator was infused into both the radial artery and the brachial artery in turn. Results in these subjects are summarized in Table 14. It may be noted that, as expected, in all cases the relative difference between indicator concentration in the two veins downstream increased during infusion into the radial artery. In one case, C.G.R., no indicator became mixed with basilic venous blood during infusion into the radial artery, whereas mixing was apparently quite good (r.d. 5.5 per cent) during infusion into the brachial artery in the same subject. In the case of

TABLE 14

MIXING DURING RADIAL ARTERIAL INFUSION OF INDICATOR

Subject	Mean Relative Difference in Per Cent Between Indicator Concentrations in Basilic Vein and Cephalic Vein	
	Infusion into Radial Artery	Infusion into Brachial Artery
J.L.G.	28.0	3.2
A.L.-2	18.0	1.7
G.E.-2	53.1	22.4
C.G.R.	100.0	5.5
$\bar{x}$	49.8	8.2

A.L.-2, m.r.d. was less than twenty per cent during infusion into the radial artery, although very little arterial mixing could have occurred.

Total upper extremity resting vascular resistance in normotensive and hypertensive subjects. Table 15 compares total resting upper extremity vascular resistance in normotensive and hypertensive subjects. It will be seen that although flows are not significantly different in the two groups, there is a difference of borderline statistical significance in vascular resistance. It should be pointed out that the hypertensive group was composed mainly of patients with "mild" essential hypertension. However there were three patients, A.L., W.J.S., and W.E.S., with "moderate" or "severe" essential hypertension, and in two of these cases there was a highly elevated vascular resistance. Mean age of the hypertensive group was 49 years, normotensives 47 years. Mean upper extremity volume in hypertensives was 1689 cc., normotensives 1530 cc. Mean brachial arterial pressure in the hypertensives was 148 mm. Hg., normotensives, 97 mm. Hg.

Anomalous bifurcation of brachial artery. Figure 21 illustrates findings in subject R.H.T. Intermittent five minute infusions of indicator were made into the "brachial artery" and blood was sampled simultaneously in the radial artery, cephalic vein, basilic vein, and contralateral brachial artery. Isotope concentrations, indicated on the ordinate, increased with time in all vessels, representing increasing recirculating concentration. The concentrations in the two veins differed greatly, indicating lack of mixing. It will be noted that isotope concentrations in radial artery and cephalic vein were almost identical with isotope concentrations in contralateral brachial artery, representing recircula-

TABLE 15

TOTAL RESTING UPPER EXTREMITY VASCULAR RESISTANCE IN HYPERTENSIVE AND NORMOTENSIVE SUBJECTS

Hypertensives				Normotensives			
Subject	$\bar{P}_A$ mm Hg	Mean Flow ml/100cc/min	Mean Resist. mm Hg/ml 100cc/min	Subject	$\bar{P}_A$ mm Hg	Mean Flow ml/100cc/min	Mean Resist. mm Hg/ml 100cc/min
A.L.-2[a]	145	3.0	44.15	C.T.L.	100	6.7	13.58
T.K.	140	10.4	12.21	W.H.	85	6.7	12.24
W.J.S.[a]	165	9.0	17.22	J.L.G.	95	5.7	13.84
L.P.	135	7.4	17.03	L.C.H.	110	5.2	19.40
G.E.-1	140	6.9	18.24	H.W.M.	95	7.3	11.92
W.E.S.[a]	150	5.1	27.45	C.G.R.	105	5.6	16.96
L.C.B.	135	5.0	15.88	W.A.F.	80	5.0	14.80
F.F.	130	10.1	11.68	J.J.	105	8.5	10.71
H.B.	190	9.1	19.23	R.C.F.C.	100	5.8	17.26
				W.C.W.	100	5.4	17.22
				L.S.	90	4.0	20.50
				E.P.	105	4.4	20.68
				E.L.F.	90	14.3	5.80
				L.M.	95	8.5	9.88
				M.P.J.	100	9.3	9.78
$\bar{x} \pm$ S.E.	148	$7.3 \pm .8$[b]	20.34 ± 3.60[b]	$\bar{x} \pm$ S.E.	97	$6.8 \pm .7$[b]	14.30 ± 1.13[b]

[a]Moderate or severe hypertension.

[b]P (Flows): N.S. ($u_1 \neq u_2$); P (Resist.): 0.1 - 0.05 ($u_1 > u_2$).

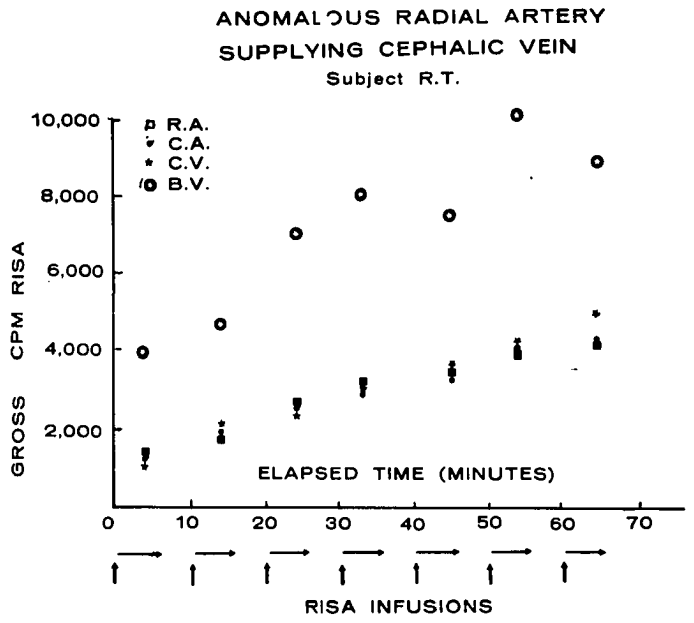

Fig. 21.-- Anomalous radial artery supplying cephalic vein

ting indicator. None of the locally infused isotope passed into the radial artery and cephalic vein and all passed into the basilic vein. Thus, the origin of this subject's radial artery was upstream to the point of indicator infusion, and the radial artery was the sole source of blood to the cephalic vein. Isotope had been infused into the ulnar rather than the brachial artery. Figure 22 illustrates findings in subject W.D., in whom most indicator passed into the cephalic vein, suggesting that in this case indicator was infused into the radial artery, rather than the brachial artery. However, radial arterial blood was not sampled, so this assumption cannot be proven.

Similar findings occurred in several other subjects and are presented in Table 7. The case of J.E.C. is particularly interesting for here no indicator was infused into the radial arterial blood and yet m.r.d. was not exceptionally high, indicating that a large amount of

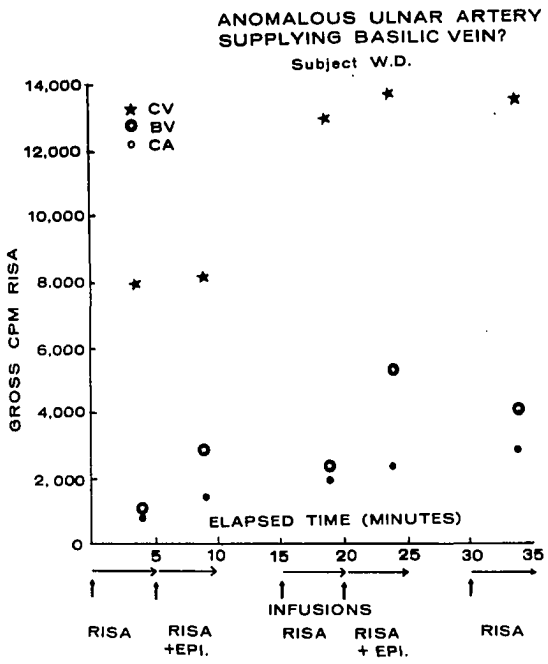

Fig. 22.-- Anomalous ulnar artery supplying basilic vein.

mixing took place in the capillary-venous bed of the limb. Findings in subjects O.H.P. and F.L.Z. suggest that, as in subject R.H.T., indicator may have been infused into the ulnar artery rather than the brachial artery.

Use of the technique to study upper extremity vascular responses to vasoactive agents. Arterial distribution of vasoactive agent during intra-brachial arterial infusions. In five subjects, indicator concentrations were measured in the ipsilateral brachial artery two centimeters downstream to the jet orifices through an arterial needle with double lumen. These concentrations were compared to those in the downstream venous samples drawn simultaneously. It was hoped that the concentrations would be similar, and that brachial arterial indicator concentration could be used to calculate flow. Table 16 presents data in these five subjects. It may be seen that indicator

TABLE 16

SIGNED RELATIVE DIFFERENCE IN PER CENT BETWEEN INDICATOR CONCENTRATION IN IPSILATERAL BRACHIAL ARTERY THREE CENTIMETERS DOWNSTREAM TO INJECTION ORIFICE AND MEAN OF PAIRED INDICATOR CONCENTRATIONS IN BASILIC AND CEPHALIC VEINS DURING RESTING FLOW

Subject	Signed r.d.% IA vs. $\frac{BV + CV}{2}$	r.d.%, BV vs. CV	Total Calc. Flow, ml/min
H.B.	+ 17.3	3.5	147.6
F.F.	+ 29.1	7.9	237.4
L.P.	+ 68.2	7.7	111.8
W.J.S.	+ 17.6	0.6	161.3
L.T.	- 7.6	8.2	150.4
Signed $\bar{x}$	+ 24.9		
$\bar{x}$	28.0	5.6	

concentration in the brachial arterial samples was quite different from that in venous blood. In four of the five subjects brachial arterial concentration was greater, in one it was less. There is no grossly apparent correlation between brachial arterial concentration and r.d. or total calculated flow.

In nine subjects indicator concentration was measured in downstream radial arterial blood and compared to mean indicator concentration in venous blood during resting flow and also during response to vasoactive infusions. Table 17 presents this data. On the average, during resting flow, concentration in the radial artery adjusted for recirculation concentration was five per cent lower than adjusted mean venous concentration. During infusion of the vasodilating agent, magnesium sulfate, adjusted concentration in the radial artery was 5.9 per cent higher on the average

TABLE 17

COMPARISON OF INDICATOR CONCENTRATION IN RADIAL ARTERIAL BLOOD WITH MEAN CONCENTRATION IN VENOUS BLOOD DURING RESTING FLOW AND DURING RESPONSE TO INFUSIONS OF VASOACTIVE AGENTS

Subject	Time After Beginning Infusion (min.)	I.A. Infusion of Isotonic NaCl			I.A. Infusion 10% $MgSO_4$ or Hypotonic NaCl			
		Signed Difference% RA vs. $\frac{BV+CV}{2}$	r.d.% BV vs. CV	Total Calculated Flow ml./min.	Agent	Signed Difference% RA vs. $\frac{BV+CV}{2}$	r.d.% BV vs. CV	Total Calculated Flow ml./min.
A.L.-2	5	+ 17.4	1.0	47.2		+ 5.3	16.7	84.5
	10	+ 15.1	1.9	47.6	$MgSO_4$	- 3.5	7.0	96.4
	15	+ 20.3	2.3	51.6		+ 32.4	1.0	104.0
	5	+ 16.7	0.1	42.5	Hypotonic	+ 12.9	0.2	32.5
	10	+ 11.1	1.4	35.9	NaCl	- 3.4	0.5	29.4
	15	+ 13.8	0.1	34.8		- 9.5	1.9	26.4
J.L.G.	5	- 36.2	2.9	76.9		+ 3.6	3.0	111.2
	10	- 34.5	3.4	75.4	$MgSO_4$	+ 18.6	4.0	110.4
	15	- 2.6	4.5	86.0		- 1.2	0.5	110.8
	5	+ 3.1	4.4	86.1	Hypotonic	- 5.5	0.2	63.6
	10	+ 12.0	4.7	83.0	NaCl	+ 0.2	5.4	54.0
	15	+ 23.5	4.1	82.0		+ 16.5	6.2	53.2

TABLE 17 - Continued

C.G.R.	5	- 0.4	5.3	49.2	$MgSO_4$	+ 43.7	19.4	144.0
	10	- 17.7	8.9	84.6		+ 40.1	17.1	143.6
	15	+ 0.3	2.3	106.9		..	..	..
	5	+ 16.8	10.9	79.2	Hypotonic NaCl	+ 36.5	4.5	64.7
	10	+ 12.2	5.7	75.9		+ 28.7	6.0	50.2
	15	+ 6.7	1.9	63.0		+ 27.7	16.4	48.8
T.K.	5	- 11.8	1.0	141.2	$MgSO_4$	- 34.0	17.2	252.4
	10	- 4.1	10.9	152.0		- 29.1	18.2	245.0
	15	- 1.9	5.8	181.8		+ 2.9	13.0	274.1
	5	- 18.6	18.6	218.6	Hypotonic NaCl	- 40.4	20.7	158.8
	10	- 12.0	20.7	189.1		- 46.3	20.1	149.8
	15	-176.4	13.5	188.0		- 46.0	17.9	141.4
W.O.D.	4	- 0.5	8.5	119.2	Hypotonic NaCl	- 2.1	4.7	72.7
	8	+ 4.2	4.9	111.4		- 37.4	5.2	58.8
	12	- 20.3	4.1	93.2		- 25.4	6.6	62.5
F.M.H.	5	..	..	..	$MgSO_4$	+ 3.9	5.7	116.0
	10	..	..	..		+ 5.6	4.0	119.1
	15	..	..	..		- 6.2	1.6	128.4

TABLE 17 - Continued

	5	- 10.4	3.2	113.8	Hypotonic	- 79.4	8.8	97.3
	10	- 27.1	3.8	110.5	NaCl	- 19.8	6.9	98.4
	15	- 21.6	5.6	108.6		- 45.1	2.6	93.8
C.T.L.	5	..	..	..		+ 9.1	4.3	112.0
	8	+ 8.6	2.4	123.3	Hypotonic	+ 2.4	5.2	151.4
	11	- 3.1	9.0	115.4	NaCl	+ 4.2	5.5	148.0
	14	+ 15.6	4.2	142.9		+ 8.3	11.6	118.6
	5	+ 7.0	7.2	144.9		..	..	..
	8	+ 12.3	4.6	143.6		..	..	..
	17	+ 3.1	10.8	129.2		..	..	..
Signed $\bar{x}$ (overall)		- 5.0				- 4.0		
$\bar{x}$ (overall)		17.2				20.5		
Signed $\bar{x}$ ($MgSO_4$)						+ 5.9		
$\bar{x}$ ($MgSO_4$)						16.4		
Signed $\bar{x}$ (Hypo.NaCl)						- 9.7		
$\bar{x}$ (Hypo.NaCl)						23.0		

TABLE 17 - Continued

Subject	Time After Beginning Infusion (min.)	I.A. Infusion of Isotonic NaCl		
		Signed Difference % RA vs. $\frac{BV + CV}{2}$	r.d. % BV vs. CV	Total Calculated Flow ml./min.
W.C.W.[a]	5	÷ 2.4	3.8	91.6
	4	- 0.9	5.1	93.9
	5	- 7.3	3.9	91.6
	5	+ 0.1	1.7	87.8
	4	- 6.8	3.3	81.9
	5	- 2.4	1.6	82.4
	4	- 9.3	1.3	85.2
	5	- 2.9	0.7	83.0
	5	- 8.3	1.1	84.4
	5	- 11.8	2.8	78.0

TABLE 17 - Continued

L.M.[a]	5	- 24.2	3.9	113.0
	5	- 33.2	5.8	119.2
	4 5	- 6.2 - 8.7	3.0 5.4	121.7 122.2
	4 5	+ 6.1 - 9.3	4.3 6.8	125.0 131.6
	4 5	+ 3.3 - 4.0	2.7 1.6	142.1 133.9
	4 5	- 1.0 - 16.5	1.0 1.5	136.4 124.8
	4 5	- 1.7 - 10.5	0.7 1.6	137.0 137.6
Signed $\bar{x}$		- 7.0		
$\bar{x}$		8.0		

[a] Seven separate consecutive five minute infusions of isotonic sodium chloride solution. Blood samples drawn at four and five minutes, or five minutes alone.

than adjusted mean concentration in the two veins. During infusion of hypotonic sodium chloride solution, which decreases blood flow, adjusted concentration in the radial artery was 9.7 per cent lower on the average than adjusted mean concentration in the two veins. However, the variation in the radial arterial concentrations was quite large, with deviations in both directions. It is of interest to note data from subjects L.M. and W.C.W., in whom a series of five minute infusions of indicator was made during resting flow and samples taken at four and five minutes. Although flow and r.d. were relatively constant, the concentration in the radial artery changed abruptly and greatly. In Table 17 the difference between indicator concentration in radial artery and veins may be compared to relative difference and to extremity blood flow. Correlation coefficient in the former case is 0.281, and in the latter case is 0.003. Correlation coefficient between relative difference and extremity blood flow is 0.0002.

Response to intrabrachial arterial infusion of angiotensin. Tables 18 and 19 present upper extremity blood flow response measured at four minutes to intrabrachial arterial infusions of angiotensin 0.025 and 0.05 μg. per minute, respectively. These observations were made in fourteen technically satisfactory procedures in eleven subjects. There is a significant decrease in blood flow at both dosage levels. In addition, there is a significant change in r.d. at both dosage levels. No gross correlation between total limb blood flow and r.d. is apparent. Figure 23 presents this data in a graphic form. It may be seen that calculated upper extremity blood flow decreased in all subjects, but that in some subjects, response to the larger dosage was less than that to the smaller

TABLE 18

UPPER EXTREMITY BLOOD FLOW RESPONSE AT FOUR MINUTES TO INTRABRACHIAL ARTERIAL INFUSION OF 0.025 μg./min. ANGIOTENSIN

Subject	Blood Flow (ml/100cc/min) During Infusion:		r.d. (per cent) During Infusion:	
	Isotonic NaCl	Angio-tensin	Isotonic NaCl	Angio-tensin
E.P.	2.7	2.2	2.7	2.8
J.J.	11.2	8.4	5.6	5.5
W.J.S.[a]	8.8	7.4	4.0	7.1
L.C.B.[a]	3.9	2.4	2.5	2.2
W.E.S.[a]	5.0	2.4	5.5	3.4
G.E.-1[a]	6.9	3.3	0.2	6.7
L.P.[a]	7.4	4.9	7.7	1.8
W.A.F.	4.5	2.9	1.0	3.0
$\bar{x}$	6.3	4.3	3.6	4.1
$\bar{d} \pm s_{\bar{d}}$	2.06 ± 0.35		2.51 ± 0.89	
P	$< .0005$[b]		.05 - .02[c]	

[a]Hypertensive subjects

[b]Alternate hypothesis $u_1 > u_2$

[c]Alternate hypothesis $u_1 \neq u_2$

TABLE 19

UPPER EXTREMITY BLOOD FLOW RESPONSE AT FOUR MINUTES TO INTRABRACHIAL ARTERIAL INFUSION OF 0.05 μ g/min. ANGIOTENSIN

Subject	Blood Flow (ml/100cc/min) During Infusion:		r.d. (per cent) During Infusion:	
	Isotonic NaCl	Angio-tensin	Isotonic NaCl	Angio-tensin
E.P.	2.5	2.2	3.2	2.1
J.J.	11.0	9.1	6.3	1.8
W.J.S.[a]	7.6	6.6	2.1	0.8
H.B.[a]	8.3	5.0	6.9	5.2
F.F.[a]	10.1	3.2	7.9	13.9
R.C.F.C.	6.8	2.6	7.1	0.4
$\bar{x}$	7.7	4.8	5.6	4.0
$\bar{d} \pm s_{\bar{d}}$	2.93 ± 0.98		3.55 ± 1.02	
P	.025 - .01[b]		.02 - .01[c]	

[a]Hypertensive subjects

[b]Alternate hypothesis $u_1 > u_2$

[c]Alternate hypothesis $u_1 \neq u_2$

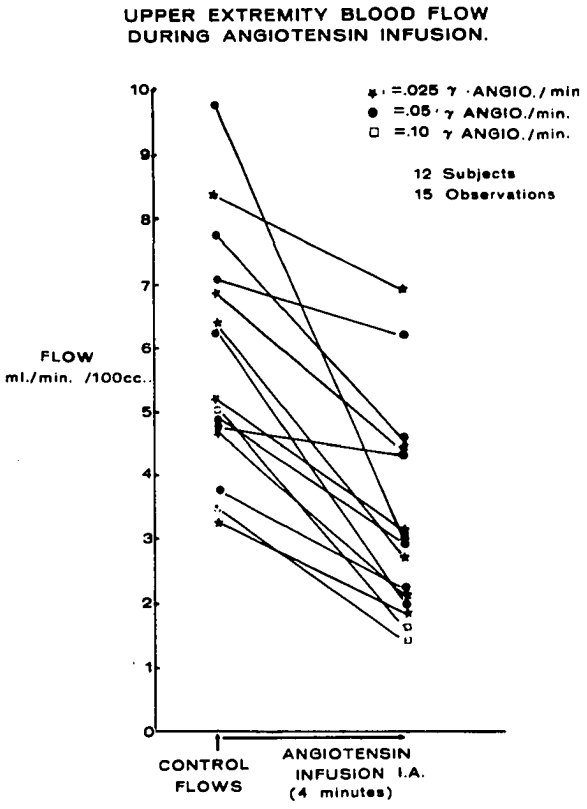

Fig. 23.-- Upper extremity blood flow during angiotensin infusion.

dosage. This finding is probably an artefact due to incomplete recovery from a previous angiotensin infusion.

Response to intrabrachial arterial infusion of epinephrine. Tables 20, 21, and 22 present upper extremity blood flow response measured at four minutes to intrabrachial arterial infusions of epinephrine 0.025, 0.05, or 0.10 μ g. per minute, respectively. These measurements were made in fifteen technically satisfactory procedures in ten subjects. No statistically significant change in upper extremity blood flow was produced by infusion of 0.025 and 0.05 μ g. per minute. Infusion of 0.10 μ g. per minute epinephrine produced a statistically sig-

TABLE 20

UPPER EXTREMITY BLOOD FLOW RESPONSE AT FOUR MINUTES TO INTRABRACHIAL ARTERIAL INFUSION OF 0.025 μg/min. EPINEPHRINE

Subject	Blood Flow (ml/100cc/min) During Infusion:		r.d. (per cent) During Infusion:	
	Isotonic NaCl	Epi-nephrine	Isotonic NaCl	Epi-nephrine
L.S.	4.0	3.3	4.6	19.8
E.L.F.	14.3	8.9	11.1	12.2
J.J.	8.5	10.5	5.7	2.9
W.A.F.	5.0	4.5	4.1	1.6
W.J.S.[a]	8.8	10.4	0.4	7.1
L.C.B.[a]	5.0	5.0	1.4	0.2
W.E.S.[a]	5.1	5.0	10.4	7.8
E.P.	4.4	4.7	1.0	13.6
$\bar{x}$	6.9	6.5	4.8	8.2
$\bar{d} \pm s_{\bar{d}}$	1.32 ± 0.63		5.59 ± 1.93	
P	.1 - .05[b]		.05 - .02[b]	

[a]Hypertensive subjects

[b]Alternate hypothesis $u_1 \neq u_2$

TABLE 21

UPPER EXTREMITY BLOOD FLOW RESPONSE AT FOUR MINUTES TO INTRABRACHIAL ARTERIAL INFUSION OF 0.05 μg/min. EPINEPHRINE

Subject	Blood Flow (ml/100cc/min) During Infusion:		r.d. (per cent) During Infusion:	
	Isotonic NaCl	Epi-nephrine	Isotonic NaCl	Epi-nephrine
W.A.F.	4.2	3.4	1.1	1.0
W.J.S.[a]	9.0	11.1	3.3	3.5
R.C.F.C.	5.8	9.8	6.7	3.2
$\bar{x}$	6.3	8.1	3.7	2.6
$\bar{d} \pm s_{\bar{d}}$	2.30 ± 0.93		1.27 ± 1.11	
P	.2 - .1[b]		0.4 - 0.3[b]	

[a]Hypertensive subject

[b]Alternate hypothesis $u_1 \neq u_2$

TABLE 22

UPPER EXTREMITY BLOOD FLOW RESPONSE AT FOUR MINUTES TO INTRABRACHIAL ARTERIAL INFUSION OF 0.10 μg/min. EPINEPHRINE

Subject	Blood Flow (ml/100cc/min) During Infusion:		r.d. (per cent) During Infusion:	
	Isotonic NaCl	Epi-nephrine	Isotonic NaCl	Epi-nephrine
L.S.	3.5	2.1	6.1	3.9
W.E.S.[a]	5.1	4.1	7.2	20.6
R.C.F.C.	6.4	9.0	8.0	4.4
H.B.[a]	9.1	10.3	0.8	5.4
$\bar{x}$	6.0	6.4	5.5	8.6
$\bar{d} \pm s_{\bar{d}}$	1.55 ± 0.36		5.95 ± 2.53	
P	.05 - .02[b]		0.1[b]	

[a]Hypertensive subjects

[b]Alternate hypothesis $u_1 \neq u_2$

nificant change in upper extremity blood flow. At all dosages, however, there was great variation in both magnitude and direction of response. Also only three observations were made at the 0.05 μ g. per minute level. Infusions at 0.025 μ g. per minute produced a statistically significant change in r.d., whereas, overall, no significant change in r.d. was produced by the two infusions of greater magnitude. However, again there was prominent variation among individual observations.

Response to intrabrachial arterial infusions of ten per cent magnesium sulfate and to hypotonic sodium chloride solution (150 mOs. per liter). Infusions of these agents were made with the specially constructed infusion pump. Figures 24 and 25 represent the experimental procedure in two subjects, J.L.G., and T.K. Brackets around points representing calculated flows signify r.d. and probably establish confidence limits for calculated flows. It will be noted that in subject J.L.G. limb resistance was in a steady state at time of beginning of infusion of magnesium sulfate, that limb resistance achieved a new steady state during infusion of magnesium sulfate, that the r.d.'s were small, and that limb resistance had returned to initial levels thirty minutes following the end of the magnesium sulfate infusion and before the beginning of the second vasoactive infusion. In contrast, in subject T.K., it will be noted that none of the above factors held. This combination of factors suggests that flow, and therefore resistance, calculations were accurate in J.L.G., but questionable in T.K. Other subjects fell between these two extremes.

Table 23 presents means of three or four measurements of upper extremity blood flow in eight subjects during fifteen minute intra-

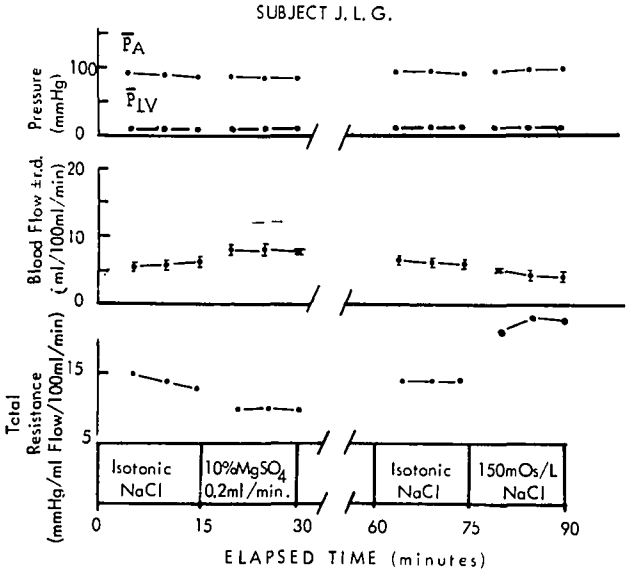

Fig. 24.-- Response to intrabrachial arterial infusions of ten per cent magnesium sulfate and to hypotonic sodium chloride solution in subject J.L.G.

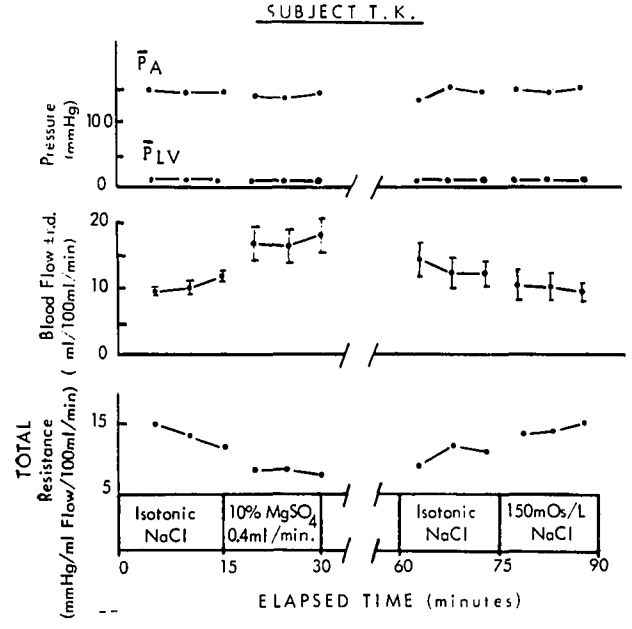

Fig. 25.-- Response to intrabrachial arterial infusions of ten per cent magnesium sulfate and to hypotonic sodium chloride solution in subject T.K.

TABLE 23

UPPER EXTREMITY VASCULAR RESPONSE DURING A FIFTEEN MINUTE INTRABRACHIAL ARTERIAL INFUSION OF TEN PER CENT MAGNESIUM SULPHATE[a]

Subject	Isotonic NaCl, 8 ml/min				10% $MgSO_4$ 0.2 ml + Isotonic NaCl 7.8 ml/min			
	Mean Calc. Blood Flow Total ml/min	ml/100cc/min	m.r.d. % BV vs. CV	Mean Total Resistance mm Hg/ml/100cc/min	Mean Calc. Blood Flow Total ml/min	ml/100cc/min	m.r.d. % BV vs. CV	Mean Total Resistance mm Hg/ml/100cc/min
W.H.	124.7	6.7	1.5	11.81	135.1	7.3	1.5	11.40
A.L.-2[b]	48.8	3.0	1.7	43.87	95.0	5.7	8.2	22.01
J.L.G.	79.4	5.7	3.6	13.58	110.8	7.9	2.5	9.62
H.W.M.	138.8	7.3	11.2	12.69	155.4	8.2	6.5	11.22
C.G.R.	80.2	5.6	5.5	18.51	143.8	10.1	18.2	9.63
C.T.L.[c]	137.2	7.2	6.8	15.87	128.8	6.8	5.1	16.72
L.C.H.[c]	92.8	5.2	9.0	18.79	143.8	8.0	11.0	12.77
T.K.[b,c]	158.3	10.4	5.9	13.04	257.2	16.9	16.1	7.71
$\bar{x}$	107.5	6.4	5.6	18.52	146.2	8.9	8.6(5.7[e])	12.64
P (Resistance):	0.025 - .01[d];		P(m.r.d.):		0.025 - 0.01[d]			

[a]Means of two to four observations during each fifteen minute infusion.
[b]Hypertensive subject
[c]0.28 to 0.40 ml. 10% $MgSO_4$ per minute
[d]Alternate hypothesis $u_1 > u_2$
[e]Mean of fifteen minute observations

brachial arterial infusions of ten per cent magnesium sulfate, 0.2, 0.28, or 0.4 ml. per minute, diluted in isotonic sodium chloride solution and infused at total volume rate of 8 ml. per minute. Decrease in mean total limb vascular resistance was observed in seven subjects during these infusions. This decrease is statistically significant. Figure 26 represents change in mean vascular resistance during these infusions. Figure 27 plots mean change in resistance against initial resistance and shows a trend toward positive correlation. Hypertensive subjects are identified by circled points on both figures.

With the exception of subject C.T.L., this infusion and its isotonic sodium chloride solution control infusion followed a pause of from five to thirty minutes after termination of the infusion of magnesium sulfate solution. Despite this time interval it is probable that limb vascular resistance had not completely returned to control levels in some of these subjects. However, increase in mean total limb vascular resistance was observed in 9 of 10 subjects during infusion of hypotonic sodium chloride solution, 150 mos. per liter, at 8 ml. per minute, as presented in Table 24. This increase is statistically significant. Figure 28 represents change in mean resistance during these infusions, and Figure 29 plots mean change in resistance against initial resistance. Hypertensive subjects are identified as above.

Infusion of hypotonic sodium chloride solution was accompanied by a significant increase in magnitude of m.r.d. Infusion of magnesium sulfate also increased the magnitude of m.r.d. during the first ten minutes of infusion. By the fifteenth minute, however, m.r.d. had returned to resting level.

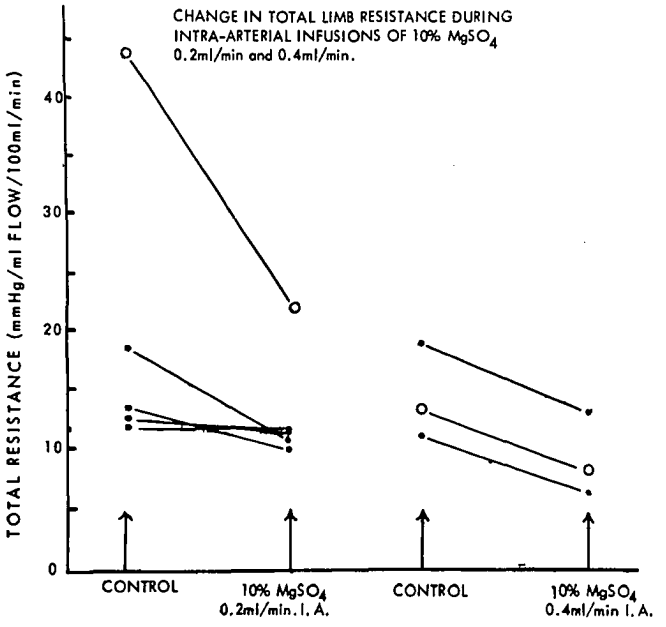

Fig. 26.-- Change in mean vascular resistance during infusions of magnesium sulfate.

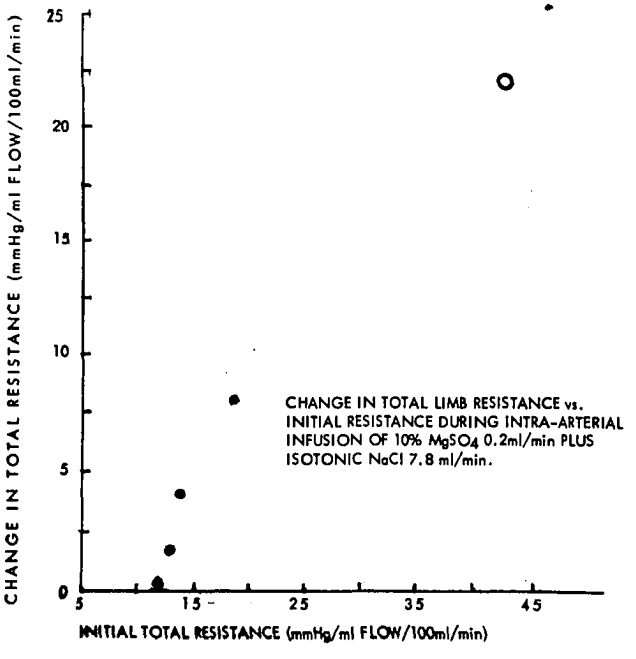

Fig. 27.-- Mean change in resistance vs. initial resistance during infusions of magnesium sulfate.

TABLE 24

UPPER EXTREMITY VASCULAR RESPONSE DURING A FIFTEEN MINUTE INTRABRACHIAL ARTERIAL INFUSION OF HYPOTONIC (150 mOs/L) SODIUM CHLORIDE SOLUTION[a]

Subject	Isotonic NaCl, 8 ml./min.				150 mOs/L NaCl, 8 ml./min.			
	Mean Calc. Blood Flow		m.r.d. % BV vs. CV	Mean Total Resistance mm Hg/ml/ 100cc/min	Mean Calc. Blood Flow		m.r.d. % BV vs. CV	Mean Total Resistance mm Hg/ml/ 100cc/min
	Total ml./min.	ml./100cc/ min.			Total ml./min.	ml./100cc/ min.		
W.H.	109.9	5.9	3.9	14.38	92.1	4.9	6.4	16.33
A.L.-2[b]	37.7	2.3	0.5	57.95	29.4	1.8	0.9	75.53
J.L.G.	83.7	6.0	4.4	13.76	56.9	4.1	3.9	21.88
H.W.M.	109.1	5.7	9.4	16.76	59.0	3.1	7.4	31.12
C.G.R.	72.7	5.1	6.2	19.82	54.6	3.8	9.0	26.50
C.T.L.	127.2	6.7	5.2	15.44	132.5	7.0	6.6	15.54
L.C.H.	121.4	6.7	2.3	15.07	84.9	4.7	10.4	22.79
W.O.D.	105.6	8.2	5.0	9.97	63.4	4.9	5.9	17.22
F.M.H.[b]	111.0	7.9	4.2	16.43	96.5	6.9	6.1	19.40
T.K.[b]	198.6	13.0	17.6	10.35	150.0	9.8	19.6	13.86
$\bar{x}$	107.7	6.8	5.9	18.99	81.9	5.1	7.6(8.8[d])	26.02

P(Resistance): 0.005 - 0.0005[c]; P (m.r.d.): 0.01 - 0.005[c]

[a]Means of three or four observations during each fifteen minute infusion.
[b]Hypertensive subject
[c]Alternate hypothesis $u_1 > u_2$
[d]Mean of fifteen minute observations

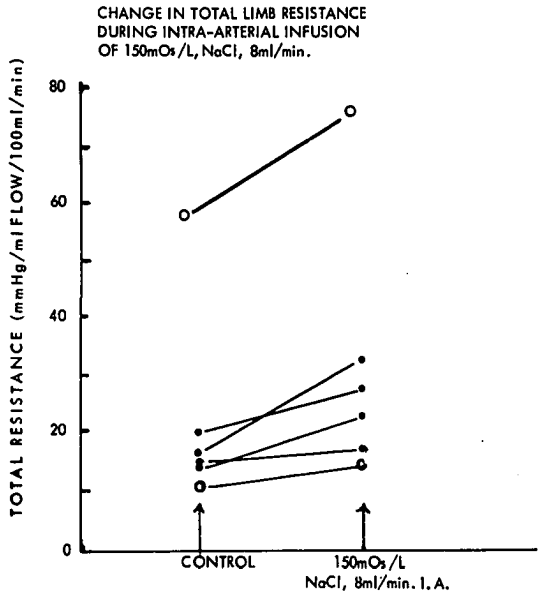

Fig. 28.-- Change in mean vascular resistance during infusions of hypotonic sodium chloride solution.

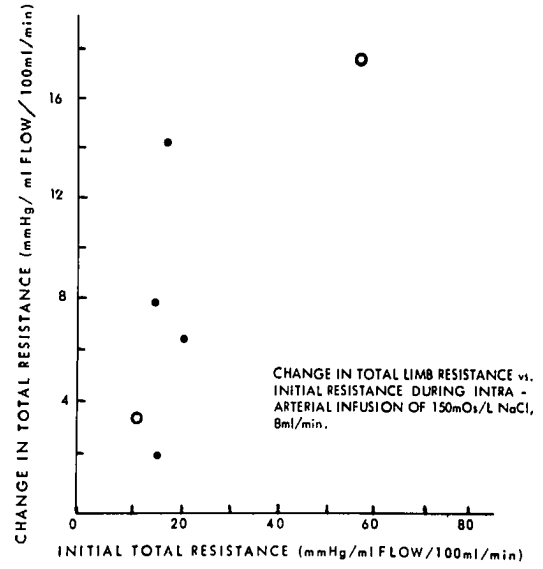

Fig. 29.-- Mean change in resistance vs. initial resistance during infusions of hypotonic sodium chloride solution.

CHAPTER IV

DISCUSSION

Validity of the Technique

There is good evidence indicating that the constant infusion indicator-dilution technique is able to measure accurately regional blood flow. Cropp and Burton (15) demonstrated a very high degree of correlation *in vitro* between actual and calculated pulsatile flow, using this technique. Accuracy, however, depends on complete mixing of indicator upstream to the sampling site. Mixing may be effected either by jet injection of indicator or by a "mixing region." Although the capillary-venous bed of the limb has dimensions quite different from those proposed by Cropp and Burton for a "mixing region," there is evidence indicating that it may function as such. This evidence includes the work of Baker and O'Brien (3) using bolus injections of indicator into the isolated forelimbs of dogs. Despite absence of any "mixing region" other than the capillary-venous bed, these authors demonstrated a high level of accuracy in calculated flow over a wide range of limb blood flows. Indeed these authors demonstrated that adding a mixer either at point of injection or at point of venous sampling did not improve accuracy. Thus complete arterial mixing is not essential for accurate calculation of blood flow *in vivo*, and satisfactory mixing must occur in the capillary and venous beds of the limbs.

The work of Andres *et al.* (2) also suggests that satisfactory mixing may occur in the capillary-venous beds of the limb despite poor arterial mixing. They demonstrated in man that m.r.d. was essentially the same whether indicator was infused via jet injector at rates calculated to produce turbulence and complete arterial mixing or via standard needle at rates too low to produce significant arterial mixing.

The present work further tests the validity of calculated flows *in vivo*, in the isolated forelimb of the dog. A high degree of accuracy was found in technically satisfactory procedures. However total venous outflow was sampled in the dog limb, whereas only a portion of venous outflow may be sampled in man. Similarly, total inflow was labeled in the dog limb, whereas undoubtedly a portion of the inflow to the upper extremity in man is not labeled. The experimental procedure could not duplicate the situation in man in these important aspects.

Mean and range resting blood flow values in the present study were slightly lower than those reported with venous occlusion plethysmography. This is of interest because most errors in the present technique, such as leakage or loss of indicator, would tend to cause high calculated flows. The comparison suggests, among other things, that during resting flow, blood viscosity, and therefore resistance, is not greatly lowered by the infusate in the present study. The reasonable agreement of values derived from these two techniques increases confidence in both methods of measurement of resting blood flow.

Theoretically the following conditions are necessary for accurate indicator-dilution measurement of blood flow: (a) steady state, (b) non-diffusible indicator, (c) supply and removal of indicator solely via

blood flow, (d) constant and equal arterial and venous blood flow, (e) constant concentration of indicator in arterial and mixed venous blood, and (f) uniform cross-sectional distribution of indicator upstream to sampling site. The findings of Andres et al. (2) and the present study regarding constancy of blood flow support the assumption that near steady state conditions exist during measurement of resting blood flow, and possibly after ten minutes during infusions of vasoactive agents. This is evidence supporting the first assumption. The data of Baker and O'Brien show that there is no trend for appreciable loss of albumin-bound indicator from the blood stream, supporting the validity of the second and third assumptions. The data of Cropp and Burton indicate that blood flow need not necessarily be constant for accurate calculations, especially if the constant infusion technique is used, satisfying the fifth assumption. Finally, the similarity in most subjects of indicator concentrations in paired venous blood samples is evidence supporting the final assumption.

Hemolysis and Mixing of Indicator

Hemolysis

Theoretically, complete brachial arterial mixing of indicator cannot be achieved without production of local turbulence. Andres et al. (2) calculated that infusion kinetic energies of at least 30,000 gm. cm.^{2}, sec.$^{-2}$ are required to exceed the Reynolds number of brachial arterial blood and produce local turbulence. Cropp and Burton (15) found in tubing and flow representative of the brachial artery that an infusion kinetic energy of 30,000 gm. cm.^{2}, sec.$^{-2}$ is necessary to achieve good cross-sectional mixing three centimeters downstream from injection orifice. Infu-

sion kinetic energies as low as 10,000 produce significant hemolysis, according to Andres et al. (2). Therefore in most cases local turbulence cannot be achieved in the brachial artery without production of significant hemolysis. Does it follow then that improved arterial mixing cannot be achieved without hemolysis? Can arterial mixing be significantly improved by jet infusions with kinetic energies below those producing turbulence? Andres et al. (2) discontinued use of the jet injector and did not investigate these questions. In the present study, there is good evidence in vitro and in vivo that r.d. and therefore arterial mixing are improved by the jet injector in the absence of hemolysis.

Thus it is possible to improve arterial mixing with infusions which do not produce turbulence, so the improved mixing must be attributed to another mechanism. From the photographs of mixing in glass tubing, it appears that indicator is mixed by being spread across the laminae of flow. In other words rather than converting laminar flow into turbulent flow, the injection system used in this present work appears to act by dispersing indicator into the various laminae of flow.

Mixing

Mixing over the entire vascular bed. For accurate calculation of blood flow there should be uniform cross-sectional distribution of indicator upstream to the sampling site. This mixing may occur on either (or both) arterial or venous sides of the limb vascular bed. Andres et al. (2) accepted as evidence that mixing had occurred similar indicator concentrations in blood from the two sampled veins. This was represented as r.d. of a low value. It is of interest to consider the significance of

the r.d. theoretically and in light of the findings of Andres et al. and the present work. A simplified approach would be to assume that the two veins sampled drain two distinct circulations, in which there is no mixing after bifurcation of brachial artery. Let us also assume that one circulation is solely supplied by the radial artery and the other solely by the ulnar artery. Given this situation, it may be calculated that the division of blood between the radial and ulnar arteries is a more important influence on accuracy of calculated flow than is the degree of mixing in the brachial artery, indicated by the magnitude of r.d. Indeed, if brachial arterial blood were evenly divided between the radial and ulnar arteries, flow would be accurately calculated even if there were no mixing prior to bifurcation and r.d. were as great as 100 per cent. If the division were forty and sixty per cent, flow would be calculated to within 10 per cent of actual flow if there were only 50 per cent mixing prior to bifurcation (r.d. $\leq$ 49 per cent). Similarly for divisions of 30%-70%, 20%-80%, 10%-90%, and 0%-100% flow would be accurately calculated to within 10% if there were at least 75%, 83%, 87%, and 90% mixing respectively. Corresponding r.d.'s would be $\leq$ 25%, 17%, 13%, and 10% respectively. The relationship is logarithmic and suggests that even in the human upper extremity, where the above assumptions do not hold, distribution of flow between the radial and ulnar arteries is probably as important a determinant of the accuracy of calculated flow as is the magnitude of r.d. It follows, then, that any vasoactive agent which changes the distribution of brachial arterial blood may change the accuracy of calculated flow, even if the value of r.d. remains constant. However, it seems unlikely that distribution of brachial arterial flow would

be rendered more disproportionate than 30%-70%. Therefore it is probable that if the value of r.d. remains equal to or less than 25 per cent, calculated flow lies within $\pm$ 10 per cent of actual flow.

The degree of mixing of indicator upstream to site of venous sampling is the major determinant of the magnitude of r.d. In this present project r.d. always increased in magnitude when indicator was infused into the radial artery rather than the brachial artery. In addition, in the present study, use of the jet injector resulted in decrease in the value of r.d. in both dogs and man over that value found using a standard needle. This result is in contrast to that of Andres et al.,who found no change during use of the jet injector. However, the veins sampled in the present study (cephalic and basilic) are different from those sampled in the study of Andres et al. (cephalic and brachial), and the study of Andres et al. excluded the hand, whereas the present study includes the hand. It is of interest to note that Andres et al., similar to the present study, found that their jet injector decreased the magnitude of r.d. if the hand was included in the circulation.

Andres et al. (2) concluded that an m.r.d. of twenty per cent or greater indicated that the indicator concentrations in the two sampled veins arose from different populations and discarded such data. However, their statistical analysis is questionable, and in Dog 1J, of the present study, r.d. exceeded twenty per cent and yet the calculated flows agreed well with the actual flows. Rather than rejecting calculated flows if the r.d. is above a certain arbitrary value, perhaps a more reasonable approach would be to consider the r.d. as an index of confidence limits for each calculated flow.

The magnitude of r.d. significantly changes during some vasoactive infusions. It might be expected that an increase in brachial arterial blood flow velocity caused by vasodilation would decrease time available for mixing of indicator with blood upstream to artery bifurcation. This should increase the magnitude of r.d. On the other hand vasoconstrictor agents should decrease r.d.by increasing mixing time. However, in the present study, hypotonic sodium chloride solution decreased blood flow, but the magnitude of m.r.d. increased. Epinephrine 0.025 μ g. per minute caused no significant change in blood flow but did significantly change the magnitude of r.d. Finally, in Table 17, there was no correlation between magnitude of blood flow and magnitude of r.d. In view of these data, changes in brachial arterial blood flow velocity within the limits observed probably do not significantly affect the magnitude of r.d.

Another variable which might affect the magnitude of r.d. during vasoactive infusions would be a temporary state of instability in venous indicator concentrations. If the two veins observed drained systems with greatly different mean transit times, the new steady state of indicator concentration would be achieved earlier in one vein than in the other. It is of interest that as noted in Table 23, if only the fifteen minute samples are considered during infusion of magnesium sulfate, there is no significant change in r.d. This finding would be evidence that in the case of magnesium sulfate infusion the change in r.d. may represent a temporarily instable state. The same finding is not present during the hypotonic infusion; however, it is possible that a steady state had not been reached even at fifteen minutes during this particular infusion.

Baltzan et al. (4) reported that a new steady state had usually been achieved after the tenth minute during an infusion of epinephrine, but this may not necessarily be true for other vasoactive agents. Indeed, the data in the present work for both magnesium sulfate and hypotonic sodium chloride solution suggest that in most subjects a really satisfactory steady state had not even been reached at fifteen minutes. However, infusions of vasoactive agents for periods longer than fifteen minutes is impractical for several reasons. First, spontaneous resistance changes over that period of time would tend to obscure changes due to the vasoactive agent. Second, systemic increments of the vasoactive agent might become significant during an infusion of longer duration.

Vasoactive agents may also change r.d. by in some way influencing the magnitude of mixing in the capillary-venous portions of the limb bed. This might be effected by opening or closing arterio-venous shunts or by diverting more or less blood into portions of the bed which have a greater number of venous anastomoses. There is no data in the present study which would support or deny this possibility.

Choice of the veins to be sampled is important. Andres et al. (2) sampled one vein draining the deep circulation and one the superficial, regardless of location of the veins in the forearm. In contrast, in the present study both veins sampled probably drain the superficial circulation, but they are located on opposite sides of the arm, the cephalic on the radial, and the basilic on the ulnar. The findings in subject R.H.T. and others that the radial artery solely or mainly supplies the cephalic vein suggests that the choice of veins in the present study was correct. However, it would be of interest to sample simultaneously

a third vein, draining the deep circulation.

Mixing on the arterial side of the bed. Data in subject A.L.-2 presented in Table 14 of the present study indicates that r.d. may be less than twenty per cent even if there is negligible mixing on the arterial side of the circulation. Thus the degree of mixing measured at the venous sampling sites is only a qualitative index of the amount of arterial mixing. Measurement of the degree of arterial mixing is necessary to the conclusion that vasoactive agent is uniformly distributed to the arteriolar bed. Sampling of brachial arterial blood is not helpful. Direct sampling of radial arterial blood is a better index of arterial mixing, but even here there are theoretical and practical problems. These may be divided into factors tending to cause flow calculated from radial arterial indicator concentrations to be erroneously high, factors tending to cause it to be "erroneously" low, and those factors which could cause errors in either direction. In the former category would be the "distance-distortion error" of Cropp and Burton (15) which could have a magnitude as great as 22.6 per cent. Also in this category would be errors caused by rapid withdrawal of radial arterial blood (increasing brachial arterial flow and decreasing indicator concentration). In the present experiments blood was rarely withdrawn from the radial artery at rates faster than four ml. per minute, but sometimes there was significant bleeding from the indwelling needle before the stylus could be reinserted. Although the total error would probably not increase brachial arterial flow by more than six or seven ml. per minute, this could decrease radial arterial indicator concentration by as much as thirty three per cent at low flow rates. One factor would tend to cause flow calcu-

lated from radial arterial indicator concentrations to appear erroneously low, failure to adjust recirculation concentration for time intercept. The equation proposed by Andres et al. (2) for calculation of blood flow subtracts an increment from recirculation concentration to take into account circulation time through the limb. Actually, however, Andres et al. neglected this correcting factor, and also in the present study this correction was not made. Baltzan et al. (4) point out that during resting flow mean forearm circulation time is approximately 2.3 minutes. Correction for this time increment would decrease calculated resting blood flow by two to six per cent. However, in order to correct recirculation concentration for this factor it would be necessary to measure steady state mean circulation time during each infusion. In the case of vasodilator infusions, where mean circulation time decreases, the correction factor would be smaller, but in cases of vasoconstrictor infusions, the correction factor could be large. Neglect of this factor tends to cause over-estimation of actual flow and might help to explain why some flows calculated on the basis of radial arterial concentrations of indicator appeared erroneously low, especially if total blood flow was low.

Factors tending to cause errors in either direction would include poor arterial mixing of indicator and failure to establish a steady state of venous indicator concentrations during vasoactive infusions. In view of these considerations, Table 25 presents means of measurements during resting flow from Table 17 and compares direction of significant difference between radial arterial and venous indicator concentrations with magnitude of r.d., and index of mixing, and with magnitude of total blood flow, an index of error due to failure to correct

TABLE 25

COMPARISON OF DIFFERENCE IN INDICATOR CONCENTRATION BETWEEN RADIAL ARTERY AND VEINS WITH TOTAL BLOOD FLOW AND WITH RELATIVE DIFFERENCE (r.d.)

	Total Blood Flow ml./min.	
	Indicator Concentration RA $> \frac{BV + CV}{2}$ by 5%	Indicator Concentration RA $< \frac{BV + CV}{2}$ by 5%
$\bar{x} \pm$ S.E.	82.5 ± 20.9	94.3 ± 29.1
P	$>0.5^a$	
	r.d. % BV vs. CV	
	Indicator Concentration RA $> \frac{BV + CV}{2}$ by 5%	Indicator Concentration RA $< \frac{BV + CV}{2}$ by 5%
$\bar{x} \pm$ S.E.	2.4 ± 0.5	4.1 ± 0.6
P	$>0.05^a$	

[a]Alternate hypothesis $u_1 \neq u_2$.

for time intercept. (The latter error should be greater at lower blood flows.) There is no significant correlation with either variable. In addition, as presented in Table 17, vasoconstriction was accompanied on the average by decreased radial arterial indicator concentrations (adjusted) relative to mean venous concentration (adjusted). This result is opposite to that to be expected from failure to adjust recirculation concentrations for time intercept. In conclusion, failure of radial arterial and mean venous indicator concentrations to be similar remains unexplained, and the evidence suggests that sampling of radial arterial blood gives only a qualitative indication of degree of arterial mixing of indicator. In addition, even in cases where there is a small difference between arterial and venous indicator concentrations such as subject W.C.W., Table 17, this may be caused by fortuitous balancing of factors tending to cause high or low radial arterial concentration. Perhaps, then, the best evidence that arterial mixing is improved by the present technique is presented in the photographs of indicator mixing in the glass tubing.

Measurement of Resting Upper Extremity Vascular Resistance in Hypertension

Observation that no increase in extremity vascular resistance is present in mild hypertension but that a significant increase is probably present in more severe hypertension confirms the findings of Brod *et al.* (10), using venous occlusion plethysmography.

Use of the Technique for Measurement of Responses to Vasoactive Agents

Use of this technique to determine vascular response to angiotensin confirms previous work indicating that this agent causes net

vasoconstriction. Data in the case of epinephrine are not clear cut and allow no conclusions. The data indicate that magnesium sulfate causes net dilation and hypotonic sodium chloride solution net increase in resistance of the upper extremity vascular bed in man. There is a trend toward positive correlation between magnitude of response to magnesium sulfate and initial vascular resistance level, as shown in Figure 28.

Errors in the Technique

The major disadvantage of the technique used to measure blood flow presented in this paper is its cumbersomeness. The technique requires three arterial and at least two venous punctures. A waiting period of at least thirty minutes following insertion of needles is required to establish a steady state. Each pair of vasoactive and control infusions requires a period of thirty minutes. A pause of at least thirty minutes is required after a vasoactive infusion, to reestablish resting flows. Taken together, study of the effects of two vasoactive infusions in one subject requires a period of from two and one-half to three hours. This is a long time for a subject to remain quietly on his back, confortable and cooperative.

Loss of Indicator

Because indicator infusions are made through a stopcock tubing arrangement at quite high pressures, there is always the possibility of indicator leakage and loss. In order to detect and prevent such loss, the system is closely observed during the procedure. Indicator may also be lost due to shunting around the elbow. Evidence for this factor has

been found in three subjects. For this reason the brachial arterial puncture is made as far distal in the forearm as practical. Finally, indicator may be lost if I^{131} is dissociated from albumin in the infusate. The manufacturer states that RIHSA dissociates less than one per cent before expiration date, so the magnitude of this loss is probably small.

Hemolysis

Complete occlusion of one orifice of the jet injector would cause the kinetic energy of infusion to rise to a level which might cause hemolysis. Occluding of the orifices is prevented by using a filter in series with the needle, by maintaining a flow of heparinized isotonic soldium chloride solution during pauses, and by monitoring infusion pressures (doubling of infusion pressure would indicate occlusion of one orifice and a four fold increase in kinetic energy of infusion). The patency of the needle is carefully checked before and after each procedure.

Viscosity Changes

Because the volume rate of the infusions is eight ml. per minute, infusate may make up as much as thirty three per cent of total forelimb blood flow during infusion of vasoconstrictor agents. This would result in a dilution of hematocrit by thirty three per cent and a fall in viscosity and resistance. In subjects with hematocrit within normal range, the change in resistance would be considerably less than thirty three per cent but still significant. In the case of agents which cause vasodilation, viscosity changes would probably not be sig-

nificant. It would be possible to correct calculated resistance changes for this dilutional effect, but instead a new jet injector needle with smaller orifices, which requires less than three ml. per minute infusion rate and delivers infusate of similar kinetic energies is being tested.

Adjustment of Recirculation Concentration for Time Intercept

(This error was discussed above.)

Establishment of Steady State Conditions

Baltzan et al. (4) point out that it is often necessary to wait as much as sixty minutes after infusion of epinephrine to reestablish control level of resistance. In the present study, resistance approximated control levels after a pause of thirty minutes in most but not all cases. Unfortunately, it is not known until after completion of the procedure, whether control levels of resistance have been reestablished. A monitoring system would be ideal to establish the correct pause, and such a system is being devised. However, it may be that infusion of more than one vasoactive solution per subject will be impractical for this reason.

Protocol for Study of Vasoconstrictor Agents

The above considerations suggest that experimental errors are of greater magnitude during vasoconstriction than during vasodilation. Infusion rates of vasoconstrictor agents which lower total extremity blood flow to less than forty ml. per minute would decrease hematocrit by more than twenty per cent and therefore should be avoided because of significant changes in blood viscosity. Mean transit time through the

extremity vascular bed would increase to approximately five minutes if vasoconstriction decreased extremity blood flow to one-half of its resting value. Approximately three mean transit times or fifteen minutes would be required to establish the new steady state of venous indicator concentration, and return to resting levels of vascular resistance would also be slow. In addition, the error caused by failure to adjust recirculation concentration for time increment could be as great as twenty per cent during vasoconstriction that reduced extremity blood flow to one-half its resting value.

In contrast, during vasodilation, viscosity changes caused by infusate would decrease. In addition, if flow doubled, the new steady state would be achieved in as little as three minutes and the error caused by failure to adjust recirculation concentration would decrease to one to three per cent. In view of these points, if two agents are to be studied in one subject, the vasodilator agent should be infused first. In addition vasoconstrictor agents should not reduce total flow to less than forty ml. per minute or one-half of resting value, and these infusions should be maintained for at least fifteen minutes.

CHAPTER V

SUMMARY AND CONCLUSIONS

The upper extremity vascular bed of man is easily accessible for study of vascular physiology and pathophysiology. Measurement of limb blood flow by a constant infusion indicator-dilution technique similar to that suggested by Andres *et al.* (2), provides an advantage over other techniques: During study of regional responses to arterial infusion of vasoactive agents, evidence of mixing of indicator suggests that vasoactive agent is also uniformly distributed throughout the limb vascular bed. The present work explores the concept that an injection system for brachial arterial infusions may be developed, which satisfactorily mixes both indicator and vasoactive substances with brachial arterial blood and does not create hemolysis.

A new jet injector needle was developed and tested *in vitro* in glass tubing and *in vivo* in the limbs of dogs and men. Constant infusions of indicator and vasoactive agents were made into the brachial artery, and cephalic and basilic venous blood was repeatedly sampled downstream for measurement of indicator concentrations. The degree of mixing was estimated by comparing concentrations of indicator in the paired venous samples. The mean venous concentration adjusted for recirculation concentration was used in calculations of flow.

In vivo investigations in dog and man indicated that the dif-

ferences between indicator concentrations in the paired venous samples were reduced by the jet injector over values obtained using a standard needle injector. In ten dogs, most calculated flows lay within $\pm$ 10 per cent of actual flows over a wide range of actual flows. Jet infusion did not change limb vascular resistance in seven dogs, indicating that no significant hemolysis was produced. In man, intra-brachial arterial infusion of angiotensin, magnesium sulfate solution, and hypotonic sodium chloride solution caused calculated limb blood flow to decrease, increase, and decrease, respectively. Fifty subjects were studied without significant morbidity.

Both in vitro and in vivo studies suggest that arterial mixing is improved by the jet injector in the absence of hemolysis. Satisfactory mixing of vasoactive agents at the arteriolar level is of great importance in study of vasular responses. The mechanism of improved mixing is probably improved dispersion of infusate in the brachial arterial blood rather than creation of local turbulent flow. Comparison of actual with calculated flows indicates that calculated flows are quite accurate. In addition, the mean and range of resting flow measurements agree well with those found by others using venous occlusion plethysmography. New findings include evidence that the cephalic and basilic veins drain beds primarily perfused by radial and ulnar arterial blood, respectively. The data indicate that magnesium sulfate and hypotonic sodium chloride solution cause net dilation and increased vascular resistance, respectively, in the upper extremity vascular bed of man. It is concluded that this technique is of value in study of the peripheral circulation in man.

REFERENCES CITED

1. Agrifoglio, G., Thorburn, G.D. and Edwards, E.A. "Measurement of Blood Flow in Human Lower Extremity by Indicator-Dilution Method," Surgery, Gynecology, and Obstetrics, 113:641, 1961.

2. Andres, R., Zierler, K.L., Anderson, H.M., Stainsby, W.N., Cader, G., Grayyib, A.S. and Lilienthal, J.L., Jr. "Measurement of Blood Flow and Volume in the Forearm of Man; With Notes on the Theory of Indicator-Dilution and on Production of Turbulance, Hemolysis, and Vasodilatation by Intra-vascular Injection," Journal of Clinical Investigation, 33:482, 1954.

3. Baker, C.H. and O'Brien, L.J. "Vascular Volume Changes in the Dog Forelimb," American Journal of Physiology, 206:1291, 1964,

4. Baltzan, M.A., Andres, R., Cader, G. and Zierler, K.L. "Effects of Epinephrine on Forearm Blood Flow and Metabolism in Man," Journal of Clinical Investigation, 44:80, 1965.

5. Barcroft, H. and Edholm, O.G. "The Effect of Temperature on Blood Flow and Deep Temperature in the Human Forearm," Journal of Physiology, 102:5, 1943.

6. Barcroft, H. and Edholm, O.G. "Temperature and Blood Flow in the Human Forearm," Journal of Physiology, 104:366, 1946.

7. Barnett, A.J. "Venous Occlusion Plethysmography: A Critical Study," Medical Journal of Australia, 2:818, 1950.

8. Bauer, F., Cassen, B., Youtcheff, E. and Shoop, L. "Jet Injection of Radioisotopes," American Journal of Medical Sciences, 225:374, 1953.

9. Bell, G. and Harper, A.M. "Measurement of Regional Blood Flow Through the Skin From the Clearance of Krypton-85," Nature, 202:704, 1964.

10. Brod, J., Fencl, V., Hejl, Z. and Jirka, J. "General and Regional Haemodynamic Pattern Underlying Essential Hypertension," Clinical Science, 23:339, 1962.

11. Brodie, T.G. and Russell, A.E. "On the Determination of the Rate of Blood Flow Through an Organ," The Journal of Physiology, 32:47, 1905.

12. Catchpole, B.N. and Jepson, R.P. "Hand and Finger Blood Flow," Clinical Science, 14:109, 1955.

13. Coles, D.R., Cooper, K.E., Mottram, R.F. and Occleshaw, J.V. "The Source of Blood Samples Withdrawn from Deep Forearm Veins via Catheters Passed Upstream from the Median Cubital Vein," Journal of Physiology, 142:323, 1958.

14. Conrad, M.C. and Green, H.D. "Evaluation of Venous Occlusion Plethysmograph," Journal of Applied Physiology, 16:289, 1961.

15. Cropp, G.J.A. and Burton, A.C. "Theoretical Considerations and Model Experiments on the Validity of Indicator Dilution Methods for Measurements of Variable Flow," Circulation Research, 18:26, 1966.

16. Dobson, E.L. and Warner, G.F. "Measurement of Regional Sodium Turnover Rates and Their Application to the Estimation of Regional Blood Flow," American Journal of Physiology, 189:269, 1957.

17. Folse, R. "Application of the Sudden Injection Dye Dilution Principle to the Study of the Femoral Circulation," Surgery, Gynecology, and Obstetrics, 120:1194, 1965.

18. Formel, P.F. and Doyle, J.T. "Rationale of Venous Occlusion Plethysmography," Circulation Research, 5:354, 1957.

19. Froněk, A. and Ganz, V. "Measurement of Flow in Single Blood Vessels Including Cardiac Output by Local Thermodilution," Circulation Research, 8:775, 1960.

20. Ganz, V., Hlavová, A., Froněk, A., Linhart, J. and Přerovský, I. "Measurement of Blood Flow in the Femoral Artery in Man at Rest and During Exercise by Local Thermodilution." Circulation, 30:86, 1964.

21. Gibbs, F.A. "Thermoelectric Blood Flow Recorder in the Form of a Needle," Proceedings of the Society for Experimental Biology and Medicine, 31:141, 1933.

22. Greenfield, D.M. "Venous Occlusion Plethysmography," In: Methods in Medical Research. Chicago, Yearbook Publishers, Inc., 8:293, 1960.

23. Greenfield, D.M. "Peripheral Blood Flow by Calorimetry," In: Methods in Medical Research. Chicago, Yearbook Publishers, Inc. 8:302, 1960.

24. Hamilton, W.F. and Remington, J.W. "Comparison of the Time Concentration Curves in Arterial Blood of Diffusible and Non-diffusible Substances when Injected at a Constant Rate and When Injected Instantaneously," American Journal of Physiology, 35:148, 1947.

25. Hensel, H. and Ruef, J. "Fortlaufende Registrierung der Muskeldurchblutung am Menschen mit Einer Calorimetersonde," Pflüger's Archiv Für Die Gesamte Physiologie, 259:267, 1954.

26. Hensel, H. and Bender, F. "Fortlaufende Bestimmung der Hautdurchblutung am Menschen mit Einem Elektrischen Wärmeleitmesser," Pflüger's Archiv Für Die Gesamte Physiologie, 263:603, 1956.

27. Hewlett, A.W. and van Zwaluwenburg, J.C. "The Rate of Blood Flow in the Arm," Heart, 1:87, 1909.

28. Hobbs, J.T., Agrifoglio, G. and Edwards, E.A. "Measurement of Femoral Artery Blood Flow Using the Indicator-Dilution Method," Journal of Surgical Research, 2:386, 1962.

29. Holling, H.E. "Observations on the Oxygen Content of Venous Blood from the Arm Vein and on the Oxygen Consumption of Resting Human Muscle," Clinical Science, 4:103, 1939.

30. Hyman, C. "Peripheral Blood Flow Measurements: Tissue Clearance," In: Methods in Medical Research. Chicago, Year Book Publishers, Inc., 8:236, 1960.

31. Kety, S.S. "Measurement of Regional Circulation by the Local Clearance of Radioactive Sodium," American Heart Journal, 38:321, 1949.

32. Kety, S.S. In: Methods in Medical Research. Chicago, Year Book Publishers, Inc., 8:248, 1960.

33. Landowne, M. and Katz, L.N. "A Critique of the Plethysmographic Method of Measuring Blood Flow in the Extremities of Man," American Heart Journal, 23:644, 1942.

34. Lassen, N.A., Lindbjerg, I.F. and Munck, O. "Measurement of Blood-Flow Through Skeletal Muscle by Intramuscular Injection of Xenon-133," The Lancet, 1:686, 1964.

35. Rapaport, S.I., Saul, A., Hyman, C. and Morton, M.E. "Tissue Clearance as a Measure of Nutritive Blood Flow and the Effect of Lumbar Sympathetic Block upon Such Measures in Calf Muscle," Circulation, 5:594, 1952.

36. Roddie, I.C., Shepherd, J.T. and Whelan, R.F. "Evidence from Venous Oxygen Saturation Measurements That the Increase in Forearm Blood Flow During Body Heating is Confined to the Skin," Journal of Physiology, 134:444, 1956.

37. Rush, B.F., Horie, N. and Klein, N.W. "Intra-arterial Infusion of the Head and Neck, Anatomic and Distributional Problems," American Journal of Surgery, 110:510, 1965.

38. Shepherd, R.C. and Warren, E. "Studies of the Blood Flow Through the Lower Limb of Man by the Nitrous Oxide Technique," Clinical Science, 20:99, 1960.

39. Shepherd, J.T. Physiology of the Circulation in Human Limbs in Health and Disease. W.B. Saunders Co., Philadelphia, Pa., 1963, p. 319.

40. Slaughter, O.L., Brown, H.S. and Wakim, K.G. "Effects of Tetraethyl-ammonium Chloride on Blood Flow in the Extremities of Man," Journal of Laboratory and Clinical Medicine, 33:743, 1948.

41. Stewart, G.N. "Researches on the Circulation Time and on the Influences Which Affect it. IV. The Output of the Heart," Journal of Physiology, 22:159, 1897.

42. Stow, R.W., and Schieve, J.F. "Measurement of Blood Flow in Minute Volumes of Specific Tissues in Man," Journal of Applied Physiology, 14:215, 1959.

43. Visscher, M.B., and Johnson, J.A. "The Fick Principle: Analysis of Potential Errors in its Conventional Application," Journal of Applied Physiology, 5:635, 1953.

44. Wahren, J. "A Dye Dilution Method for the Determination of Brachial Artery Blood Flow During Forearm Exercise in Man," Acta Physiologica Scandinavica, 64:477, 1965.