Hypertonic saline method accurately determines parallel conductance for dual-field conductance catheter

Paul Steendijk, Eva Staal, J. Wouter Jukema, and Jan Baan. Hypertonic saline method accurately determines parallel conductance for dual-field conductance catheter. Am J Physiol Heart Circ Physiol 281: H755–H763, 2001.—Conversion of conductance catheter data to absolute ventricular volumes requires assessment of parallel conductance ($G_P$). We determined the accuracy of $G_P$ obtained by the hypertonic saline method ($G_{P\text{saline}}$) compared with angiographically derived $G_P$ ($G_{P\text{Angio}}$) and quantified the variabilities of $G_P$ for the dual-field conductance catheter method in nine anesthetized sheep studied at baseline, treated with dobutamine, and subjected to volume loading and β-blockade. $G_{P\text{saline}}$ and $G_{P\text{Angio}}$ showed an excellent linear correlation ($G_P = 1.002G_{P\text{Angio}} + 0.001\ \Omega^{-1},\ R^2 = 0.92$), and Bland-Altman analysis yielded a nonsignificant bias and narrow limits of agreement (bias ± 2SD = 0.002 ± 0.112 Ω⁻¹). Within-animal variability of $G_P$ was very similar with both methods and was due to changes in blood conductivity rather than geometrical changes. Variability between animals was significant (26.3% of mean for $G_{P\text{saline}}$ and 25.7% for $G_{P\text{Angio}}$) and thus warrants individual assessment. Variations during the cardiac cycle were not significantly different from zero. With biplane angiography used as gold standard, the hypertonic saline method accurately determines $G_P$ for the dual-field conductance catheter over a wide range of hemodynamic conditions.

The conductance catheter method provides a continuous on-line measurement of left ventricular (LV) volume by means of a multielectrode catheter positioned in the LV. In combination with simultaneous measurement of LV pressure through a sensor on the same catheter, this instrument enables quantification of ventricular function by means of pressure-volume relations. Such relations have proven to be particularly useful, because they provide indexes of systolic and diastolic ventricular function that are relatively independent of loading conditions and, as such, mainly reflect intrinsic myocardial properties.

The conductance catheter method is based on the continuous measurement of the electrical conductance of the blood in the LV. This signal is converted to a volume signal on the basis of a stacked-cylinder model and by taking into account the specific conductivity of blood and the catheter electrode spacing. However, the conductive tissues and fluids surrounding the LV cavity [e.g., myocardial wall, blood in the right ventricle (RV), lung] also contribute to the measured conductances and introduce an offset in the relation between true LV volume and conductance-derived volume. Therefore, to obtain an absolute volume signal, this parallel conductance needs to be determined and subtracted from the raw conductance signal.

To assess parallel conductance or the corresponding offset volume, several methods have been used. These methods are based on a direct comparison with an independent method for absolute volume measurement (5, 6, 20) or do not require an independent volumetric measurement, such as the dual-frequency method (12), the suction method (3), or the volume reduction technique (31). However, in most studies, parallel conductance is determined by a method, previously developed in our laboratory, that consists of injecting a small bolus of hypertonic saline through a balloon-flotation catheter in the pulmonary artery (3). The highly conductive saline transiently changes the conductivity of the blood, practically without affecting parallel conductance. The contribution of parallel conductance to the total conductance signal can be derived from a registration of the conductance signal during the passage of the bolus through the LV. A number of studies indicate that the saline method provides reliable estimates of parallel conductance, but generally the evidence is indirect, since these studies were not specifically designed to test the accuracy of the saline method (1, 7, 20, 26, 28, 29). Some studies were more specifically related to the calibration factors of the conductance catheter method, but those studies largely focused on whether parallel conductance remains constant during preload reduction by caval occlusion (2, 5, 27). The present study, in anesthetized sheep, was designed specifically to test the accuracy of assessment of parallel conductance by the hypertonic saline method and used biplane contrast cineangiography as an independent method for absolute volume measurements. In addition, in the present study we used the dual-field excitation mode to generate the electric field required for the conductance measurements. Dual-field excita-
tion, developed by our group, is used in most studies, because it has been shown to improve the accuracy of the conductance catheter method (10, 24, 25, 35). However, most studies regarding parallel conductance have been performed before the introduction of the dual-field excitation method. The more uniform electric field, which is the rationale for using dual-field excitation, improves the linearity of the relation between conductance and true volume (25), and thus, at least theoretically, the possible dependence of parallel conductance on volume could be expected to be reduced. The more uniform electric field implies more uniform intracavitary current density, but the “wider” field is expected to lead to more current flow through external structures and, thus, higher parallel conductance. A possible advantage, however, is that volume dependence and variability of parallel conductance between hemodynamic conditions may be reduced. To investigate the need for repeated assessments, we determined the variability of parallel conductance between animals and between hemodynamic conditions. Measurements were obtained over a wide range of hemodynamic conditions, induced by dobutamine infusion, volume loading, and β-blockade. Finally, it is generally (implicitly) assumed that parallel conductance remains constant during the cardiac cycle. This assumption has been tested by analyzing hypertonic saline injections for individual points in the cardiac cycle (19, 33, 34); however, this analysis may be theoretically incorrect (see DISCUSSION). Therefore, we directly compared the time-varying angiographic and conductance-derived volume signals. After subtraction of a (constant) parallel conductance factor, the remaining difference between the two signals can be interpreted as (time-dependent) variations in parallel conductance during the cardiac cycle.

METHODS

Animals

The study was approved by the Animal Research Committee of the University of Leiden. The investigation conforms with the Guide for the Care and Use of Laboratory Animals (National Institutes of Health Publication No. 85-23, Revised 1996). Nine sheep (body mass 38.6 ± 2.9 kg, age 3–6 mo) were premedicated with ketamine (40 mg/kg im) and atropine (0.05 mg/kg im). The animals were intubated and ventilated with a Servo 900B (Siemens, Elema, Sweden). On ventilation, atracurium (0.25 mg/kg iv) was administered to achieve adequate muscle relaxation. General anesthesia was maintained with isoflurane (1%) and continuous infusion of ketamine (4–10 mg·kg⁻¹·h⁻¹ iv). Arterial PO₂ and PCO₂ and pH were checked every 30 min and kept within normal ranges by adjusting the fraction of inspiratory O₂ and tidal volume as necessary.

Instrumentation

An electrocardiogram was obtained using subcutaneous needle electrodes. To facilitate catheter placement, sheaths were placed in the left and right femoral artery, the left and right femoral vein, the left carotid artery, and the left jugular vein. A balloon-floation catheter was positioned in the pul-

Conductance Catheter

The conductance catheter technique has been described in detail previously (3, 25). Briefly, a catheter with 10 or 12 electrodes is positioned along the longitudinal axis of the LV. The electrode distance is chosen such that, with electrode 1 within the apex, electrode 9 is situated just above the aortic valve. Through the two most proximal and the two most distal electrodes, two 20-kHz currents (current ratio 1:0.25) opposite in polarity are applied, creating a dual electric field in the ventricular cavity (24, 25). The interspaced electrodes are used to measure the conductances of five intraventricular segments. Total time-varying LV conductance (G(t)) is calculated as the sum of these five segmental conductances. Time-varying LV volume is calculated as follows: V_{cath}(t) = (1/α)(L^2/σ_0)[G(t) - G_p^p], where α is a dimensionless slope factor (see below), σ_0 is the specific conductivity of the blood measured from a blood sample using a special cuvette, L is the catheter electrode spacing, and G_p^p is the parallel conductance (see below).

Nomenclature. In the literature, the term parallel conductance is often used loosely for the “physical” parallel conductance (G_p^p) and for the correction volume (V_c). In the present study, the results are described mainly in terms of G_p^p and the relation between absolute “true” volume and conductance (G(t)) is written as follows: V(t) = (1/α)(L^2/σ_0)[G(t) - G_p^p]. Originally, Baan et al. (3) used the following equation: V(t) = (1/α)(L^2/σ_0)G(t) - V_C; other groups have used the following equation: V(t) = (1/α)(L^2/σ_0)G(t) - V^p. Obviously, V_C and V^p can be written in terms of G_p^p, but since α and σ_0 may vary, these terms cannot be used interchangeably. To avoid confusion, we will use the nomenclature as follows: G(t) for conductance, G_p^p for parallel conductance, (L^2/σ_0)G(t) for conductance volume, V^p = (L^2/σ_0)G_p^p for parallel conductance volume, and V_C = (1/α)(L^2/σ_0)G_p^p for correction volume.

The Leycom Sigma-5 DF signal processor requires the user to dial in a value for L and for blood resistivity (ρ = 1/σ_0), and the analog output of the system equals (L^2/σ_0)G(t), rather than the raw conductance [G(t)].

Slope factor. After correction for G_p^p, the signal obtained by the conductance catheter is directly proportional to absolute ventricular volume but generally underestimates true volume by a fixed percentage. To correct the underestimation, α...
was introduced. In practice, $\alpha$ is determined by comparing
the conductance-derived volume (or stroke volume (SV)) with
an independent measurement such as angiography or ther-
modilation. In animals such as dogs or sheep, $\alpha$ is typically
0.8 (3, 24). A key point in the present study is that the
approaches are used to determine $G^P$, i.e., the saline
dilution method and the angiographic method (see below),
do not require that $\alpha$ be assessed and are independent of
the actual value of $\alpha$. Both methods analyze the raw conductance
signals, rather than the calibrated conductance volume sig-
als, to determine $G^P$. The only implicit assumption is that $\alpha$
can be regarded as constant during the cardiac cycle, but it
does not necessarily need to be 1.0. This assumption is
validated by testing the linearity of the relation between
conductance and angiographic volume.

$G^P$ Obtained by Hypertonic Saline Injection

The electric field generated by the conductance catheter is
not entirely restricted to the ventricular blood volume, but
current also passes through the ventricular wall, other car-
diac chambers, and, to some extent, through all electrically
conducting structures surrounding the heart. As a conse-
quence, the total conductance measured is the sum of the
conductance of the blood in the LV and the “parallel” conduc-
tance of the surrounding structures. Baan et al. (3) devised a
method to determine $G^P$ by injecting a small bolus (2–3 ml) of
hypertonic (10%) saline through a balloon-flotation catheter
in the pulmonary artery. This procedure can be explained as
follows. If blood conductivity in the LV could be reduced to 0,
the measured total conductance would represent $G^P$ only.
In practice, this is not possible, but we can transiently change
conductivity (by the hypertonic saline injection), plot mea-
sured total conductance vs. blood conductivity, and extrapo-
late these data to the point where conductivity hypotheti-
ically would be 0 and obtain $G^P$ in this way. This approach (see APPENDIX) requires a beat-to-beat estimate of blood conduc-
tivity in the LV, which can be obtained as follows: $\sigma_N = \frac{1}{\rho_0}(G_{SV}/SV)\times SG$, where $SV = V_{ED} - V_{ES}$, and “stroke conductance”
(SG) = $G_{ED} - G_{ES}$, where the subscripts ED and ES repre-
sent end diastole and end systole, respectively. The equation
for $\sigma_N$ shows that if hemodynamics (and thus SV) are con-
stant during the passage of the bolus, $\sigma_N$ is directly propor-
tional to the amplitude of the conductance signal (SG). Thus
$G^P$ can be obtained by plotting $G_{ED}$ vs. SG for each beat
during the change in blood conductivity and extrapolating
this relation to SG = 0. This point corresponds to the hypo-
thetical situation with $\sigma_N = 0$ and, therefore, yields $G^P$.

In the present study, $G^P$ obtained by hypertonic saline injection ($G^P_{saline}$) was determined as the mean of three repeat
hypertonic saline injections analyzed by custom-made soft-
ware.

Angiography

Angiography was performed with a Philips DCI SX biplane
X-ray system with a biplane frame rate of 25 frames/s and
7-in. image intensifiers. Simultaneous biplane images from
standard 30° right anterior oblique (RAO) and 60° left ante-
rior oblique (LAO) projections were obtained after injection of
15 ml of nonionic contrast material (Iomeron, Bracco-Byk
Gulden, Konstanz, Germany) at a flow rate of 6 ml/s. All data
were acquired after the respirator was disconnected at end
expiration. Images were stored on CD ROM at the end of the
study for off-line analysis.

Angiographic dimensions were calibrated on the diameter
of the angiographic catheter using QCA-CMS View software
(Medis, Leiden, The Netherlands). Angiographic LV volumes
($V_{Angio}$) were calculated using the area-length method (9, 36)
as follows: $V_{Angio} = \left(\frac{2}{3}\pi L_{RAO} A_{LAD} + L_{LAO} A_{LAD}\right)$, where $A_{LAD}$ and
$L_{LAD}$ are the areas enclosed by the LV contours in the RAO
and LAO projections, respectively, and $L_{RAO}$ and $L_{LAO}$ are
the lengths of the LV long axis in the RAO and LAO projec-
tions, respectively. The contours were drawn manually in all
frames from two consecutive, well-opacified cardiac cycles
using custom-made software.

Protocol

Measurements were performed at baseline, after treat-
ment with dobutamine (2.5 μg·kg$^{-1}$·min$^{-1}$·iv), after volume
load (200 ml iv gelofusine over a 12-min period), and after
infusion of propranolol (1 mg/kg iv). In each condition, $\sigma_N$
was measured, three consecutive hypertonic saline injections
(2 ml, 10% saline) were performed to determine $G^P$, and
biplane angiography was performed. Recording of simulta-
neous conductance signals was started ~5 s before contrast
injection and continued during the acquisition of angiog-
graphic images. Because respiration may affect $G^P$ and ac-
tual end-diastolic volume, all data were acquired during
apnea at end expiration. The respirator was disconnected
3–5 s before the actual data acquisitions.

Comparison of Angiographic and Conductance Signals

Angiographically derived parallel conductance and $\alpha$. Con-
trast medium changes the conductivity of blood; therefore, we
compared the angiographically derived volume signals with
the conductance signals obtained just before contrast injec-
tion. To smooth the conductance signal and to avoid selection
bias, four consecutive cardiac cycles were selected and tem-
porally averaged. The conductance and angiographic signals
were synchronized by matching the peaks (end-diastolic vol-
ume) of the two signals. The temporal resolution of the
angiographic signal was 40 ms and that of the conductance
signal was 4 ms; consequently, comparisons were made using
data points at 40-ms intervals only. The data points from
both signals were plotted vs. each other, and a linear regres-
sion was performed. The y-intercept represents the hypothet-
cal conductance when angiographic volume equals zero and
thus represents an estimate of $G^P$ based on direct comparison
with instantaneous angiographic volume. Statistical analysis
of the comparative data [$G^P_{saline}$ vs. $G^P_{obtain}$] was performed by standard linear regression
and Bland-Altman analysis (4).

The slope of the relation d$dG$/d$V_{Angio}$ was used to determine $\alpha$. Given the definition of $\alpha$, $\alpha = (L^2/\sigma_N)(dG/dV_{Angio})$.
In addition, the linear correlation coefficient ($R^2$) and the
standard error of the $y$ estimate were used to test the linearity
of the relation and, thus, support the assumption that $\alpha$ re-
mains constant during the cardiac cycle.

Time-varying $G^P$. The analysis described above determines
$G^P$ averaged over the full cardiac cycle. To investigate
whether $G^P$ varies during the cardiac cycle, the conductance
signal was calibrated using the coefficients (slope and inter-
cept) obtained from the linear regression, and subsequently
the angiographic signal was subtracted from this calibrated
conductance-volume signal. The resulting difference signal
represents the errors remaining after correction for mean $G^P$
and can be interpreted as variations in $G^P$ during the cardiac
cycle. Mathematically, $G^P$ can be written as follows: $G^P(t) = G^P + dG^P(t)$, were $G^P$ is the average (constant) parallel
conductance and $dG^P(t)$ the time-varying component. $V_{Angio}$
is taken as the gold standard; thus it is assumed that
$V_{Angio}(t) = (1/\sigma_N)(L^2/\sigma_N)G^P(t) - [G^P + dG^P(t)]$; $\alpha$ and $G^P$
are obtained as the slope and intercept, respectively, of the rela-

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tion between angiographic and uncalibrated conductance volume, as described above. Thus \(dG^P(t) = [V_{\text{Cath}}(t) - V_{\text{Angio}}(t)]/ \left[\frac{1}{a} \left(\frac{L^2}{s_b}\right)\right]\), where the calibrated conductance volume \(V_{\text{Cath}}(t) = \left(\frac{1}{a} \left(\frac{L^2}{s_b}\right)\right)G(t)\). To enable comparisons between animals and between conditions despite changes in heart rate, the conductance and angiographic signals were fitted with a cubic spline, resampled at 500 time points, and plotted on a normalized time scale.

Variability of \(G^P\)

Variabilities of \(G^P\) (\(G_{\text{saline}}^P\) and \(G_{\text{Angio}}^P\)) were quantified in the following multiple linear regression model: \(G^P = a^0 + \sum a^A_i A_i + \sum a^C_i C_i\). The dummy variables \(A_i\) account for between-animal differences, allowing each animal to have a different mean value (effects coding). The standard deviation of the group of animal coefficients, \(a^A_i\), is a measure of inter-animal variability of \(G^P\). The dummy variables \(C_i\) code the various conditions (baseline, dobutamine, gelofusine, and propranolol), with reference cell coding with the baseline condition as the control group (14, 22). Consequently, the offset \(a^0\) yields mean \(G^P\) at baseline and the coefficients \(a^C_i\) quantify the differences in the various conditions compared with baseline. The same statistical analysis was also applied for \(G_{\text{saline}}^P\) and parallel conductance volumes obtained by hypertonic saline injection (\(V_{\text{saline}}^P\)) and by angiography (\(V_{\text{Angio}}^P\)).

RESULTS

Typical examples of uncalibrated conductance volume signals, \(L^2/s_b G(t)\), and angiographic volume signals in the four hemodynamic conditions are shown in Fig. 1. Linear extrapolation of all data points during the cardiac cycle, \(G(t)\) vs. \(V_{\text{Angio}}(t)\), yielded \(G_{\text{Angio}}^P\), as shown in Fig. 2. These values were compared with \(G_{\text{saline}}^P\). All pooled data are shown in Fig. 3 (linear regression) and Fig. 4 (Bland-Altman plot). These findings indicate an excellent linear relation between the two methods. Bland-Altman analysis reveals a bias (± 2 SD) of 0.002 ± 0.112 \(\Omega^{-1}\), indicating an essentially zero bias with a standard deviation of 8.4% of mean \(G^P\).

Statistical analysis of the variabilities of \(G_{\text{saline}}^P\) and \(G_{\text{Angio}}^P\) between animals and between hemodynamic conditions is given in Table 1. The results show a mean \(G_{\text{saline}}^P\) of 0.661 ± 0.015 \(\Omega^{-1}\) at baseline. This standard deviation does not include the interanimal variability. The variability between animals was 0.17 \(\Omega^{-1}\), or 26% of baseline \(G_{\text{saline}}^P\). Variation between conditions was substantially less, but \(G_{\text{saline}}^P\) was significantly smaller during dobutamine (−0.065 \(\Omega^{-1}\), or −9.8% of baseline).
and significantly larger during propranolol (+0.058 \(\Omega^{-1}\), or +8.7% of baseline). Results for \(G^P_{Angio}\) were essentially the same (Table 1), as also illustrated by Fig. 5, which shows mean \(G^P_{Angio}\) and \(G^P_{Saline}\) at the various conditions. \(\sigma_b\) varied significantly between animals (8.4%), although much less than \(G^P\). The condition variabilities of \(\sigma_b\), however, were very similar to those of \(G^P\), suggesting that these changes in \(G^P\) were largely due to underlying changes in \(\sigma_b\). This suggestion is substantiated by the fact that parallel conductance volume (\(V^P\)), which is equal to \((L^2/\sigma_b)G^P\), did not differ significantly between conditions.

Figure 6 shows the variations of \(G^P\) during the cardiac cycle as a percentage of its mean value. The average variation was small and not significantly different from zero; it ranged from +1.8 ± 2.4% in the mid-ejection phase to −1.3 ± 2.6% during early filling.

**DISCUSSION**

Conversion of raw conductance catheter data to calibrated absolute volumes requires the assessment of \(G^P\). \(G^P\) can be determined by direct comparison of the conductance signals with an independent measurement of absolute LV volume, such as by angiography (20), dimension crystals (2), or balloon volume (6). In addition, several methods have been proposed that enable assessment of \(G^P\) without the need for independent volumetric measurements. Baan et al. (3) used a suction method in which cavity volume was reduced to zero by means of a multihole catheter in the LV or by manual compression. The results indicate reasonable correspondence with the saline method, but the nature of this intervention clearly limits its practical application. Recently, White et al. (31) introduced a novel approach that relies on the analysis of the transient reduction of volume induced, e.g., by balloon occlusion of the inferior vena cava. By extrapolating the relation between end-diastolic volume and end-systolic volume during the intervention to a point where these volumes are equal, an estimate of the offset volume corresponding to \(G^P\) is obtained. The key assumption in this approach is that ejection fraction is constant during the volume reduction. This hemodynamic requirement may explain why the results show good correlation.

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**Table 1. Multiple linear regression analysis**

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<th>(G^P_{Saline})</th>
<th>(G^P_{Angio})</th>
<th>(\sigma_b)</th>
<th>(V^P_{Saline})</th>
<th>(V^P_{Angio})</th>
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<td>(a_0) (Dobu)</td>
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Model: \(y = a_0 + \sum a_i x_i + \sum a_{i-2} C_i\). \(G^P_{Saline}\) and \(G^P_{Angio}\), parallel conductance by hypertonic saline method and by angiography; \(\sigma_b\), specific conductivity of blood; \(V^P_{Saline}\) and \(V^P_{Angio}\), parallel volume by hypertonic saline method and by angiography; Dobu, dobutamine; Gelo, gelofusine; Prop, propranolol; \(\%iav\), percent interanimal variability compared with baseline; \(\%iav\), percent intercondition variability compared with baseline. Offset \(a_0\) predicts mean baseline value of dependent \(y\). Animal variables \(a_i\) quantify differences between animals; condition variables \(a_{i-2}\) predict mean changes in corresponding conditions compared with mean baseline. Thus predicted value in, e.g., animal 3 in gelofusine condition is calculated as \(a_0 + a_2 + a_5\). *\(P < 0.05\) vs. baseline; †\(P < 0.01\) vs. baseline.
with the saline method in some study groups but poor agreement in several other groups. Gawne et al. (12) introduced the so-called dual-frequency method, which exploits the fact that, in the 2- to 100-kHz frequency range, blood conductivity is essentially constant whereas muscle conductivity varies. By comparing measured conductance catheter signals at two frequencies (3.3 and 33 kHz), an estimate of $G^P$ was obtained.

An inherent disadvantage of this method is that changes in frequency-independent components of $G^P$ (i.e., the blood in the RV) will not be picked up. Although the results by Gawne et al. were promising, more recent studies in neonatal and adult pigs (32) have been disappointing. However, the method appears to be applicable in mice, presumably because $G^P$ in these hearts resides almost exclusively in the relatively thick myocardial wall (13). However, the method most widely used to determine $G^P$ is the hypertonic saline method (3). The present study addressed the following three issues with regard to this method: the absolute accuracy of the hypertonic saline method for assessment of $G^P$, the variability of $G^P$ between animals and between hemodynamic conditions, and the variability of $G^P$ during the cardiac cycle.

**Accuracy of the Hypertonic Saline Method**

Testing of the absolute accuracy of the hypertonic saline method requires an independent measurement of absolute volume. In the present study, the uncalibrated conductance data were plotted vs. absolute volumes obtained by biplane cineangiography for all time points during a full cardiac cycle. Extrapolation of this relation to a hypothetical zero angiographic volume yields an estimate of $G^P$. This estimate was compared with the value obtained by saline dilution. Our results show an excellent agreement between these two methods. Linear regression showed a good correlation ($R^2 = 0.92$), an essentially zero offset, and slope equal to 1.0, whereas Bland-Altman analysis yielded a nonsignificant bias ($\pm 2$ SD) of $0.002 \pm 0.112\ \Omega^{-1}$. Previous results using the single-field conductance method in patients vs. monoplane cineangiography (3) indicated a nonsignificant underestimation of 6.5%. A somewhat larger and significant underestimation of 14% was found by Boltwood et al. (5). As in our study, biplane angiography was used to obtain an independent estimate of $G^P$, but hemodynamic conditions were altered by partial balloon occlusions of the aorta, inferior vena cava, or pulmonary artery, whereas we used pharmacological interventions and volume infusions. We chose the latter, because, in our experience, it is rather difficult to produce well-defined steady-state conditions with partial occlusions, and thus it was anticipated that it would be difficult to perform (multiple) saline injections while maintaining steady-state hemodynamics. In the study of Boltwood et al., $G^P_{\text{Angio}}$ and $G^P_{\text{saline}}$ were obtained from repeated occlusions; although the aim of these occlusions was to reach the same hemodynamic condition, they may have contributed to a correlation between individual $G^P_{\text{saline}}$ and $G^P_{\text{Angio}}$ that was substantially less than in our study. Another factor may have been that, in our study, $G^P_{\text{saline}}$ was obtained as the mean of three repeat injections, whereas the analysis of Boltwood et al. was based on single injections. Burkhoff et al. (6) tested the saline method in isolated rabbit hearts in which the independent volume measurement was obtained from an intraventricular balloon. Their results indicate a relatively small, nonsignificant underestimation (8% of mean) by the saline method. These findings were later confirmed by Lankford et al. (19). Applegate et al. (2) found no significant differences between $G^P$ calculated by the saline method and estimated by the regression of conductance vs. LV volume by ultrasonic endocardial crystals in intact dogs. Studies in the intact piglet heart presented by Cassidy and Teitel (7) compared conductance volume with biplane cineangiography. End-systolic and end-diastolic volumes were obtained, and the pooled data from all animals in various hemodynamic conditions were used to determine the relation with angiographic volumes. They showed that, after individual correction for $G^P$ obtained with the saline method, the linear relation between the two methods had a small offset (1.2 ml), which may be interpreted as an underestimation of “true” $G^P$ (8% of mean). The variability of the offset between animals was 0.94 ml, indicating that the underestimation was systematically found in most animals. Similar results were obtained by Szwarz et al. (27) in a comparison of conductance-derived and radionuclide volumes in dogs: The relation between end-diastolic volumes by both methods showed a nonsignificant offset of 7.5 ml, again suggesting an underestimation of true $G^P$ by the saline method of ~8%. In several other studies, absolute volume obtained with the conductance catheter was compared with independent methods such as echocardiography (1), angiography (3), and magnetic resonance imaging (21). These studies, generally, show a
good correspondence with conductance-derived volumes and thus provide (indirect) evidence for the accuracy of the saline method. It is important to note that all cited studies were performed with single-field excitation of the conductance catheter, in contrast with the present study, where dual-field excitation was used. The improved linearity of the dual-field method (24) may explain the more accurate $G_{\text{saline}}$ estimates.

A limitation in most studies lies in the dependence of the results on $\alpha$. In principle, the error in the $G_{\text{saline}}$ can be determined directly by subtracting an independent volume measurement from the conductance-derived absolute volume. However, this approach requires that $\alpha$ is determined, which introduces an error source. Alternatively, analysis of conductance volumes corrected for $G^p$, but not for $\alpha$, is possible, but only when multiple data points are available from different animals or from different hemodynamic conditions. In that case, the error in $G^p$ can be derived from the offset of the relation between volume by conductance and by the independent method. Although the latter approach does not require measurement of $\alpha$, it does implicitly assume that the variability in $\alpha$ (between animals or between conditions) is small. In contrast, in our study, the relation with angiography was obtained by using all data points from a full cardiac cycle. Thus the only implicit assumption is that $\alpha$ is constant (but not necessarily 1.0) within the cardiac cycle, and neither the absolute value of $\alpha$ nor variations of $\alpha$ between animals or between hemodynamic conditions affected our results. A previous study by Szwarc et al. (26), using single-field excitation, yielded significant, but small, changes in $\alpha$ during ejection. Our results (Table 2), with dual-field excitation, show an excellent linear relation between conductance and angiographic volume and indicate that, for all practical purposes, $\alpha$ can be regarded as constant during the cardiac cycle.

Variability of $G^p$ Between Animals and Between Hemodynamic Conditions

Quantification of the variability of $G^p$ is essential to determine the need for repeated assessments. Previous studies have clearly indicated that the variability between animals is substantial even when the age and weight of the study group are within fairly narrow ranges. The most likely causes for this variability are differences in the catheter position, the size and geometry of the heart, the position of the heart in the thorax, and the geometry of other structures in the thorax. In addition, there will be between-subject differences in the conductive properties of blood and tissues. In the present study, the interanimal variability of $G_{\text{saline}}^p$ was 26.3% of the mean value (for $G_{\text{Angio}}^p$ the interanimal variability was 25.7%). These results match findings in previous studies in sheep (23) (20.0%), dogs (5, 17) (20.2 and 27.3%, respectively), and newborn lambs (18) (29.7%). The variability in patient studies is generally somewhat wider, most likely due to the less uniform study group: a between-patient variability of 30.0% can be derived the study of White et al. (34) in children; Baan et al. (3) reported 24.8% in adult patients; and Kass et al. (16) show a variability of 22.6% in normal patients and 24.1% in patients with LV hypertrophy. These results indicate that assessment of $G^p$ in individual subjects is required.

Within-subject variability of $G^p$ was investigated in the present study by repeating the assessments after interventions aimed at inducing different hemodynamic conditions. The results in Table 1 indicate that, during dobutamine infusion, $G_{\text{saline}}^p$ was decreased by $-9.8\%$ ($G_{\text{Angio}}^p$ by $-8.6\%$), whereas after propranolol infusion, $G_{\text{saline}}^p$ increased by $+8.7\%$ ($G_{\text{Angio}}^p$ by +6.4%). Previous studies have shown that $G^p$ is sensitive to changes in RV volume (5, 6, 23); thus these changes may reflect the effects of dobutamine and the effects of propranolol on the latter. However, $\sigma_b$ was also changed: during dobutamine infusion, $\sigma_b$ decreased by $-8.5\%$ compared with baseline, whereas after propranolol infusion, $\sigma_b$ was +8.3% higher than control. These changes in $\sigma_b$ reflect changes in hematocrit, which, during dobutamine infusion, may be increased because of an increased transcapillary fluid shift or red cell recruitment from the spleen (8, 11, 15, 30), whereas, conversely, gelofusine infusion leads to a decrease in hematocrit and, thus, an increase in $\sigma_b$. Thus, because the physical structures that contribute to $G^p$ (i.e., myocardial wall, RV cavity, lungs) contain blood, the changes in $G^p$ may, in fact, be largely due to changes in $\sigma_b$ rather than geometrical changes. Interestingly, this means that the parallel conductance volume, which is calculated as $V^p = (L^2/\sigma_b)G^p$, would be much less affected. Indeed, statistical analysis (Table 1) shows that changes in $V^p$ were <2.4% and not statistically significant.

Table 2. Slope factor, $R^2$, and SEE of relation between conductance volume and angiographic volume

<table>
<thead>
<tr>
<th></th>
<th>$\alpha$</th>
<th>$R^2$</th>
<th>SEE, ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>0.67 ± 0.20</td>
<td>0.983 ± 0.012</td>
<td>1.50 ± 0.65</td>
</tr>
<tr>
<td>Dobutamine</td>
<td>0.65 ± 0.22</td>
<td>0.983 ± 0.008</td>
<td>1.05 ± 0.38</td>
</tr>
<tr>
<td>Gelofusine</td>
<td>0.89 ± 0.25*</td>
<td>0.981 ± 0.011</td>
<td>2.03 ± 0.47</td>
</tr>
<tr>
<td>Propranolol</td>
<td>0.90 ± 0.31*</td>
<td>0.981 ± 0.017</td>
<td>1.67 ± 0.37</td>
</tr>
</tbody>
</table>

Values are means ± SE, $\alpha$, slope factor; $R^2$, standard error of y estimate. Conductance volume $= (L^2/\sigma_b)G(t)$, where $L$ is catheter electrode spacing, $\sigma_b$ is specific conductivity of blood, and $G(t)$ is time-varying conductance. *P < 0.05 vs. baseline.
The dependence of $G_P$ (or $V_P$) on hemodynamic conditions has been investigated in several previous studies. Szware et al. (27) measured $V_P$ by the saline method in intact dogs at baseline, after volume loading, and after bleeding and did not find significant differences despite large changes in hemodynamic status. Boltwood et al. (5) estimated $V_P$ by repeated saline injections under a variety of loading conditions: compared with control, $V_P$ was unchanged during occlusion of the pulmonary artery or the aorta but was significantly reduced (~9%) during occlusion of the inferior vena cava. This may reflect the influence of reduced RV volume during caval occlusion. The influence of RV volume was also demonstrated in a study from our group (23) where embolization of the right coronary artery, dilating the RV, caused a 20% increase in $G_P$. In general, these changes in $G_P$ do not invalidate the method but indicate the need to reassess $G_P$ after substantial changes in hemodynamic conditions.

**Variability of $G_P$ During the Cardiac Cycle**

The saline method yields a single value for $G_P$; however, RV filling and ejection, atrial filling, changes in myocardial shape, and blood content could potentially cause changes in $G_P$ during the cardiac cycle. Our results, however, indicate that such changes are very small and can, in practice, be neglected. Previously, cyclic $G_P$ (or $V_P$) variations have only been studied for single-field excitation. White et al. (33, 34) assessed cyclic variation of $V_P$ by plotting isochronal uncalibrated conductance volume vs. conductance stroke volume: the $y$-intercept for each set of isochronal points was used as an estimate of $V_P$ at the time during the cardiac cycle corresponding to the isochrone. The method, however, contains a theoretical flaw: on the one hand, it aims to determine a time-varying $G_P$; on the other hand, it assumes that conductance stroke volume is directly proportional to blood conductivity and, thus, implicitly requires that $G_P$ is equal at end diastole and end systole (or any other pair of points during the cardiac cycle that are used to calculate “apparent” stroke volume). Thus, rather than at apparent stroke volume equals zero, the $y$-intercept should be determined at $x = V_{ED} - V_{ES}$, or the difference in $V_P$ between end diastole and end systole. Despite this problem, the results probably give a reasonable estimate of the relative magnitude of the cyclic variation of $V_P$, which was found to be 5.8% of end-diastolic volume in the human RV (33) and 4.3% in the LV (34). A similar approach by Lankford et al. (19) yielded non-significant variations on the order of 4% of end-diastolic volume. These authors also directly compared conductance-derived and intraventricular balloon volume in isolated heart, which yielded similarly small cyclic variations in $V_P$.

**Conclusions**

The main finding in this study is that the hypertonic saline method accurately determines $G_P$ for dual-field conductance catheter compared with biplane angiography. The dual-field method produced $G_P^{saline}$, which, compared with $G_P^{Angio}$, showed an essentially zero bias and narrow limits of agreement. Compared with previous studies using single-field excitation, which generally show a slight underestimation of $G_P$, dual-field excitation appears to be superior. The variability between animals was substantial and could not be explained by variability in blood conductivity. The within-animal variability was much smaller and was largely related to changes in blood conductivity. Clearly, the variability in $G_P$ between animals and between conditions is not due to inaccuracy of the saline method but is a true biological variability, since the angiographic method shows almost identical changes. Finally, the variations in $G_P$ during the cardiac cycle were found to be negligible.

**APPENDIX**

The conventional method for determination of $G_P$ from end-diastolic conductances ($G_{ED}$) and end-systolic conductances ($G_{ES}$) obtained after hypertonic saline injections consists of plotting $G_{ED}$ vs. $G_{ES}$ for all beats during the saline washin period and calculating the intercept of the relation through these points with the line of identity. The alternative approach used in the present study (see METHODS) is mathematically identical, but it better illustrates that $G_P$ is determined at the hypothetical point where $s_b = 0$, and thus the only remaining conductance is $G_P$. The $s_b$ cannot be directly measured, but, as shown below, “stroke conductance” ($SG = G_{ED} - G_{ES}$) is directly proportional to $s_b$. Therefore, rather than $s_b$, $G_{ED}$ is plotted vs. $G_{ES}$ and the relation is extrapolated to the point where $G_{ED} - G_{ES} = 0. This way, similar to the conventional method, this alternative approach determines $G_P$ at the hypothetical point where $G_{ED} = G_{ES}$ (Fig. 7).

Absolute end-diastolic volume ($V_{ED}$) = ($L/a)(L^2/\sigma_b)(G_{ED} - G_P$), and absolute end-systolic volume ($V_{ES}$) = ($L/a)(L^2/\sigma_b)(G_{ES} - G_P$). Thus $SV = V_{ED} - V_{ES} = (L/a)(L^2/\sigma_b)(G_{ED} - G_{ES})$, from which it can be derived that $\sigma_b = (L/a)L^2\sigma_b/SV$. Because $\alpha$, $L$, and $SV$ are constant, $SG$ is directly proportional to $\sigma_b$.

**REFERENCES**

PARALLEL CONDUCTANCE BY THE SALINE METHOD