Effective cerebral perfusion pressure: does the estimation method make a difference?

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ABSTRACT

**Introduction:** The effective cerebral perfusion pressure (CPPe), zero-flow pressure (ZFP), and the resistance area product (RAP) are important determinants of cerebral blood flow. ZFP and RAP are estimated by linear regression analysis of pressure–velocity relationships of the middle cerebral artery. Aim of the study was to validate four other estimation methods against the linear regression method.

**Methods:** In a former study, recordings of EEG, blood pressure and mean cerebral artery velocity (transcranial Doppler) were obtained in patients, during internal cardioverter defibrillator implantation procedures in order to determine EEG frequency ranges that represent ischemic changes during periods of circulatory arrest. In this secondary analysis CPPe, ZFP and RAP estimated by four different methods, the 3-point intercept calculation (LR3, systolic/mean/diastolic), Czosnyka (CZO, systolic/diastolic), Belford (BEL, mean/diastolic), and Schmidt (SCH, systolic/diastolic) were validated against the reference method (LR, linear regression). CPPe is calculated as the difference between mean arterial pressure and ZFP. Primary endpoint was difference, correlation, and agreement of these CPPe measurements.

**Results:** In total, 174 measurements in 35 patients under steady state conditions during general anesthesia before the first cardiac arrest phase were collected. CPPe ZFP and RAP measurements based on LR3 and CZO calculation methods showed small mean differences, good agreement, low percentage errors, and an excellent correlation when compared to references method. Agreement and correlation were moderate for the BEL method, and unsatisfying for the SCH method.

**Conclusions:** CPPe, ZFP, and RAP measurements based on LR3 and CZO calculation methods are comparable to the LR reference method.

**Keywords:** Transcranial Doppler sonography, Cerebral circulation, Zero-flow pressure, Critical closing pressure, Cerebral perfusion pressure, Resistance area product, Agreement
INTRODUCTION

Knowledge of the cerebral perfusion pressure (CPP) and its components are important in understanding and treating patients with intracranial pathologies in the perioperative period. When calculating the CPP, the mean arterial pressure (MAP) has been used as effective upstream pressure and the intracranial pressure (ICP) as effective downstream pressure of the cerebral circulation, because of a Starling resistor phenomenon located at the level of cerebral veins ('classical model' \( \text{CPP}_{i} = \text{MAP} - \text{ICP} \)). However, the “classical model” has limitations, because it assumes that cerebral blood flow or blood flow velocity only reach zero when the arterial blood pressure is zero, which is unlikely. Using solely intracranial pressure as a measure of the effective downstream pressure of the brain, would neglect vascular tone properties of the vessels.

In vivo pressure-flow relationships per cardiac cycles are approximately straight lines in many vascular beds such as the cerebral vessels. Thus, the zero-flow pressure (ZFP), the pressure when flow ceases, can be extrapolated by linear regression of instantaneously obtained data pairs of pressure and flow (velocity). The ZFP represents the vasomotor tone. The inverse slope of the pressure-flow plot represents vascular bed resistance. The effective cerebral perfusion pressure (CPPe) is thus better defined by the difference between mean arterial pressure (MAP) and cerebral ZFP ('alternative model' \( \text{CPP}_{e} = \text{MAP} - \text{ZFP} \)). Assessment of ZFP may help to evaluate cerebral vascular reserve, indicating the degree of vasomotor tone. If tone is low this would suggest maximal vasodilation and indicate that the patient may not tolerate anemia or hypotension.

Since the introduction of transcranial Doppler sonography, a number of methods have been developed to assess cerebral ZFP, CPPe, and vascular resistance by pressure-velocity relationships (figure 1). The “resistance area product (RAP)”, a Doppler ultrasound index, characterizes vascular resistance according to the inverse slope of the linear pressure-velocity relationship. Deciding which method is the most appropriate for ZFP, RAP and CPPe measurements is still challenging.

We used data from a prospective, controlled, observational clinical study detecting cerebral ischemia caused by short periods of circulatory arrest during internal cardioverter defibrillator device (ICD) implantation and testing. In this study electroencephalogram (EEG), transcranial Doppler sonography (Vmca) and invasive arterial blood pressure was continuously measured under general anesthesia. In a secondary analysis we estimated CPPe, ZFP, and RAP by four different methods and validated them against the reference method (LR, linear regression). We hypothesized that estimation based on three points (systolic, mean, and diastolic) is superior to a two-point estimation method (systolic/diastolic or mean/diastolic) when compared to regression analysis of the whole wave tracings (reference method).
Figure 1. Estimation of cerebral zero flow pressure (ZFP) and resistance area product (RAP). Arterial blood pressure (ABP) and velocity of the mean cerebral artery (Vmca) are usually graphically plotted against time (A). The time delay between the two waveforms has been synchronized before ZFP estimation. The zero-flow pressure (ZFP), the pressure when flow ceases, can be extrapolated by linear regression of the instantaneous pairs of the pressure-flow (velocity) relationship, which represents vasomotor tone while its inverse slope (RAP) represents the value of vascular bed resistance (C). The beat-to-beat values of ZFP and RAP provide an indication of the short-term variability of these estimates. Therefore, many research groups use multiple cardiac cycles (i.e. 10-12 heartbeats within two breaths) to determine the ZFP and RAP. The ZFP_{LR3} was estimated by x-axis intercept determination from the linear regression line based on corresponding data pairs of systolic, mean and diastolic values of ABP (abscissa) and Vmca (ordinate, panel D). Other methods use a formula to calculate ZFP, which is based on the slope-intercept form. With corresponding pairs of systolic/diastolic (ZFP_{CD}, ZFP_{SCD}, panel E) or mean/diastolic (ZFP_{BEL} panel F) values of blood pressures and MCA velocity of the same time period it is also possible to calculate the intercept pressure at zero flow.

METHODS

Design

In a former study, recordings of EEG, arterial blood pressure and mean cerebral artery velocity (transcranial Doppler) were obtained in patients, during the whole internal cardioverter defibrillator implantation procedure in order to determine EEG frequency ranges that represent ischemic changes during short periods of circulatory arrest. Patients were eligible for inclusion if scheduled for ICD implantation or replacement. Exclusion criteria were patient refusal, active neurological disease, and a history of cerebrovascular disease, brain injury, or intracranial surgery. All patients were informed of the purpose of the study and provided written informed consent before being enrolled. The Medical Ethical Committee of the University of Utrecht provided ethical approval for the original study (Nr.:92/59). The study project followed the recommendations of the
Declarations of Helsinki and the European Union Commission and European Medicines Agency. The trial was planned and done before CONSORT 2010. Dutch laws did not require international registration of this type of clinical trial at that time.

This report is a secondary analysis of the former recordings of arterial blood pressure and mean cerebral artery velocity in the phase before the induction of cardiac arrest by ventricular fibrillation. We estimated CPPe, ZFP, and RAP by four different methods and validated them against the reference method.

Endpoints
The main endpoint of this investigation was the CPPe based on five different ZFP estimations methods (CPPe=MAP–ZFP). The ZFP and RAP were estimated by five different methods: (1) the 3-point intercept estimation (LR3), (2) Czosnyka (CZO, 2 point systolic/diastolic), (3) Belford (BEL, 2 point mean/diastolic), and (4) Schmidt (SCH, 2 point systolic/diastolic), which were validated against the (5) reference method (LR, linear regression).

Anesthesia Procedure
Anesthesia was standardized to reduce the variance in measurements. Patients were premedicated orally with 2.5 mg lorazepam 2 hours before arrival in the operating theatre. Anesthesia was induced by intravenous administration of 4 µg/kg fentanyl, 0.1 mg/kg midazolam 0.1-0.2 mg/kg etomidate, and 0.07 mg/kg pancuronium. Anesthesia was maintained with 7-10 µg/kg/h of fentanyl and 150 µg/kg/h of midazolam. All patients received an endotracheal tube, a nasogastric tube, a urine catheter, a peripheral intravenous line, an arterial catheter, a Swan-Ganz catheter, and a central venous catheter. For jugular venous bulb oximetry measurements, a fiberoptic catheter was inserted retrogradely into the right internal jugular vein. Positioning of the tip of the catheter into the jugular bulb was confirmed by fluoroscopy. Ventilation was performed with a mixture of oxygen in air (FIO₂ 0.35). Steady state end-tidal partial pressure of carbon dioxide was kept normal between 30 and 43 mmHg (4.0 and 5.6 kPa). Nasopharyngeal temperature was maintained between 35.5°C and 37.0°C.

Study protocol & measurements
All measurements were performed during hemodynamic and respiratory steady-state conditions by the same neurophysiological research team.

Arterial blood pressure (ABP) was monitored via a cannula placed in the radial artery with the transducer positioned at the level of the base of the skull/ear. Cerebral blood flow velocity in both middle cerebral arteries (Vmca) was monitored with transcranial Doppler sonography device (DWL multidop-X device with two 2-MHz pulsed Doppler probes, DWL TCD 7 software, DWL Elektronische Systeme GmbH, Sipplingen, Germany).
All patients were positioned supine on an operation table. The patient’s heads were positioned and fixed in-line in midline position on a pillow. The ultrasound transducer was placed over the temporal area just above the zygomatic arch and in front of the tragus of the ear. After identification of the middle cerebral artery, the depth of insonation was adjusted in 1-2 mm increments to obtain signals from the proximal segment (M1) of the MCA. Accurate care was taken to ensure a stable position of the ultrasound probe during the study period. The ultrasound probe was therefore fixed using a monitoring probe holder attached to the patient’s head (DWL, Sipplingen, Germany). Doppler ultrasound variables such as depth, gain, sample volume, and power of the ultrasound beam were unchanged during the measurement procedure. At each measurement, simultaneous data recordings of Vmca, arterial blood pressure (ABP, mmHg), expiratory concentration carbon dioxide concentration (PeCO₂, mmHg), and jugular venous bulb oxygen saturation (SjO₂, %) were acquired and stored on a microcomputer via analogue/digital converters.

**Estimation of the ZFP by pressure-flow velocity plot analysis.**

The analyses of the ZFP, CPPe and RAP have been performed after the study period. For the secondary analyses, we looked at the period after induction of anesthesia and before ICD-testing. We extracted five consecutive data sets of 10-12 seconds (two breaths) each within 3 min from the raw data files of the primary investigation.

For the CPPe/ZFP analysis we used dedicated custom-developed software (LabView®, USA, written by Dr. E.G. Mik, Erasmus MC). After visual inspection of the pressure curves and the velocity waveforms, we used corresponding pairs of ABP (abscissa) and Vmca (ordinate, figure 1 C) curves from 2 breathing cycles, (10-12 cardiac cycles) to determine ZFP and RAP. Hysteresis in the pressure-flow velocity plots because of a time delay between pressure and flow velocity recordings was minimized for each heartbeat by synchronizing the point of maximal positive slope of both waveforms. ZFP and RAP were calculated for every heart cycle by regression analysis of the pressure-flow velocity plot. Extrapolated ZFP and RAP data of all heart beats within these respiratory cycles were averaged for further analysis.²⁷¹⁶ For a detailed description we refer to appendix 1 in the supplemental information content section.

The ZFPLR was estimated by x-axis intercept determination from the linear regression line based on corresponding data pairs of systolic, mean and diastolic values of ABP (abscissa) and Vmca (ordinate, figure 1 D). The other methods primarily use formulas to calculate ZFP. With diastolic, mean or systolic values of blood pressures and flow-velocity from the same time period it is possible to calculate the intercept pressure at zero flow. All formulas used in this investigation had been suggested by Belfort and colleagues¹⁴, Czosnyka and colleagues¹⁰ and Schmidt and colleagues¹¹¹⁵ (see table 1). For a detailed description of the mathematical basis of the calculation methods we refer to appendix 1 and 2 in the Supplemental Digital Content (SDC) section.
When using transcranial Doppler sonography, an estimate of vascular resistance has been defined as the inverse slope of a linear regression line of the driving blood pressure and flow velocity, the resistance area product (RAP).\textsuperscript{6,12} Subsequently, we calculated the RAP as: $\text{RAP} = \frac{\text{MAP} - \text{ZFP}}{\text{mean } V_mca}$. The CPPe of all methods was calculated from the difference between MAP and the respective ZFP. For a detailed description of the CPPe model we refer to appendix 1 in the Supplemental Digital Content (SDC) section.

**Statistical analysis**

This analysis was based on the recommendation of current guidelines for reporting agreement studies.\textsuperscript{17,18} In this secondary analysis study setting a prior sample size calculation could not be applied.\textsuperscript{13} Each patient served as his own control.

Normal distribution of data was assessed both visually by inspection of histograms and with the D'Agostino-Pearson omnibus K2 method.\textsuperscript{19} Results are presented as mean values (standard deviation, SD) unless otherwise stated. Four methods of CPPe, ZFP and RAP estimation (LR3, BEL, CZO SCH) were compared to the reference method (LR) by ANOVA with correction for multiple comparison (Dunnett). The relationship between the four methods of CPPe, ZFP and RAP estimation (LR3, BEL, CZO SCH) and the reference method (LR) was examined by regression analysis and by using Pearson's or Spearman correlation tests, depending on data distribution.

To evaluate the *accuracy* (i.e. the closeness of ZFP, RAP or CPPe measurements to its reference value) of the $ZFP_{LR}$ versus $ZFP_{LR3}$, $ZFP_{BEL}$, $ZFP_{CZO}$, and $ZFP_{SCH}$ we described the *agreement* between these paired data using the Bland-Altman analysis with correction for multiple measurements per subject.\textsuperscript{20} The *bias* (mean difference between the reference and test-method) represents the systematic error between the two methods. As a measure of *precision* (i.e. the distribution of repeated measurements), the limits of agreement (LoA) were commonly calculated as bias +/- 1.96 SD, defining the range in which 95% of the differences between the methods are expected to lie.\textsuperscript{21} In this analysis

### Table 1. ZFP estimation by formulas

<table>
<thead>
<tr>
<th>Method</th>
<th>Formula</th>
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<tbody>
<tr>
<td>Belfort\textsuperscript{19}</td>
<td>$ZFP_{BEL} = \text{MAP} - \text{Vm} \cdot \frac{\text{MAP} - \text{DAP}}{\text{Vm} - \text{Vd}}$</td>
</tr>
<tr>
<td>Czosnyka\textsuperscript{15}</td>
<td>$ZFP_{CZO} = \text{SAP} - \text{Vs} \cdot \frac{\text{SAP} - \text{DAP}}{\text{Vs} - \text{Vd}}$</td>
</tr>
<tr>
<td>Schmidt\textsuperscript{16,20}</td>
<td>$ZFP_{SCH} = \text{MAP} - \frac{\text{Vd}}{\text{Vm}} \cdot (\text{MAP} \cdot \text{Vd} + 14)$</td>
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The effective zero flow pressure (ZFP) estimated by different methods: Belford (BEL, 2-point: mean/diastolic), Czosnyka (CZO, 2-point: systolic/diastolic), and Schmidt (SCH, 2-point: systolic/diastolic). DAP, MAP, SAP = diastolic, mean, and systolic arterial pressure, Vd, Vm, Vs = diastolic, mean, and systolic blood flow velocity. One CPPe measurement corresponds to the cumulative analysis of 10-12 cardiac cycles within two breaths. 174 measurements were performed on 35 cardiovascular patients (5 each per patient) under general anesthesia and normocapnia.
we used the LoA calculation approach with correction for multiple measurements per subject.\textsuperscript{20}

Subsequently, the percentage error (PE) was calculated as $1.96 \times \text{SD of the bias of the tested method divided by the mean value of the tested and the reference method.}$\textsuperscript{21} To the best of our knowledge, an acceptable PE and acceptable agreement for ZFP, RAP and CPPe measurements have not yet been described. A precision (LoA) of less than 10\,mmHg for a CPPe of around 60\,mmHg could be clinically acceptable.

All statistical analyses were performed two-sided, and a P value of less than 0.05 was considered to be significant. Database sheets were created with MS Excel for Mac2011 (Microsoft Corp, USA). Statistical analysis and graphs were executed using Prism7.0 (GraphPad Software, USA).

RESULTS

Eligibility
Over a period of 17 months (02-08-1994 until 9-12-1995), 56 consecutive cardiovascular patients were scheduled for implantation (n=32) or replacement (n=24) of an ICD.\textsuperscript{13} Out of these 56 patients we excluded 19 patients for secondary analysis. One patient had an insufficient arterial pressure curve. 17 patients had a no or a very small acoustic window for transcranial Doppler sonography examinations, insufficient to use for pressure-flow velocity relationship analysis.

37 patients were eligible for secondary analysis. One patient had a labile arterial blood pressure, another patient showed atrial fibrillation. Both datasets were insufficient to use for pressure-flow velocity relationship analysis (two outliers). During the period of measurements, we obtained five data sets for each patient. In one patient, we could obtain only four measurements; one measurement has been excluded due to supraventricular extra-systolic beats. Secondary analysis was thus performed in 35 patients with 174 measurements (Figure 2: STROBE flow chart of participants).

Patients
The group consisted of 6 women and 29 men with a median age of 50 years (range, 15-73 years). The underlying cardiac diseases were: ischemic heart disease in 14 patients, primary electrical heart disease in 13 patients, and cardiomyopathy in 7 patients; 1 patient received an ICD for other reasons. Left ventricular pump function was normal in 14 patients, moderately reduced in 13 patients, and severely reduced in 7 patients. Arterial hypertension was treated in 8 patients. In 25 patients, the ICD was placed for the first time; in 10 patients, the ICD was replaced.
In 35 patients (5 measurements per patient, in total 174 observations) mean heart rate was 65 bpm with and a standard variation (SD) of 9 bpm (range from 44 to 79 bpm). Jugular venous oxygen saturation was 66 (9)% (range from 51 to 78 %). End tidal partial pressure of carbon dioxide was 33 (2) mmHg (range from 29 to 38 mmHg).

The values of effective cerebral perfusion pressure according to the reference method ($CPP_{LR}$) ranged from 32 to 84 mmHg (CV 21%), with a mean (SD) of 56 (13) mmHg. Descriptive analysis per subject of the five $CPP_{LR}$ measurements showed again a small variability (subject means: 35 mmHg, averaged $CPP_{LR}$ 56 mmHg, range 53–58 mmHg, average SD
2, CV 4%). The characteristics of variation and dispersion within the other CPPe, ZFP and RAP were small too. Hemodynamic data are summarized for all observations and averaged subject means in Table 2.

Bland-Altman analysis of CPPe, corrected for repeated measurements, revealed a small positive mean bias (SD) and good precision when comparing CPP_LR measurements with CPP LR3 (mean bias 1.2 (4.1) mmHg, LoA -6.8 to 9.3 mmHg, PE 15%) and with CPP CZO measurements (mean bias 1.8 (3.2) mmHg, LoA - 4.5 to 8.0 mmHg, PE 11%). Comparing CPP_LR measurements with CPP_BEL mean bias was moderately higher with more widespread LoA (mean bias 3.7 (5.6) mmHg, LoA -7.4 to 14.8 mmHg, PE 21%). Analysis of CPP SCH and the reference method showed less agreement with a negative mean bias of -10.6 (8.7) mmHg and wide-ranging LoA of -27.5 to 6.4 mmHg, PE 28%.

As expected, Bland Altman analysis of the five ZFP estimation methods showed similar results, because CPPe is defined as difference between MAP and ZFP while MAP values were unchanged ((1) ZFP LR versus ZFP LR3: mean bias -0.95 (4.88) mmHg, LoA -10.51 to 8.61 mmHg, PE 40%); (2) ZFP LR versus ZFP CZO: mean bias -1.90 (3.42) mmHg, LoA -8.60 to 4.81 mmHg, PE 27%; (3) ZFP LR versus ZFP BEL: mean bias -3.44 (5.81) mmHg, LoA -14.83 to 7.96 mmHg, PE 45%, (4) ZFP LR versus ZFP SCH: mean bias -11.14 (8.10) mmHg, LoA -4.73 to 27.01 mmHg, PE 88%).

Biases and LoA between RAP LR values (reference method) and RAP LR3, RAP CZO as well as the RAP BEL calculation methods have been small. However, the accuracy and precision for RAP SCH method was more wide-ranging ((1) RAP LR versus RAP LR3: mean bias 0.022 (0.115) mmHg, LoA -0.204 to 0.248 mmHg, PE 16%); (2) RAP LR versus RAP CZO: mean bias 0.051 (0.080) mmHg, LoA -0.108 to 0.206 mmHg, PE 12%; (3) RAP LR versus RAP BEL: mean bias 0.102 (0.147) mmHg, LoA -0.354 to 0.558 mmHg, PE 24%).

Figure 3. Comparison CPPe LR versus CPPe LR3, CPPe CZO, CPPe BEL, and CPPe SCH
The effective cerebral perfusion pressure (CPPe) estimated by four different methods, the 3-point intercept estimation (LR3, systolic/mean/diastolic), Czosnyka (CZO, 2 point systolic/diastolic), Belford (BEL, 2 point mean/diastolic), and Schmidt (SCH, 2 point systolic/diastolic) were validated against the reference method (LR, linear regression). One CPPe measurement corresponds to the cumulative analysis of 10-12 cardiac cycles within two breaths. 174 measurements were performed on 35 cardiovascular patients (5 each per patient) under general anesthesia and normocapnia. The figures explain differences (top rows: A-D), correlations (middle rows E-H) and agreement (bottom rows I-L) of the CPPe measurements. All statistical analyses were performed two-sided, and a P value of less than 0.05 was considered to be significant. All results are presented as mean (standard deviation).

Differences between the four tests methods of CPPe estimation (LR3, BEL, CZO SCH) and reference method (LR) were tested by ANOVA with correction for multiple comparison (Dunnett); § significant for all measurements; # significant for subject means.

Pearson r correlation coefficients between four CPPe values and the reference method (LR) of all observation were computed to assess their relationship. Agreement was analyzed by Bland-Altman plot analysis with correction for multiple measurements per subject (LL = lower limits of agreement, UL = upper limits of agreement, PE = percentage error).
The effective cerebral perfusion pressure (CPPe), the zero flow pressure (ZFP) and the resistance area product (RAP) estimated by four different methods, the 3-point intercept estimation (LR3, systolic/mean/diastolic), Czosnyka (CZO, 2-point: systolic/diastolic), Belford (BEL, 2-point: mean/diastolic), and Schmidt (SCH, 2-point: systolic/diastolic) were validated against the reference method (LR, linear regression). One CPPe measurement corresponds to the cumulative analysis of 10-12 cardiac cycles within two breaths. 174 measurements were performed on 35 cardiovascular patients (5 each per patient) under general anesthesia.
and normocapnia. All statistical analyses were performed two-sided, and a P value of less than 0.05 was considered to be significant. All results are presented as mean (standard deviation). Differences between the four tests methods of estimation (LR3, BEL, CZO SCH) and reference method (LR) were tested by ANOVA with correction for multiple comparison (Dunnett). CV = coefficient of variation (%), MAP = mean arterial pressure, MD = mean differences, Min = minimum, Max = maximum, OB = observations (results within all 174 measurements), SM = subject means (results within the 5 measurements per subject), SD = standard deviation, Vmca = blood flow velocity of the mean cerebral artery.

bias 0.097 (0.154) mmHg, LoA -0.204 to 0.398 mmHg, PE 22%; (4) RAP LR versus RAP SCH: mean bias -2.58 (0.244) mmHg, LoA -0.735 to 0.220 mmHg, PE 31%).

Pearson r correlation coefficients between four CPPe values and the reference method (LR) of all observation were computed to assess their relationship (n=174, 2-tailed). CP-P_{LR3}, CPP_{CZO} and CPP_{BEL} values showed a very strong positive correlation when compared to the reference method ((1) CPP_{LR3} r=0.95 ranging from 0.93-0.96, p<0.001, R^2 0.90; (2) CPP_{CZO} r=0.96 ranging from 0.95-0.97, p<0.001, R^2 0.93; (3) CPP_{BEL} r=0.89 ranging from 0.85-0.91, p<0.001, R^2 0.79). The correlation analysis between CPP_{LR} and CPP_{SCH} revealed a moderate positive relationship (r=0.69 ranging from 0.61-0.76, p<0.001 R^2 0.48).

Correlation analysis of ZFP values showed almost similar results. Correlation analysis of ZFP_{LR3} and ZFP_{CZO} showed a strong relationship versus the reference method ZFP_{LR}. The correlation of ZFP_{BEL} was moderate, and of ZFP_{SCH} was weak ((1) ZFP_{LR} versus ZFP_{LR3} r=0.87 ranging from 0.83-0.90, R^2 0.76; (2) ZFP_{LR} versus ZFP_{CZO} r=0.91 ranging from 0.88-0.93, R^2 0.83; (3) ZFP_{LR} versus ZFP_{BEL} r=0.69 ranging from 0.60-0.76, R^2 0.47; (4) ZFP_{LR} versus ZFP_{SCH} r=0.32 ranging from 0.18-0.45, R^2 0.10).

RAP computations showed fewer differences. They all showed very strong or strong positive correlations, when compared to the reference method ((1) RAP_{LR} versus RAP_{LR3} r=0.97 ranging from 0.96-0.98, R^2 0.94; (2) RAP_{LR} versus RAP_{CZO} r=0.99 ranging from 0.98-0.99, R^2 0.97; (3) RAP_{LR} versus RAP_{BEL} r=0.95 ranging from 0.94-0.96, R^2 0.91; (4) RAP_{LR} versus RAP_{SCH} r=0.87 ranging from 0.82-0.90, R^2 0.75).

The CPPe analysis of differences, correlation and agreement are graphically summarized in figure 3. For further figures regarding analyzes of ZFP and RAP, we refer to appendix 4 of the SDC.

DISCUSSION

In a secondary analysis, we validated four different estimation methods of effective cerebral perfusion pressure, zero-flow pressure, and resistance area product against a reference method in 35 cardiovascular patients under steady state conditions during fentanyl-midazolam anesthesia and normocapnia. The study was performed to identify differences, correlation and agreement of these methods.
The most prominent results of the study are:

i. CPPe, ZFP and RAP measurements based on LR3 and CZO calculation methods showed small mean differences, good agreement, low percentage errors, and an excellent correlation when compared to LR reference method.

ii. Agreement and correlation of CPPe ZFP and RAP was moderate for the BEL method, and weak for the SCH method.

iii. It is possible and safe enough to estimate CPPe, ZFP and RAP with simpler approaches like the CZO method or the LR3 method when compared to the LR method.

iv. ZFP estimation by intercept calculation based on a 3 point-extrapolation (systolic/mean/diastolic LR3) does not seem to be more advantageous than by a 2 point-extrapolation (systolic/diastolic CZO).

v. The RAP seems to be rather unaffected by the method of estimation

Mainly, there are two methods for determining the zero-flow pressure using transcranial Doppler sonography. The most fundamental method estimates the ZFP by regression analysis of the pressure-flow velocity plot derived from complete pressure and flow velocity tracings. The second group of ZFP estimation methods were based on the slope-intercept-form (figure 1). These methods are much simpler to implement than regression analysis of digitized arterial pressure and Vmca curves and can thus easily be used for bedside assessments. Furthermore, a correction of the time delay between arterial blood pressure and velocity of the mean cerebral artery is not necessary. For further mathematical explanation we refer to the appendix 1 of this report in the SDC section.

Linear pressure-flow velocity relationships in an elastic vessel based on systolic/diastolic (CZO), mean/diastolic values (BEL) or all 3 values (LR3) are based on the assumption that the vessel’s resistance behavior remains constant within the whole cardiac circle and throughout its diameter. Here, our study data demonstrated that ZFP estimation by intercept calculation based on a 3 point-extrapolation (systolic/mean/diastolic LR3) seems to be a little more advantageous when compared by a 2 point-extrapolation based on systolic/diastolic data (CZO). Using only mean and diastolic values and thus skipping the systolic values of the ABP and Vmca, on the other part, will shorten the distance of the velocity plot, which might weaken the estimation of ZFP and RAP. This could be a reason for the wider limits of agreement and thus moderate agreement of the BEL method. Thus, the systolic part of the pressure and velocity curves seems to be important for the accuracy of the ZFP calculation.

To the best of our knowledge, there are only a few (method comparison) on ZFP, CPPe or RAP studies using pressure-flow velocity analysis in patients without intracranial pathologies. They are difficult to compare with our results because they unfortunately differ in study design, study population, statistical analysis and used CPPe, ZFP and RAP estimation methods. Recently Baker et al. compared ZFP obtained by optical diffuse correlation spectroscopy (DCS) measurements with ZFP measurements based on Dop-
pler sonographic pressure-velocity plots in 18 awake healthy adults laying supine at rest, which showed good agreement.

Panerai and colleagues\textsuperscript{23} compared seven different ZFP/RAP estimation methods to assess their robustness and reproducibility in ten healthy subjects. Their data showed a high incidence of negative ZFP, which could be related on the calculation method. The authors suggested that the classical use of LR of the instantaneous pressure-flow velocity relationship of the entire cardiac cycle should not be used for static applications due to poor reproducibility and high occurrence of negative values of ZFP. They recommended further that first harmonic fitting (H1) and 2-point mean/diastolic values (2Pm) perform better than LR analysis and should be used for the estimation of ZFP and RAP for both static and dynamic applications. In our study, we did not find any negative ZFP values in our study. Negative ZFP values have been described during hyperemia, vasospasm, or with artificially elevated diastolic Vmca.\textsuperscript{24-26} However, at present, there is no physiologically relevant interpretation for these findings and they are commonly viewed as methodological limitations. Interestingly, the ZFP(LR) values from Baker et al.\textsuperscript{22} and Panerai et al.\textsuperscript{23} were about 10 mmHg lower compared to our data, which perhaps could be related to differences in study population, non-invasive arterial pressure measurements (Finapress\textsuperscript{®}), general anesthesia and artificial ventilation.

Some methodological aspects of our study have to be considered. A major problem in defining pressure-flow relationships in living vascular beds is that autoregulation can alter bed characteristics during the period of observation. In situations of CPP below 40 mmHg, which corresponds to a maximal vasodilation of small pial arteries, autoregulation is progressively exhausted. Here the shape of the pressure-flow plot could be nonlinear. Most methods proposed for estimation of ZFP can only represent the linear range of the pressure-flow (or velocity) relationship. Thus, “in vivo”-estimation of ZFP by linear pressure-flow(velocity) plot analysis can give only “presumptive” or “apparent” values rather than “true” values.\textsuperscript{3} To define the ZFP\textsubscript{LR} as reference method is thus still debatable.

Analysis of the long diastolic fall in pressure and flow after cessation of cardiac function was the “classic” method the zero-flow pressure estimation of the coronary circulation in dogs\textsuperscript{27} and later in humans. Aaslid et al. transferred this approach for the cerebral perfusion with transcranial Doppler sonography.\textsuperscript{28} The ZFP based on long diastoles with an duration of was about 6 mmHg lower when compared to the ZFP\textsubscript{LR} method. In order to verify the ZFP\textsubscript{LR} method as suitable reference method in our patients we additionally measured ZFP based on the first long diastole during ICD testing-induced cardiac arrest (ZFP\textsubscript{LD} and RAP\textsubscript{LD}). We performed pressure-velocity plots analyses at different durations of the long diastole (1s-10s, and the whole long diastole). Our additional analysis of ZFP based on long diastoles demonstrated that pressure velocity relationships with a duration of 4s or longer showed a less linear relationship. Subsequently, the extrapolated ZFP\textsubscript{LD} (4s or longer) showed lower values when compared to ZFP\textsubscript{LR}. The use ZFP\textsubscript{LR} as
reference method seems to be safe and a good alternative to ZFP_{LD} measurements. Based on our results we recommend a standard diastolic time of 3s when ZFP is assessed by long diastoles. For further information we refer to Appendix 3 of the SDC section.

CONCLUSIONS

We validated four different estimation methods of CPPe pressure, ZFP, and RAP against a reference method in 35 cardiovascular patients under steady state conditions during fentanyl-midazolam anesthesia and normocapnia. CPPe ZFP and RAP measurements based on LR3 and CZO calculation methods showed small mean differences, good agreement, low percentage errors, and an excellent correlation when compared to references method. Agreement and correlation were moderate for the BEL method, and unsatisfying for the SCH method. The RAP seems to be rather unaffected by the method of estimation. Our study results demonstrate that it is possible and safe enough to estimate CPPe, ZFP and RAP with simpler approaches like the CZO method or the LR3 method.

REFERENCES


APPENDIX 1 - ZERO FLOW PRESSURE (ZFP) ESTIMATION

Background

Models defining cerebrovascular tone

The classic concept defining cerebrovascular tone is cerebral vascular resistance (CBF = CPP / CVR). It assumes that perfusion pressure and flow are linearly and proportionally related. When calculating the CPP, the MAP has been used as effective upstream pressure (EUP) and the intracranial pressure (ICP) as effective downstream pressure (EDP) of the cerebral circulation, because of a Starling resistor phenomenon located at the level of cerebral veins (classical model CPpi = MAP - ICP). Patients without cerebrovascular disease are expected to have a normal ICP between 7-15 mmHg in supine position. When ICP is elevated by i.e. intracranial bleeding or hydrocephalus, CPpi will decrease unless reflex arterial hypertension occurs. If MAP increases less than ICP beyond this point, CBF will decrease.

However, the “classical model” has limitations. Using solely the ICP as effective downstream pressure (EDP) of the cerebral circulation, would neglect vascular tone properties of the vessels. Arteriolar wall tension arises from a combination of the stretched elastic components of the vessel wall and active contraction of vascular smooth muscle. Subsequently, a lot of studies of organ perfusion of the i.e. lung, myocardium, liver, muscle, skin, and the brain, have shown, that the EDP is more determined by a critical closing pressure located at arteriolar level.

The origin of the term critical closing pressure (CrCP) or later zero flow pressure (ZFP) is often attributed to Alan Burton. He suggested the use of Laplace’s law to explain the influence of active wall tension on collapsible vessels. As perfusion pressure is reduced, there will be a point where transmural pressure will not be sufficient to counteract the active tension imposed by the smooth muscle layer. Then, the vessel will collapse. At this point blood flow will cease and the corresponding perfusion pressure is the CrCP or ZFP.

When dynamic measurement techniques are used, such as electromagnetic flowmetry or ultrasound Doppler, the limitations of the classical CPpi concept become evident. Dynamically, flow may stop at pressure levels significantly higher than zero. The arterial blood pressure (ABP) level at which flow stops is defined as the CrCP or zero-flow pressure (ZFP). Graphically, this value is associated with the pressure-axis intercept of a linear regression plot of the blood flow (or velocity) as a function of ABP. The ZFP represents vasomotor tone while its slope represents the value of vascular bed resistance. Hence the diameter of the resistance vessel is the relationship between vasomotor tone and vessel diameter. That means that flow is linearly (but not proportionally) related to pressure and that it can be regulated by changes in both, ZFP (the x intercept) and slope. The pressure-flow relation is mainly a function of the periph-
eral resistance. Thus, the driving pressure for the flow through arterioles is, under many conditions, not the difference between arterial (inflow) pressure and venous (outflow) pressure, but rather the difference between arterial pressure and ZFP (alternative model CPPe = MAP - ZFP).

In a former investigation, we suggested the hypothesis of two Starling resistors in a series connection, one (proximal) at the precapillary level of cerebral resistance vessels (CrCPart) and a second (distal) at the level of collapsible cerebral veins (CrCPven). The effective downstream pressure of the cerebral circulation may be determined by CrCPart, CrCPven (i.e. ICP), or jugular venous pressure, depending on which one is the highest (figure S1). In the light of this concept, some researchers have created the term “effective cerebral perfusion pressure” to refer to the difference between MAP and ZFP, by considering the tone of the vessels.

Another assumption, sometimes found in the literature, is that ZFP can be used as a substitute for ICP. This is possible only in situations where active wall tension remains constant, which seems to be unlikely unless in patients with impaired autoregulation. The difference between ZFP and ICP is explained by the tone of the small vessels, i.e. wall tension.

![Cerebral vascular waterfall of the circulation](image)

**Figure S1: Cerebral vascular waterfall of the circulation**
Hypothesis of two Starling resistors in a series connection, one (proximal) at the precapillary level of cerebral resistance vessels (CrCPart) and a second (distal) at the level of collapsible cerebral veins (CrCPven). The effective downstream pressure of the cerebral circulation may be determined by CrCPart, CrCPven (i.e. ICP), or jugular venous pressure, depending on which one is the highest.

**Resistance area product (RAP)**
The slope of the pressure-flow velocity plot is inversely related to arterial resistance. Many research groups used the inverse of the slope of the pressure-velocity plot to de-
termine cerebral vascular resistance by transcranial Doppler sonography measurements. In 1988, David Evans et al. introduced the Doppler sonographic index, the resistance area product (RAP). Derived from Hagen-Poiseuille law, the slope of a pressure-velocity relationship could also be equated to the product of velocity and the cross-sectional area of the vessel at the site of insonation. This definition is a misnomer, because it is not possible to measure the diameter of the artery by transcranial Doppler sonography at the point of insonation. Nevertheless, the resistance area product (RAP) is an important estimate of vascular resistance, because it takes the cerebral zero flow pressure into account. The driving pressure of the brain is the effective perfusion pressure rather mean arterial pressure, as mentioned above.

Simplified, the RAP can also be calculated as a quotient of effective cerebral perfusion pressure and mean MCA velocity (\(\text{RAP} = \text{CPPe} / \text{mean Vmca}\)). This definition or calculation of the RAP however, is not consistently used. 18-20

**Methods for non-invasive assessment of the EDP of the cerebral perfusion**
Several methods for non-invasive assessment of the effective downstream pressure of the cerebral perfusion have been described by using: sensing tympanic membrane displacement 21, skull vibrations 22, otoacoustic emissions 23, magnetic resonance imaging to estimate intracranial compliance 24, brain tissue resonance 25, transcranial time of flight 26, recordings of visual evoked potentials 27, optic nerve sheath diameter assessment 28, venous ophthalmodynamometry 29, and ultrasound-guided eyeball compression 30, and the optical diffuse correlation spectroscopy (DCS) 31. Most these techniques are more appropriate for one-point assessment of instant value of EDP/ CrCP/ ZFP/ ICP and subsequently CPP rather than continuous monitoring. 32

Transcranial Doppler sonography (TCD) allows non-invasive, continuous measurements of the flow velocity of the middle cerebral artery (Vmca), which represents with 80% the major portion of global cerebral blood flow. In a number of previous studies, relative changes in CBF were reflected by changes in Vmca in a proportional manner, suggesting that MCA diameter remains constant. 33 A good correlation between changes in Vmca and changes in CBF has been found during carotid endarterectomy. 34

Since the introduction of TCD, a number of methods have been developed to assess cerebral ZFP by pressure-(flow)velocity relationship analysis. 15 16 35-43

Other research groups tried to develop alternative mathematical models for EDP / CrCP / ZFP calculations including values of ICP, ABP, autoregulation and intracranial compliance. 41 44-46 However, these models are rather complex and therefore less suitable for the clinical setting.
Linear relationship between blood flow (velocity) and blood pressure

It has been proven that in vivo dynamic pressure-flow relationships are straight lines for many vascular beds even for the cerebral vessels.\textsuperscript{3,10-12,47-50} The most common approach to estimate the cerebral CrCP or ZFP is thus to perform a linear regression on continuous measurements of ABP and Vmca taken from a single cardiac cycle. These waveforms oscillate between the diastolic and systolic values. Therefore, the linear regression line uses only the values represented by circles, corresponding to i.e. $80 > Pa < 120$ mmHg.

A lot of animal studies support the linear relationship between flow and pressure even for situations of very low arterial blood pressures (ABP). Walter Ehrlich evaluated instantaneous femoral artery pressure-flow relations during cardiac arrest in anesthetized dogs. He could demonstrate, that in all pressure-flow plots obtained under conditions of normal or elevated venous pressure, either with or without alpha- blockade, the pressure-flow relations were linear, and the zero-flow intercept on the pressure axis was reached in less than 3 seconds after the onset of cardiac arrest, which was markedly higher than the simultaneous venous pressure.\textsuperscript{49} Richard C. Dewey evaluated cerebral pressure-flow relationships in anesthetized monkeys. Changes from arterial hypertension to hypotension did not affect the linearity of the measured pressure-flow plots of carotid artery.\textsuperscript{10}

The linear pressure-flow(velocity) plot model may has limitations. Cardiac arrhythmia or function tests with implantable cardioverter defibrillators devices are situations with very long diastolic phases and low diastolic pressures.

This approach has been used to estimate and to confirm a “true” ZFP in the coronary circulation, systemic and even cerebral circulation in humans.\textsuperscript{5,17,51} Analysis of prolonged diastolic pressure and flow curves however showed non-linear parts of the pressure-flow curve. Some “non-linear” factors (i.e. cerebral venous circulation, vascular capacitance, elasticity of the insonated part of the vessel, rheological properties of the blood, pulse rate, radius-dependent active wall tension etc.) of the cerebrovascular tree regulation are less represented in this model.\textsuperscript{52-54} Comparable factors may also disturb the arterial pressure waveform like changes in cardiac output or stroke volume. When using normal ranges of systolic and diastolic of arterial blood pressures and Vmca, any extrapolation to calculate ZFP is likely to vary significantly given the wide ranges of the normal values within these variables. This may explain the wide ranges of ZFP reported in our study as well in previous studies (figure S2).\textsuperscript{55}

However, the linearity of pressure-flow plots seems to be stable within normal physiologic values. There are some experimental data about vascular compliance available, predominantly for the coronary perfusion.\textsuperscript{8,56-60} It has been shown that the compliance of the cerebral vessels, however, seems to be lower than other vascular beds.\textsuperscript{61} Thus, the influence of capacitance effects under “in vivo” conditions appears to be small. Variations of the partial pressure of carbon dioxide, a strong vasodilator, could affect vascular ca-
pacitance of the middle cerebral artery, which in turn may impair the accuracy of linear extrapolation of ZFP. In a prior investigation we could demonstrate that the linearity of pressure-flow velocity relationships have not been influenced by PaCO2 and its potential effects on compliance are thus assumed to be of minor methodological importance.62

Figure S2: Possible bias in ZFP and rAP estimation by linear regression

This figure explains the possible wide ranges of estimated cerebral zero flow pressures (ZFP) in a normal population. The dot boxes show all possible combinations of the systolic (upper box) and the diastolic (lower box) values of blood pressure (ABP) and middle cerebral artery flow velocity (Vmca) in a normal population (ranges of ABP, systolic = 145-100 mmHg, diastolic = 90-50 mmHg; ranges of the Vmca, systolic = 90-80 cm/s, diastolic 46-33 cm/s).63 The solid line arrows point to the extrapolated ZFP, if the values of blood pressure and flow velocity are close to the average. The dotted line arrows point to the possible range of extrapolated ZFP if the values of blood pressure or flow velocity (and the pulsatility of their waveforms) approach the limits of their normal range (modified from Athanassiou et al. 2005.55

“Apparent” versus “true” ZFP

Most methods proposed for estimation of ZFP can only represent the linear range of the pressure-flow (or velocity) relationship. As a consequence, only estimates of a “presumptive” or “apparent” ZFP can be obtained, and these tend to be significantly higher than the “true” ZFP.5

In the past, many research groups have tried to identify a “true” ZFP. In animal studies, it is possible to isolate brain circulation from other systems (e.g., by ligating the carotid artery) and thus to perform repeated measurements of blood pressure, blood flow, and flow velocity under different conditions. Later, analyzes of the long diastolic fall in pressure and flow after cessation of cardiac function (i.e. induced by ventricular fibrillation) was used for zero-flow pressure estimation of the coronary circulation in dogs and later in humans.56 64

Eva Kottenberg-Assenmacher and her group estimated the ZFP/CrCP of the systemic circulation with different mathematical models during internal cardiac defibrillator

Erasmus University Rotterdam
testing in 10 patients under general anesthesia. Interestingly, they could demonstrate that arterial ZFP based on heart beating data, was substantially higher than ZFP values found during cardiac arrest. They explained the discrepancy between heart beating and cardiac arrest values by a leak in the waterfall. In an intact circulation the height of the waterfall will be intact as long as the volume supply exceeds the volume loss. However, when supply becomes less than the volume loss, as is the case during a cardiac arrest, the drain of arterial blood through those vascular waterfalls with lower local ZFP values will result in a reduction of measured ZFP. They could demonstrate that the ZFP measured or calculated by curve fitting after prolonged periods of circulatory arrest was lower than that with a circulatory arrest of shorter duration.

*Rune Aaslid et al.* transferred this approach for the cerebral perfusion with transcranial Doppler sonography. Their study aim was to validate estimates of ZFP based on regular heartbeat data (linear regression method) against ZFP estimates based on long diastole determinations (about 3 seconds) after induced ventricular fibrillation (VF) during internal cardiac defibrillator testing in 13 patients under general anesthesia. The ZFP based on long diastoles was about 6 mmHg lower when compared to the ZFP<sub>LR</sub> method.

Obtaining ZFP from long diastoles induced by ventricular fibrillation has also some disadvantages:

- Any circulatory arrest is an abnormal state likely to change vascular properties even in the brain. There are no oxygen stores in the brain in contrast to myoglobin, which stores oxygen in the muscle. For example, ischemia during circulatory arrest might induce peripheral arteriolar vasodilation within seconds, which subsequently will result in a too-low ZFP.
- Although the venous compliance is certainly greater than the compliance of the arterial vascular tree, the further fall in arterial pressure after circulatory arrest may be related to a reduction in vascular tone rather than drainage of the remaining arterial flow.
- Conversely, the induction of ventricular fibrillation with a quick reduction in arterial pressure could also induce intense efferent neural sympathetic outflow and consequently arteriolar vasoconstriction, giving rise to a too-high ZFP.
- Furthermore, aortic valve flutter has been described using transesophageal echocardiography during ventricular fibrillation. In this case, aortic valve regurgitation in addition to peripheral arterial flow runoff could speed aortic pressure decay and shorten the decay time constant, and consequently underestimate calculated ZFP.

However, measurements of a “true” ZFP might be an elusive concept. A major problem in defining pressure-flow relationships in living vascular beds is that autoregulation can alter bed characteristics during the period of observation. Any circulatory arrest is an abnormal state likely to change vascular properties even in the brain. In situations of CPPs below 40 mmHg, which corresponds to a maximal vasodilation of small pial arter-
ies, autoregulation is progressively exhausted. Here the shape of the pressure-flow plot could be non-linear.

It is thus debatable whether fibrillation-induced circulatory arrest allowed measurements of the ZFP that prevails physiologically.31

Mathematical principle of the ZFP calculation

Two respiratory cycles of each measurement period are randomly selected and extrapolated ZFP data of all heartbeats within these respiratory cycles are averaged for further analysis of ZFP, RAP and CPPe. In algebraic geometry, a straight line in an axis system is described by the linear function. In two dimensions, the equation for non-vertical lines is often given in the slope-intercept form (see figure S8):

\[ y = a \cdot x + b \]  \hspace{1cm} (1)

In figure S3 \( a \) is the slope and \( b \) is the y-intercept of the line. If \( P1 = (x1, y1) \) and \( P2 = (x2, y2) \) are points on the non-vertical line \( L \) then the equation of the line can be found using the point-slope-formula or the two-point form.

\[ y - y1 = a \cdot (x - x1) \] \hspace{1cm} (2a)

\[ y - y1 = \frac{(x2 - x1)}{(x2 - x1)}\cdot(x - x1) \] \hspace{1cm} (2b)

To calculate the line \( L \) intercept, compare equation (1) to:
The intersection of line L with the x axis [(X, 0), c in figure S6] can be found by equating (1) equal to 0 and solving:

\[ 0 = a \cdot x + b \]  

(4a)

\[ -b = a \cdot x \]  

(4b)

\[ a = \frac{b}{x} \]  

(4c)

With this background, it is possible to redirect a graph as in figure S4 to a formula, as done by Belfort et al, to calculate the ZFP.66

For a pressure-flow plot analysis the diastolic and mean flow velocity (Vd, Vm) have to be put on the y-axis as the independent variable. The diastolic and mean arterial blood pressure (DAP, MAP) are put on the x-axis as dependent variable. Thus, the linear graph of a pressure-flow(velocity)-plot could be defined regarding to equation (1) as:

\[ V_m = a \cdot MAP + b \]  

(5a)

Then the y-intercept would be:
The slope could be defined as:

\[ a = \frac{(V_m - V_d)}{(MAP - DAP)} \]  

(5c)

To calculate the intercept a zero velocity (P0 = ZFP) we have to rearrange the forms 5a-c with the following steps:

\[ 0 = a \cdot P_0 + b \]  

(6a)

\[ -b = a \cdot P_0 \]  

(6b)

\[ P_0 = -\frac{b}{a} \]  

(6c)

\[ ZFP = -b \cdot \frac{(MAP - DAP)}{(V_m - V_d)} \]  

(6d)

\[ ZFP = -(V_m - a \cdot MAP) \cdot \frac{(MAP - DAP)}{(V_m - V_d)} \]  

(6e)

\[ ZFP = -V_m \cdot \frac{(MAP - DAP)}{(V_m - V_d)} + a \cdot MAP \cdot \frac{(MAP - DAP)}{(V_m - V_d)} \]  

(6f)

\[ ZFP = -V_m \cdot \frac{(MAP - DAP)}{(V_m - V_d)} + MAP \cdot \frac{(V_m - V_d)}{(MAP - DAP)} \cdot \frac{(MAP - DAP)}{(V_m - V_d)} \]  

(6g)

\[ ZFP = -V_m \cdot \frac{(MAP - DAP)}{(V_m - V_d)} + MAP \cdot 1 \]  

(6h)

\[ ZFP = MAP - V_m \cdot \frac{(MAP - DAP)}{(V_m - V_d)} \]  

(6i)

Then CPPe can be calculated by:

\[ CPPe = MAP - ZFP \]  

(7a)

Or by:

\[ CPPe = V_m \cdot \frac{(MAP - DAP)}{(V_m - V_d)} \]  

(7b)
APPENDIX 2 STUDY PROTOCOL CPPE, ZFP, AND RAP ANALYSIS

“In vivo” estimation of ZFP by linear pressure-flow(velocity) plot analysis

Arterial blood pressure measurements
Arterial blood pressure (ABP) was monitored via a cannula placed in the radial artery with the transducer positioned at the level of the base of the skull/ear.

Blood flow velocity measurements of the middle cerebral artery
Blood flow velocity of the middle cerebral artery (Vmca) was measured via the temporal window with a commercially available 2 MHz transcranial Doppler device (DWL multidop-X device with two 2-MHz pulsed Doppler probes, DWL TCD 7 software, DWL Elektronische Systeme GmbH, Sipplingen, Germany).

Figure S5: TCD measurements of the mean cerebral artery
(with permission from Mr. Arnoud Steutel)

The ultrasound transducer was placed over the temporal area just above the zygomatic arch and in front of the tragus of the ear. After identification of the middle cerebral artery, the depth of insonation was adjusted in 1-2 mm increments to obtain signals from the proximal segment (M1) of the MCA. Accurate care was taken to ensure a stable position of the ultrasound probe during the study period. The ultrasound probe was therefore fixed using a monitoring probe holder attached to the patient’s head (see figure S5). Doppler ultrasound variables such as depth, gain, sample volume, and power of the
ultrasound beam were unchanged during the measurement procedure. Determinations of flow velocity were based on envelope curves of maximum intravascular velocity.

Data recordings and storage
Simultaneous data recordings of ABP and Vmca envelope curves were acquired over periods of 2-5 minutes. At each measurement, simultaneous data recordings of Vmca, arterial blood pressure (ABP, mmHg), expiratory concentration carbon dioxide concentration (PeCO₂, mmHg), and jugular venous bulb oxygen saturation (SjO₂, %) were acquired and stored on a microcomputer via analogue/digital converters. These files are still available (DWL software format).

The analyses of the ZFP, CPPe and RAP have been performed after the study period. For the secondary analyses, we looked at the period after induction of anesthesia and before ICD-testing. We extracted five consecutive data sets of 10-12 seconds (two breaths) each within 3 min from the raw data files of the primary investigation. These data sets were exported (ASCII format, see figure S6). Recordings that contained ABP and Vmca measurements during ICD tests (ventricular fibrillation) were not used for this secondary analysis.

Figure S6: Exported TCD data-set (ASCII format)
**CPPe / ZFP analysis**

For the CPPe / ZFP analysis we used dedicated custom-developed software (LabView®, USA, written by Dr. E.G. Mik, Erasmus MC).

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**Figure S7: CPPe estimation software based on LabView® (window 1)**

The following steps were performed (Figures S7 and S8):

1. **Import**
   An ASCII files was imported.

2. **File detail string**
   The imported file was checked and verified using the preview window.

3. **Sample rate**
   The used TCD sample rate was noted.

4. **Inspection**
   Vmca and ABP waveforms were visually inspected for artifacts or arrhythmias, which could distort further analysis.

5. **Determine the start of the measurement period (Offset)**
   The start of measurement period within the whole data tracings was chosen.

6. **Determine the duration of the measuring period (Zoom function)**
   A data set of 10-12 seconds (two breaths, 9 beats) was selected.

7. **Synchronization (Offset fine adjustment)**
   ABP and Vmca waveforms were synchronized: To compensate for the time delay between ABP and Vmca at the radial and middle cerebral artery, respectively, flow velocity curves have to be shifted.

8. **Visual control pressure flow plot**
   The correct compensation of time delay was calculated by iterative regression analysis until hysteresis of ABP / Vmca plots completely disappeared. A correlation
coefficient >0.94 was used as a cut-off value to select heartbeats with adequate signal quality.

Figure S8: CPPe estimation software based on LabView® (window 2)

9. Visual control of the selected measurement period
   On the following screen (window 2) measurement tracings of ABP and Vmca and their synchronization were visually controlled.

10. Verification of ABP and Vmca values
    Systolic, diastolic, and mean values of ABP and Vmca of the selected measurement period were averaged.

11. CPPe, ZFP_{LR} estimation by linear regression of ABP and Vmca
    The pressure axis intercepts of the ABP / Vmca plot of the selected measurement period represent the ZFP_{LR} of the cerebral circulation (step 8 and 9). CPPe was calculated by MAP-ZFP_{LR}.

12. CPPe, ZFP_{CZO} calculation by Czosnyka formula
    ZFP_{CZO} was calculated by a formula reported by Czosnyka et al. Here, averaged systolic and diastolic values of ABP and Vmca (step 10) were used. CPPe was calculated by MAP-ZFP_{CZO}.

\[
ZFP_{CZO} = \frac{SAP - Vs \cdot (SAP - DAP)}{(Vs - Vd)}
\]

13. CPPe, ZFP_{SCH} calculation by Schmidt formula
    ZFP_{SCH} was calculated by a formula reported by Schmidt et al. Here, averaged mean and diastolic values of ABP and Vmca (step 10) were used. CPPe was calculated by MAP-ZFP_{SCH}. 


$$ZFP_{SCF} = \text{MAP} - \left( \text{MAP} \cdot \frac{Vd}{Vm} + 14 \right)$$

14. **CPPe, ZFP<sub>BEL</sub> calculation by Belfort formula**

$ZFP_{BEL}$ was calculated by a formula reported by Belfort et al. Here, averaged mean and diastolic values of ABP and Vmca (step 10) were used. $ZFP$ was calculated by $\text{MAP} - \text{CPP}_{BEL}$.

$$ZFP_{BEL} = \text{MAP} - Vm \cdot \frac{(\text{MAP} - \text{DAP})}{(Vm - Vd)}$$

15. **Calculations of resistance index (RI) and pulsatility index (PI)**

The Resistance index (RI) was calculated by a formula reported by Pourcelot et al. The Pulsatility index (PI) was calculated by a formula reported by Gosling et al. Here, averaged systolic, mean and diastolic values of Vmca (step 10) were used. These results were not used in the current manuscript.

$$\text{PI} = \frac{Vs - Vd}{Vm}$$

$$\text{RI} = \frac{Vs - Vd}{Vs}$$

16. **Export of measured data**

The results of the analysis were saved and exported as text file (*.txt).

**Additional calculations**

All measurements files were imported in a spreadsheet for further calculation and analysis (Excel 2011®, Microsoft Cooperation, USA).

**CPPe, ZFP<sub>LR3</sub> calculation**

$ZFP_{LR3}$ was calculated by a 3-point intercept estimation (LR3) based on the averaged dataset of systolic, mean and diastolic ABP and Vmca values of each measurement period (from step 10).

The Excel "Intercept function" calculates the intercept (the value at the intersection of the y axis) of the linear regression line through a supplied set of x- and y- values (URL: https://support.office.com/en-US/article/INTERCEPT-function-2A9B74E2-9D47-4772-B663-3BCA70BF63EF)

**RAP calculations**

The RAP<sub>LR3</sub> was defined as the inverse slope of the linear regression line of ABP and Vmca values. Thus, the RAP<sub>Lr</sub>, RAP<sub>LR3</sub>, RAP<sub>BEL</sub>, RAP<sub>CO</sub> and RAP<sub>SCH</sub> were calculated by the quotient of the respective CPPe and mean Vmca. (\(\text{RAP} = (\text{MAP} - \text{ZPF}) / \text{mean Vmca}\)).
APPENDIX 3 PRESSURE-VELOCITY RELATIONSHIP ANALYSIS OF LONG DIASTOLES

**Background:**
In order to verify the ZFP\textsubscript{LR} method as suitable reference method in our patients we additionally measured ZFP based on the first long diastole during ICD testing-induced cardiac arrest (ZFP\textsubscript{LD} and RAP\textsubscript{LD}).

**Methods:**
Our own ZFP analysis software (made with LabView \textsuperscript{®}) could not fit the job. This ZFP analysis of long diastoles has been performed offline with EXCEL and PRISM (see Figure S1). We performed pressure-velocity plots analyses at different durations of the long diastole in 10 patients (1s - 10s, and the whole long diastole).

Normal distribution of data was assessed both visually by inspection of histograms and with the D’Agostino-Pearson omnibus K2 method. Results were presented as mean values (standard deviation, SD) unless otherwise stated.

Values of ZFP\textsubscript{LD} and RAP\textsubscript{LD} estimations (1s -10s and the whole LD) were compared to the reference method (LR) by paired tests and ANOVA for repeated measurements with correction for multiple comparison (Dunnett).

**Results:**
The first long diastole of patient 4 and patient 13 showed a lot of artifacts regarding the TCD signal. They have been excluded from further analysis of long diastoles.

*Figure S9* showed the measurement phase the first long diastole of patient 8. *Figure S10* showed the ZFP analysis at 1s - 10s, and for the whole long diastole.

*Table S1/S2 and figure S11/S12* show the values of ZFP\textsubscript{LD} / RAP\textsubscript{LD} analyses of the first long diastole after induction of ventricular fibrillation of 10 patients.

In our study ZFP\textsubscript{LD} (3s) was 3 mmHg lower (MD 2.8 (4.), 95% CI -2.7 to 8.3 mmHg, P 0.51) and ZFP\textsubscript{LD} (4s) was 6 mmHg lower when compared to ZFP\textsubscript{LR} (MD 5.5 (4.0), 95% CI 1.1 to 9.9, P = 0.016). The analysis of long diastoles of our patients with a duration >4 seconds showed a decrease in ZFP\textsubscript{LD} when compared to the ZFP\textsubscript{LR} based on instantaneous pressure-velocity relationships. The longer the duration of the measurement phase, the lower the ZFP\textsubscript{LD} was. The RAP showed an increase with the similar characteristics.

**Conclusions**
Our additional analysis of ZFP\textsubscript{LD} demonstrated that pressure velocity relationships with a duration of 4 s or longer showed a less linear relationship. Subsequently, the extrapolated ZFP\textsubscript{LD} (4s or longer) showed lower values when compared to ZFP\textsubscript{LR}.
The use $ZFP_{LR}$ as reference method seems to be safe and a good alternative to $ZFP_{LD}$ (2-3s) measurements.

Figure S9: ZFP and RAP analyses based on long diastoles (patient 8).
Values of ZFP and RAP by pressure-velocity plots analyses at different durations of the long diastole (1s - 10s, and the whole long diastole).
Figure S10: ZFP and RAP analyses based on long diastoles (patient 8).

Values of ZFP and RAP by pressure-velocity plots analyses at different durations of the long diastole (1 s, 2s, 3s, 5s, 10s, and the whole long diastole).
Figure S11: ZFP analyses based on long diastoles.

Table S1: ZFP analyses based on long diastoles.

<table>
<thead>
<tr>
<th>PAT</th>
<th>ZFP-LR</th>
<th>ZFP-LD1s</th>
<th>ZFP-LD2s</th>
<th>ZFP-LD3s</th>
<th>ZFP-LD4s</th>
<th>ZFP-LD5s</th>
<th>ZFP-LD10s</th>
<th>ZFP-LD-all</th>
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<td>14.8</td>
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<td>19.2</td>
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<td>25.9</td>
<td>23.4</td>
<td>20.8</td>
<td>18.1*</td>
<td>16.0*</td>
<td>11.2*</td>
<td>9.7*</td>
</tr>
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<td>(SD)</td>
<td>(5.8)</td>
<td>(4.3)</td>
<td>(3.8)</td>
<td>(3.3)</td>
<td>(3.3)</td>
<td>(3.2)</td>
<td>(4.8)</td>
<td>(4.8)</td>
</tr>
</tbody>
</table>

**ZFP-LR** = values of zero flow pressure based on 5 “in vivo” instantaneous beat to beat pressure-velocity plot analyses within 2 breaths,  
**ZFP LD1=** values of ZFP pressure-velocity plots analyses at different durations of the long diastole (1 s, 2s, 3s, 5s, 10s, and the whole long diastole). * significant different when compared to ZFP LR (two tailed paired t test and RM ANOVA, P< 0.05)
Table S2: RAP analyses based on long diastoles.

<table>
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<th>RAP-LR</th>
<th>RAP LD1s</th>
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<th>RAP LD3s</th>
<th>RAP LD4s</th>
<th>RAP LD5s</th>
<th>RAP LD10s</th>
<th>RAP LD-all</th>
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<td>1.58*</td>
<td>1.64*</td>
<td>1.83*</td>
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<td>(SD)</td>
<td>(0.40)</td>
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<td>(0.32)</td>
<td>(0.38)</td>
<td>(0.42)</td>
<td>(0.43)</td>
<td>(0.51)</td>
<td>(0.57)</td>
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</table>

RAP-LR = values of zero flow pressure based on “in vivo” instantaneous beat to beat pressure-velocity plot analyses within 2 breaths,
ZFP LD1= values of ZFP pressure-velocity plots analyses at different durations of the long diastole (1s -10s, and the whole long diastole). * significant different when compared to ZFP LR (two tailed paired t test and RM ANOVA, P< 0.05)
APPENDIX 4: ADDITIONAL FIGURES

A

B

C

D

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Erasmus University Rotterdam
Figure S13: Comparison ZFP<sub>LR</sub> versus ZFP<sub>LR3</sub>, ZFP<sub>CZO</sub>, ZFP<sub>BEL</sub>, and ZFP<sub>SCH</sub>.

The cerebral zero flow pressure (ZFP) estimated by four different methods, the 3-point intercept estimation (LR3, systolic/mean/diastolic), Czosnyka (CZO, 2 point systolic/diastolic), Belford (BEL, 2 point mean/diastolic), and Schmidt (SCH, 2 point systolic/diastolic) were validated against the reference method (LR, linear regression). One ZFP measurement corresponds to the cumulative analysis of 10-12 cardiac cycles within two breaths. 174 measurements were performed on 35 cardiovascular patients (5 each per patient) under general anesthesia and normocapnia. The figures explain differences (top rows: A-D), correlations (middle rows E-H) and agreement (bottom rows I-L) of the ZFP measurements. All statistical analyses were performed two-sided, and a P value of less than 0.05 was considered to be significant. All results are presented as mean (standard deviation).

Differences between the four tests methods of ZFP estimation (LR3, BEL, CZO SCH) and reference method (LR) were tested by ANOVA with correction for multiple comparison (Dunnett); § significant for all measurements; # significant for subject means.

Pearson r correlation coefficients between four ZFP values and the reference method (LR) of all observation were computed to assess their relationship. Agreement was analyzed by Bland-Altman plot analysis with correction for multiple measurements per subject (LL = lower limits of agreement, UL = upper limits of agreement, PE = percentage error).

Figure S14: Comparison rAP<sub>LR</sub> versus rAP<sub>LR3</sub>, rAP<sub>CZO</sub>, rAP<sub>BEL</sub>, and rAP<sub>SCH</sub>.

The resistance area product (RAP) estimated by four different methods, the 3-point intercept estimation (LR3, systolic/mean/diastolic), Czosnyka (CZO, 2 point systolic/diastolic), Belford (BEL, 2 point mean/diastolic), and Schmidt (SCH, 2 point systolic/diastolic) were validated against the reference method (LR, linear regression). One RAP measurement corresponds to the cumulative analysis of 10-12 cardiac cycles within two breaths. 174 measurements were performed on 35 cardiovascular patients (5 each per patient) under general anesthesia and normocapnia. The figures explain differences (top rows: A-D), correlations (middle rows E-H) and agreement (bottom rows I-L) of the RAP measurements. All statistical analyses were performed two-sided, and a P value of less than 0.05 was considered to be significant. All results are presented as mean (standard deviation).

Differences between the four tests methods of RAP estimation (LR3, BEL, CZO SCH) and reference method (LR) were tested by ANOVA with correction for multiple comparison (Dunnett); § significant for all measurements; # significant for subject means.

Pearson r correlation coefficients between four RAP values and the reference method (LR) of all observation were computed to assess their relationship. Agreement was analyzed by Bland-Altman plot analysis with correction for multiple measurements per subject (LL = lower limits of agreement, UL = upper limits of agreement, PE = percentage error).
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