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Transfer-function-free technique for the noninvasive determination of the human arterial pressure waveform

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Abstract

The estimation of central aortic blood pressure is a cardinal measurement, carrying effective physiological, and prognostic data beyond routine peripheral blood pressure. Transfer function-based devices effectively estimate aortic systolic and diastolic blood pressure from peripheral pressure waveforms, but the reconstructed pressure waveform seems to preserve features of the peripheral waveform. We sought to develop a new method for converting the local diameter distension waveform into a pressure waveform, through an exponential function whose parameters depend on the local wave speed. The proposed method was then tested at the common carotid artery. Diameter and blood velocity waveforms were acquired via ultrasound at the right common carotid artery while simultaneously recording pressure at the left common carotid artery via tonometer in 203 people (122 men, 50 ± 18 years). The wave speed was noninvasively estimated via the lnDU-loop method and then used to define the exponential function to convert the diameter into pressure. Noninvasive systolic and mean pressures estimated by the new technique were 3.8 ± 21.8 ($p = 0.015$) and 2.3 ± 9.6 mmHg ($p = 0.011$) higher than those obtained using tonometery. However, differences were much reduced and not significant in people >35 years (0.6 ± 18.7 and 0.8 ± 8.3 mmHg, respectively). This proof of concept study demonstrated that local wave speed, estimated from noninvasive local measurement of diameter and flow velocity, can be used to determine an exponential function that describes the relationship between local pressure and diameter. This pressure-diameter function can then be used for the noninvasive estimation of local arterial pressure.

KEYWORDS
carotid artery, hypertension, local wave speed, noninvasive estimation of pressure, pressure

1 | INTRODUCTION

The World Health Organization estimates that ~15% of the population worldwide suffers from high blood pressure, and only 20% of these are effectively managing their condition. According to the European Society of Hypertension, the current definition of hypertension entails having a brachial systolic blood pressure (P_s) ≥140 mmHg and/or diastolic blood pressure (P_d) ≥90 mmHg (Williams et al., 2018). In contrast to central aortic pressure, which may only be accurately measured invasively, brachial P_s and P_d can easily be assessed noninvasively via cuff measurement and are the gold standard in daily clinical practice because, for nearly 100 years, each was consistently shown to predict adverse cardiovascular outcomes.

Mean blood pressure (P_m) and P_d and are relatively constant throughout most of the arterial tree (Pauca et al., 2001; Wang et al., 2011), but P_s increases as the measurement site moves distally from the ascending aorta (Reference Values for Arterial Measurements Collaboration, 2014; Segers et al., 2009), most likely due to wave reflections and higher wall stiffness in the distal arteries compared to the aorta. Further, the magnitude of the pressure amplification is age, sex, and pathology dependent (McEniery et al., 2014; Reference Values for Arterial Measurements Collaboration, 2014). Therefore, using brachial pressure to estimate pressure in other regions of the arterial tree will generally be poor (Sharman et al., 2017).

Several studies have shown the added predictive value of central blood pressure for future cardiovascular events and stroke mortality, beyond brachial pressure and independent from established cardiovascular risk factors (Cheng et al., 2013; Chirinos et al., 2013). Indeed, it is expected that central blood pressure reflects the hemodynamic load on the left ventricle more accurately than brachial pressure (Roman et al., 2007). Further, central and peripheral blood pressure can be differentially affected by antihypertensive therapy, with potential clinical implications on patients management of hypertension and heart failure (Borlaug et al., 2014; Sharman et al., 2013; Williams et al., 2006). These findings promoted the development of methods to noninvasively estimate aortic pressure.

Transfer function-based techniques are currently the most commonly used methods for the noninvasive determination of aortic pressure. These functions describe the relationship between the central aortic pressure and the pressure measured at a peripheral site, and can be used to convert peripheral pressure waveforms into central aortic pressure (Costello et al., 2015; Ghasemi et al., 2017). While several commercial devices have been produced to solve this task (Ding et al., 2011) and empirical evidence suggests that some devices accurately predict the P_s–P_d range in the aorta (Ding et al., 2011), the estimated waveforms are similar to and preserve features of the measured peripheral pressure waveform (Millasseau et al., 2003; Segers et al., 2005). Hence, the waveforms estimated by the transfer functions might better represent the peripheral waveform than aortic root waveforms.

To overcome this issue, alternative methods have been introduced to directly estimate pressure from local arterial waveforms acquired noninvasively and a local estimate of wave speed (c) (Beulen et al., 2011; Vennin et al., 2015). Similar to transfer functions, these methods typically rely on the assumption that P_d and P_m are the same in most arterial locations. Vennin et al. (2015) proposed a method to reconstruct the aortic pressure waveform (P) from noninvasive acquisition of aortic blood flow velocity waveform (U), peripheral P_s and P_d, and features of the exponential decay of peripheral pressure in diastole. The method relies on values of c and the water hammer equation (Khir et al., 2001) to convert the systolic ejection in the velocity waveform into the pressure upstroke. While this method provided plausible estimations of aortic P both in computational and in vivo settings, the pressure waveform involved in the determination of c (Davies et al., 2005) was recorded invasively. This renders the technique unsuitable for routine examination, although its relative accuracy is yet to be determined if c is estimated noninvasively. Beulen et al. (2011) used simultaneous ultrasound measurements of U and diameter distension waveform (D) to estimate P in flexible tubes. The flow-area (Q–A) method (Rabben et al., 2004) was used to determine c noninvasively, and the relationship between c and distensibility (Ds), described by the Bramwell–Hill equation (Bramwell et al., 1923), was used to calculate pressure by integrating changes in tube cross-sectional area with respect to the diastolic reference. This method assumes that c is constant across the pressure range of the entire cardiac cycle. While this assumption might be correct in the case of flexible tubes with a linear P–A relationship, the latter is nearly exponential in arteries (Gavish & Izzo, 2016), implying that c does increase with increasing pressure.

The aim of this study was to develop a noninvasive method for estimating arterial pressure from local hemodynamic waveforms. Our ultimate goal was to provide an alternative to transfer functions using local noninvasive measurements for estimating central aortic pressure. To that end and as a proof of concept, we tested the new technique using data measured at the common carotid artery (CCA) and compared the results against applanation tonometry.

2 | METHODS

The general methodology of the technique is to construct the exponential relationship between P and A in arteries using

local c, which can be determined noninvasively from ultrasound measurement of local D and U using the lnDU-loop method (Feng & Khir, 2010). Once the P–A relationship is established, P is estimated using noninvasive local measurement of D and peripheral P_d. We applied this approach to carotid artery data, comparing the new technique with applanation tonometry, a well-established method for the recording of pressure in superficial arteries (Segers et al., 2009).

2.1 | Theoretical background

Tube laws describe the relationship between P and A, or D of a flexible tube. Assuming that arteries are cylindrical, the P–A relationship of arteries, closely resembling an exponential function (Fung, 1967; Spronck et al., 2015), can be written in terms of P and D^2. The tube law used in this study is that proposed by Meinders and Hoeks (2004) (Equation 1):

$$P(D) = P_d \cdot e^{\gamma\left(\frac{D^2}{D_d^2} - 1\right)}, \tag{1}$$

where P_d is the diastolic pressure, D_d is the diastolic diameter (i.e., the diameter at P_d), and γ is an exponential gain defining the relationship between P and D^2. The objective of the following derivation is to re-write Equation 1 using noninvasive parameters; thus allowing for the noninvasive determination of pressure.

Arterial distensibility is defined as $Ds = dA/(AdP)$, where dA is the change in the vessel cross-sectional area in response to a change in pressure (dP). The relationship between Ds and c is expressed in the Bramwell–Hill equation (Equation 2) (Bramwell et al., 1923):

$$c = D\sqrt{\frac{dP}{\rho \cdot d(D^2)}}, \tag{2}$$

where ρ is the blood density. Equation 2 states that the wave speed c at any given pressure P_c can be expressed as a function of the slope of the tangent to the P–D^2 relationship at the pressure level P_c (and corresponding D_c so that $P(D_c) = P_c$). Therefore, for any estimate of c there must be a P_c satisfying Equation 2. Using Equation 1, the derivative term of Equation 2 can be rearranged as

$$\frac{dP}{dD^2} = \frac{\gamma \cdot P_d}{D_d^2} e^{\gamma\left(\frac{D^2}{D_d^2} - 1\right)}. \tag{3}$$

Inverting Equation 1 to express the diameter as a function of pressure, we obtain

$$D(P) = D_d\sqrt{1 + \frac{\ln\left(\frac{P}{P_d}\right)}{\gamma}}, \tag{4}$$

and substituting D in Equation 3 with Equation 4, the derivative term is furtherly manipulated in:

$$\frac{dP}{dD^2} = \frac{\gamma \cdot P_d}{D_d^2} \cdot e^{\gamma\left[\frac{\ln\left(\frac{P}{P_d}\right)}{\gamma}\right]} = \frac{\gamma \cdot P_d}{D_d^2} \cdot e^{\ln\left(\frac{P}{P_d}\right)} = \frac{\gamma \cdot P}{D_d^2}. \tag{5}$$

Substituting Equation 5 into Equation 2 and knowing that for any given c Equation 2 is verified for $P = P_c$, we obtain:

$$c^2 = \frac{D_c^2 \cdot \gamma \cdot P_c}{D_d^2 \cdot \rho}. \tag{6}$$

Then, replacing D_c in Equation 6 with Equation 4 and solving for γ leads to the following relationship:

$$\gamma = \frac{\rho \cdot c^2}{P_c} - \ln\left(\frac{P_c}{P_d}\right), \tag{7}$$

where P_c is the pressure level at which c is calculated. Hence, it is expected that P_c is the representative pressure for pressure range pertaining to the method chosen for the estimation of c.

2.2 | Study population and acquisition protocol

The data used in this study were acquired at the University Hospital of Pisa. The study population included 203 people (122 men, 51±17 years, age range 16–78 years) undergoing standard out-patient cardiovascular risk assessment, all free of major cardiovascular events, atrial fibrillation, malignancy, or chronic inflammatory disease. All subjects were referred for a complete cardiovascular examination to the Clinic for Cardiometabolic Risk Prevention of the Department of Surgical and Medical Pathology, University of Pisa. The protocol of the study followed the principles of the Declaration of Helsinki and was approved by the institutional ethics committee "Comitato Etico di Area Vasta Nord Ovest" (reference number: 3146/2010). Everyone gave their informed consent to participate. Clinical characteristics of the study participants are reported in Table S1 (https://figshare.com/s/4aab7f7fd026d8fbb761).

P, D, and U waveforms of the CCAs were acquired simultaneously by a single experienced operator (C.M.), following an earlier reported protocol (Giannattasio et al.,

2008). Simultaneous ultrasound acquisition of D and U was performed on the right CCA using a 10.0-MHz linear array probe with radiofrequency data output at the frequency of 1 kHz connected to an Aloka Alpha10 Prosound system (Hitachi Ltd., Tokyo, Japan) as reported previously (Giudici et al., 2020). Given the impossibility of placing a pressure tonometer on the right CCA due to the presence of the ultrasound probe, P was acquired at the left CCA using a PulsePen (DiaTecne, Milan, Italy) with sampling frequency 1 kHz. Acquisitions lasted for approximately 10 s, granting at least seven heartbeats where P and D-U were recorded simultaneously.

The carotid ultrasound/tonometer acquisitions were performed after the subject had rested in the supine position for at least 10 min. Brachial P_s and P_d ($_bP_s$ and $_bP_d$) were measured by an electronic digital manometer (Omron, model 705cp, Kyoto, Japan) and the average of two consecutive measurements was used for calibration.

The data that support the findings of this study are available from the corresponding author upon reasonable request.

2.3 | Noninvasive estimation of the local wave speed ($_nc$) and exponential gain ($_n\gamma$)

The local CCA wave speed was estimated using the lnDU-loop method, whose complete derivation is described in earlier work (Feng & Khir, 2010). Briefly, when arterial waves are unidirectional (i.e., travelling only from the heart toward the periphery or vice versa), the relationship between the U and the natural logarithm of D is linear and proportional to the wave speed (Equation 8).

$$_nc = \pm \frac{1}{2} \frac{dU_\pm}{d(\ln D_\pm)}, \quad (8)$$

where subscripts $+$ and $-$ indicate forward (from the heart to the periphery) and backward (from the periphery to the heart) direction of wave travel. Following cardiac ejection, a forward travelling compression wave is generated. Assuming only forward waves exist in early systole, as it will be too early for reflected waves to return from the periphery, the unidirectionality of the waves is reasonable, and $_nc$ can be determined with Equation 8 (Figure 1b).

The exponential gain $_n\gamma$ can then be calculated using Equation 7 and $_nc$ if P_c for the lnDU method is known. Given the inherent assumption in the loop methods that c is constant during that pressure range, we take P_d as fiducial marker of the early systolic pressure range that is easily obtained noninasively since constant throughout the circulation. Hence, we assume that $P_c = P_d = {_bP_d}$ and Equation 7 reduces to

$$_n\gamma = \frac{\rho \cdot {_nc^2}}{_bP_d}. \quad (9)$$

with $\rho = 1060$ kg/m³. A similar approach has been described previously for regional pulse wave velocity (Spronck et al., 2017).

To provide a means of comparison for $_n\gamma$, the exponential gain was also calculated using the tonometer waveform; inverting Equation 1 and considering the systolic pressure and diameter leads to

$$_t\gamma = \frac{\ln\left(\frac{_tP_s}{_tP_d}\right)}{\frac{D_s^2}{D_d^2} - 1}, \quad (10)$$
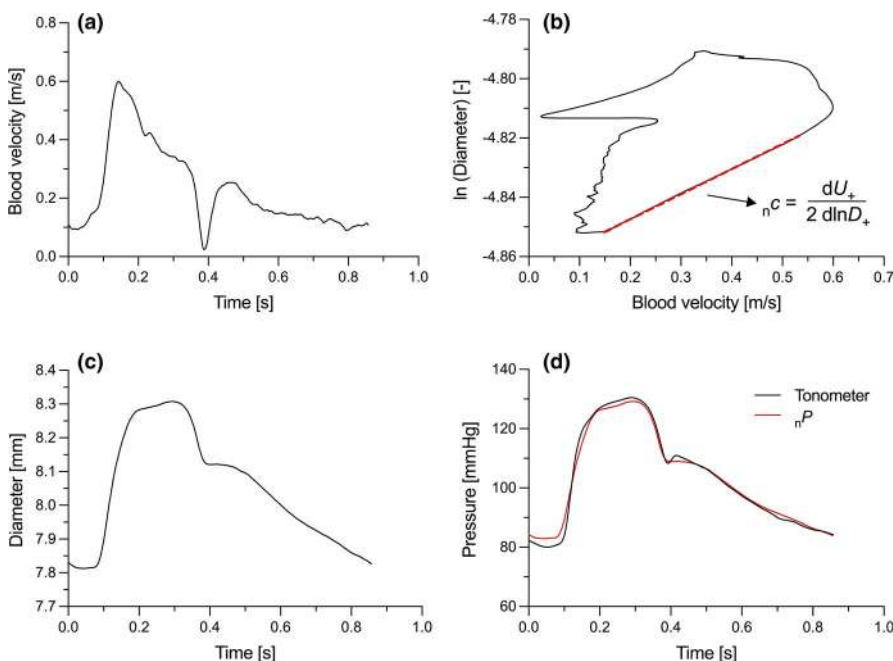
FIGURE 1 Ensemble averaged measurements of blood flow velocity (U) (a) and diameter (D) (c) waveforms measured at the carotid artery for a 74 years old patient. The lnDU-loop and noninvasive pressure ($_nP$) estimation are presented, respectively in (b, d). The noninvasive wave speed ($_nc$) is estimated from the slope of the initial linear part of the lnDU-loop (Equation 8). $_nc$ is then used to estimate the exponential gain $_n\gamma$ (Equation 9) and convert the diameter waveform into a pressure waveform using Equation 11

where $_tP_s$ and $_tP_d$ are the average P_s and P_d of all the heartbeats ($N = 7$–10) of the tonometer acquisition (i.e., the peak and the minimum pressure in each cardiac cycle), and D_s and D_d are the average systolic and diastolic D determined from the ultrasound acquisition.

2.4 | Noninvasive estimation of pressure

Assuming a uniform P_d throughout the arterial system, the diameter waveform can be converted into a pressure waveform using Equation 1 and $_n\gamma$.

$$_nP(D) = {_bP_d} \cdot e^{n\gamma\left(\frac{D^2}{D_d^2}-1\right)} \qquad (11)$$

As $_bP_d$, D and $_n\gamma$ are all determined noninvasively, $_nP$ can therefore be determined entirely noninvasively. $_nP_s$ was calculated as the average of the peaks of all the cardiac cycles ($N = 7$–10) of the estimated $_nP$ waveform. $_nP_m$ was the arithmetic mean of all the data points of $_nP$.

2.5 | Statistical analysis

Data are reported as mean ± standard deviation (SD). $_nP_s$ and $_nP_m$ were compared with tonometer measurements of $_tP_s$ and $_tP_m$. The tonometer acquisition of pressure was calibrated using $_bP_d$ and $_bP_m$ and assuming constant P_m and P_d throughout the arterial system. $_bP_m$ was estimated using a form factor (FF) of 0.43 as $_bP_m = {_bP_d} + 0.43$ $(_bP_s - {_bP_d})$ (Segers et al., 2009). The noninvasive carotid FF ($_nFF$) was used as an additional parameter to quantify the accuracy of the new technique for estimating the pressure waveform $_nFF = \frac{_nP_m - {_bP_d}}{_nP_s - {_bP_d}}$, and compared to FF calculated from the left CCA tonometry ($_tFF$).

Comparison between noninvasive and tonometer output variables was initially performed using paired sample t-test and Bland–Altman plots (Bland & Altman, 1986), first on the entire cohort and then stratifying the population in three age groups: young (<35 years), middle-aged (35–59 years), and older adults (≥70 years). This allowed a first evaluation of the effect of age on the accuracy of the pressure estimation.

Then, age was used as a continuous variable in multivariate regression analysis, including $_nP$ as dependent variable and corresponding $_tP$ value (i.e., P_s and P_m independently), age, type 1 diabetes mellitus (T1DM), type 2 diabetes mellitus (T2DM), antihypertensive treatment, and dyslipidemia as independent variables.

Linear regression and correlation analysis were performed where appropriate. $p \leq 0.05$ was considered statistically significant.

3 | RESULTS

The hemodynamic characteristics of subjects included in this study are reported in Table 1. $_bP_s$ and $_bP_d$ were 122.1 ± 16.8 mmHg and 75.2 ± 10.3 mmHg, respectively. Using a form factor 0.43 (Equation 11) lead to $_bP_m = 95.4 ± 12.0$ mmHg.

Average noninvasive wave speed $_nc$ was 5.67 ± 1.45 m/s. $_n\gamma$ was comparable to $_t\gamma$ estimated using applanation tonometry (3.60 ± 1.75 vs. 3.53 ± 1.48, limits of agreement: −2.42 to 2.54, $p = 0.49$) and the two metrics showed strong correlation (Figure 2). When stratifying our cohort in age groups, $_n\gamma$ was significantly higher than $_t\gamma$ in young people (≤35 years, $p = 0.006$), but not in middle-aged and older adults ($p = 0.73$ and $p = 0.51$, respectively) (Table 1 and Figure S1, https://figshare.com/s/4aab7f7fd026d8f bb761). However, in the multivariate regression analysis, no significant interaction was found between age and $_n\gamma$ ($\beta = 0.056$, 95% confidence interval (CI) [−0.093–0.196], $p = 0.47$) (Table S2, https://figshare.com/s/4aab7f7fd0 26d8fbb761).

Figure 1d shows the comparison between $_nP$, estimated by the new technique, and P, acquired with tonometry, for a 74 year old subject included in this study. On average, noninvasive $_nP_s$ and $_nP_m$ were 3.8 (limits of agreement: −39.9 to 47.4) mmHg ($p = 0.015$) and 2.3 (−17.0 to 21.5) mmHg ($p = 0.011$) higher than $_tP_s$ and $_tP_m$ acquired via tonometry, respectively (Table 1 and Figure 3b–d). Correlation between the two techniques was strong for P_m (Figure 3a) and moderate for P_s (Figure 3c). Further, the Bland–Altmann plots (Figure 3b–d) showed weak correlations between the difference and average of P_m and P_s determined with the two techniques ($r = 0.35$, $p < 0.001$ and $r = 0.18$, $p = 0.010$, respectively). Overall, $_nFF$ was slightly higher than $_tFF$ ($p < 0.001$).

As for γ, the pressure estimation using the new method performed better, on average, in middle-aged and older adults; the average differences with tonometry for $_nP_s$ and $_nP_m$ were 1.8 (limits of agreement: −29.9 to 33.6) mmHg ($p = 0.31$) and 1.3 (limits of agreement: −12.6 to 15.2) mmHg ($p = 0.11$) in middle-aged subjects, and −0.6 (limits of agreement: −42.8 to 41.6) mmHg ($p = 0.80$) and 0.3 (limits of agreement: −18.5 to 19.1) mmHg ($p = 0.76$) in older adults (Figure S2 (https://figsh are.com/s/4aab7f7fd026d8fbb761) and Figure 4 for P_s and P_m, respectively). However, as for γ, the interaction between age and $_nP_s$ or $_nP_m$ was not significant ($\beta = -0.078$, 95% CI [−0.260–0.104], $p = 0.40$ and $\beta = -0.057$, 95% CI [−0.198–0.084], $p = 0.43$, respectively). Further, the clinical background did not affect the pressure estimation (Table S2 https://figshare.com/s/4aab7f7fd026d8f bb761).

TABLE 1 Measured and noninvasively estimated hemodynamic characteristics of the people included in the study

	All	≤35 years	36–59 years	≥60 years
N (male)	203 (60%)	47 (59%)	78 (53%)	78 (68%)
Age [years]	51 ± 17	24 ± 5	51 ± 6	67 ± 5
Brachial artery				
$_bP_s$ [mmHg]	122.1 ± 16.8	111.4 ± 11.7	120.9 ± 14.7	129.8 ± 17.6
$_bP_d$ [mmHg]	75.2 ± 10.3	67.0 ± 8.5	77.8 ± 9.4	77.5 ± 9.5
Carotid artery				
$_nc$ [m/s]	5.67 ± 1.45	4.45 ± 0.73	5.58 ± 1.12	6.49 ± 1.50
$_t\gamma$ [−]	3.53 ± 1.48	2.11 ± 0.54	3.29 ± 0.99	4.64 ± 1.43
$_n\gamma$ [−]	3.60 ± 1.75	$2.43 \pm 0.80^\dagger$	3.33 ± 1.30	4.53 ± 1.98
$_tP_s$ [mmHg]	120.3 ± 17.3	110.5 ± 13.0	118.5 ± 15.0	128.1 ± 18.1
$_nP_s$ [mmHg]	$124.1 \pm 23.6^*$	$122.2 \pm 25.6^\dagger$	120.4 ± 18.9	127.5 ± 23.3
$_tP_m$ [mmHg]	95.4 ± 12.0	86.1 ± 8.4	96.3 ± 11.0	100.0 ± 11.7
$_nP_m$ [mmHg]	$97.6 \pm 13.6^*$	$91.7 \pm 12.1^\ddagger$	97.6 ± 11.8	100.3 ± 13.7
$_tFF$ [−]	0.45 ± 0.04	0.45 ± 0.03	0.46 ± 0.04	0.45 ± 0.04
$_nFF$ [−]	$0.46 \pm 0.03^\ddagger$	0.46 ± 0.03	$0.47 \pm 0.03^*$	$0.46 \pm 0.03^\dagger$

Comparison between tonometry and new method: $^*p < 0.05$, $^\dagger p < 0.01$, and $^\ddagger p < 0.001$.

Abbreviations: $_bP_d$, brachial diastolic blood pressure; $_bP_s$, brachial systolic blood pressure; $_nc$, noninvasive wave speed; $_nP_m$, estimated carotid mean pressure; $_nP_s$, estimated carotid systolic pressure; $_n\gamma$, exponential gain estimated from $_nc$; $_tFF = (_tP_m - _bP_d)/(_tP_s - _bP_d)$, $_nFF = (_nP_m - _bP_d)/(_nP_s - _bP_d)$; $_tP_m$, tonometer carotid mean blood pressure; $_tP_s$, tonometer carotid systolic blood pressure; $_t\gamma$, exponential gain estimated from the tonometer pressure and ultrasound diameter waveforms.
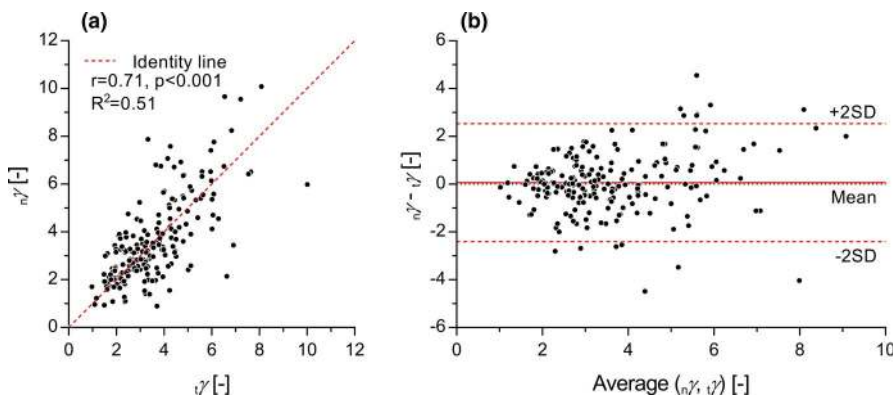

FIGURE 2 Correlation (a) and Bland–Altman (b) plot between exponential constants estimated from the tonometer pressure, $_t\gamma$ (Equation 10), and from the noninvasive wave speed, $_n\gamma$ (Equation 9), of the $N = 203$ subject included in this study. Limits of agreement were calculated as ± 2 standard deviations (SD)

4 | DISCUSSION

In this study, we proposed a new technique where the local wave speed, estimated by noninvasive local measurements of diameter distension and blood flow velocity waveforms, is used to estimate the parameters of an exponential function that allows converting the diameter waveform into a pressure waveform. In this proof of concept study, we compared the performance of the proposed method with CCA pressure measured using applanation tonometry in a group of healthy controls and hypertensive and diabetic patients. Results of the new technique compared well, on average, with those measured using applanation tonometry, but limits of agreements between the two techniques were high, especially for P_s.

In the past two decades, several commercial TF-based devices have been developed to estimate pressure noninvasively in the aorta. Although they are the most commonly used, their accuracy is still called into question. Ding et al. (2011) compared invasively measured central aortic pressure with estimates provided by two commercial devices, SphygmoCor and Omron HEM-9000AI, both relying on the measurement of radial pressure waveforms calibrated with cuff measurement of brachial pressure. The first underestimated aortic P_s by 15 mmHg and the limits of agreement in the Bland–Altman plot were approximately −33 to 3 mmHg. The Omron device performed slightly better on average, with limits of agreement of approximately ±20 mmHg. Laugesen et al. (2014) showed that calibrating the radial pressure waveform

FIGURE 3 Correlation (a, c) and Bland–Altman (b, d) plots between tonometer pressure and pressure estimated with the new technique in the $N = 203$ subject included in this study: mean pressure (P_m) (a, b) and systolic pressure (P_s) (c, d). Limits of agreement were calculated as ± 2 standard deviations (SD)
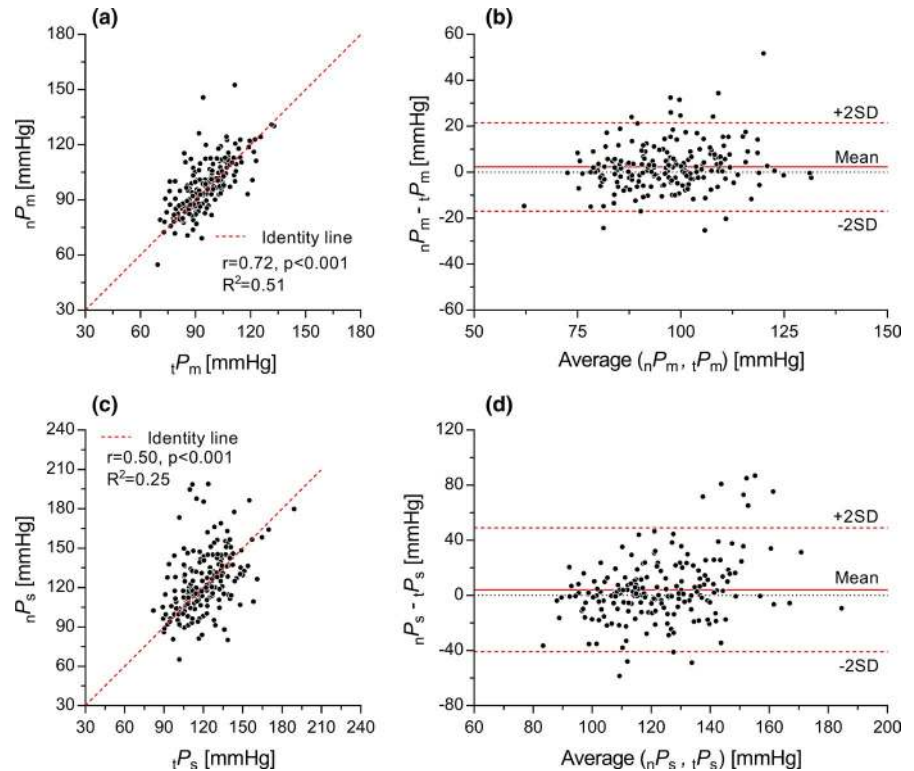

with oscillometric brachial pressure did reduce, on average, the underestimation of the SphygmoCor synthesized central P_s, but retained similarly wide limits of agreement (± 22 mmHg). Only the calibration using invasively measured aortic P_s and P_d considerably improved the accuracy (± 11 mmHg).

Compared to applanation tonometry measurements, our method overestimated P_s and P_m by 3.8 and 2.3 mmHg in the overall study population. Although multivariate regression analysis did not yield any significant interaction between noninvasive parameters and age, these differences were largely attributable to young subjects, where P_s and P_m were overestimated by ~11% and ~7%, respectively, while smaller and nonsignificant differences were found in middle-aged and older adults. However, limits of agreement for P_s were wider than that reported for commercial devices (-36.8 to 38.0 mmHg in middle-aged and older adults). These results might be due to the fact that the brachial pressure waveform was not acquired in our study. Conversely, $_bP_m$ used for calibrating the tonometer pressure waveform was estimated using average FF previously reported for the brachial artery; $FF = 0.43$ (Segers et al., 2009). Here, carotid $_tFF$, determined from the acquired tonometer waveforms, averaged 0.45 but ranged between 0.36 and 0.57, and a similar variability was reported for the brachial artery (Grillo et al., 2020). Hence, neglecting the age-dependence and inter-subjects variability of the brachial FF likely affected the accuracy of the calibration of the tonometer pressure waveforms. Grillo et al. (2020) recently proposed an alternative

method for the estimation of a subject-specific brachial FF derived from $_bP_d$ and gender. They showed that the method predicts age-differences of brachial FF more effectively in middle-aged and older adults. However, as conceded by the authors, the accuracy of the proposed formula remained sub-optimal and unable to capture the high inter-subject variability of the brachial FF. When applied to our data, estimation of $_bP_m$ from $_bP_d$ and gender did not improve the agreement between $_nP$ and $_tP$ in any of the age groups considered here.

The accurate estimation of P_s is undoubtedly important, however little attention is generally given to the shape and high frequency components of the pressure waveform synthesized from distal measurements. The pressure waveform at any arterial location is widely accepted to be the linear summation of the forward travelling pressure waves, generated by left ventricular contraction, and the backward travelling waves, originated at reflection sites when the forward travelling wave meets discontinuities (i.e., mismatched bifurcations and downstream tapering of the arterial tree) (Abdullateef et al., 2020; Khir & Parker, 2005). Given the complex structure of the arterial tree, the magnitude and timing of reflected waves are highly location-dependent, making the estimation of pressure at any location from pressure acquired elsewhere in the arterial tree a complicated task. Indeed, the accuracy of estimating aortic waveform from radial measurements using transfer functions remains controversial. Segers et al. (2005) found that the augmentation index (AIx), an estimate of the relative magnitude of the
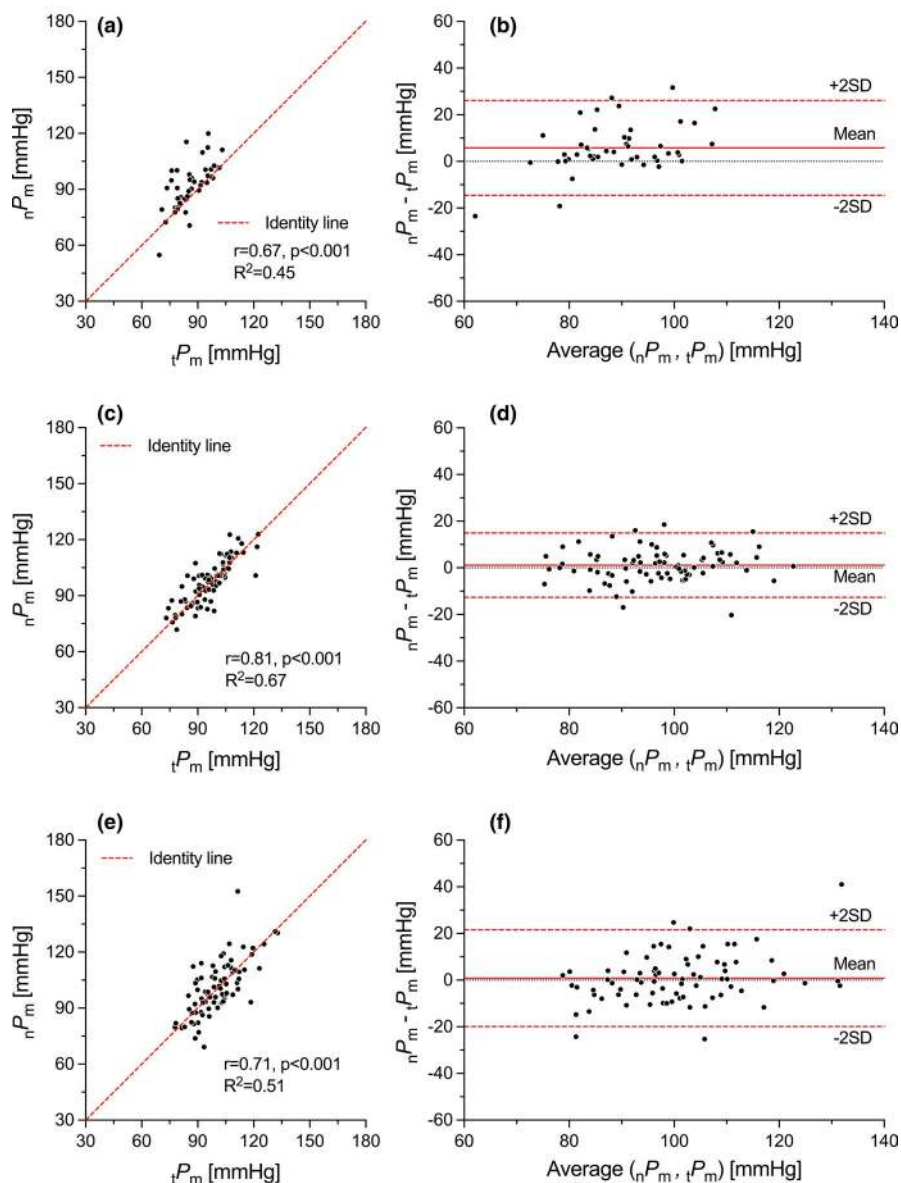
FIGURE 4 Figure S2—Correlation (a, c, and e) and Bland–Altman (b, d, f) plots between tonometer mean pressure ($_tP_m$) and pressure estimated with the new technique ($_nP_m$). (a, b) Young people (<35 years, $N = 47$); (c, d) middle-aged people (35–59 years, $N = 78$); (e, f) older adults (≥60 years, $N = 78$)

reflected wave, calculated from the TF-synthesized aortic pressure mildly correlated with the carotid AIx acquired using tonometry, although strongly correlated with that of the radial pressure waveform used in the transfer function. This result contradicts previous findings showing that carotid AIx strongly correlates with that of invasively measure aortic pressure waveforms (Chen et al., 1996), casting further doubts on the accuracy of generalized TF-based aortic waveforms (Millasseau et al., 2003; Segers et al., 2005).

The magnitude and timing of reflected waves in central arteries has been positively associated with ventricular function (Park et al., 2020) and the incidence of cardiovascular events (Sugawara et al., 2009; Wang et al., 2010). Therefore, findings by Segers et al. (2005) suggest that the pressure waveform estimated via transfer function might carry information on reflections at peripheral

sites but be less than ideal to evaluate the subject-specific cardiac risk. Additionally, a previous study from our group showed good agreement between wave intensity analysis, using standard invasive P and U and noninvasive D and U methods (Li & Khir, 2011). This suggests that the information on the complex interaction between forward and backward waves is better captured by using local measurements of U together with either P or D, further supporting the new approach presented here; using the local lnDU-loop to facilitate the estimation of local pressure waveform.

Methods that noninvasively estimate pressure from local arterial waveforms are likely to provide more accurate alternatives to transfer functions, following the rationale that local waveforms necessarily carry more representative information on local hemodynamics than peripheral pressure. Vennin et al. (2015) used the "water

hammer" equation (Khir et al., 2001) to convert the up-stroke of the flow velocity waveform into the pressure upstroke, and then modelled the elastic recoil in diastole with an exponential decay function and the pressure peak in late systole. The method yielded good results both in a one-dimensional computational model of the arterial tree and in vivo. However, the use of invasively measured aortic P for the estimation of c using the sum of squares method (Davies et al., 2005) makes this technique less likely to be used clinically. Also, the accuracy of their technique remains to be examined when using a noninvasive estimation of c.

Beulen et al. (2011) were the first to use the relationship between c and arterial distensibility to convert the arterial diameter/area distension waveform into P. It is worth noting, however, that the underlying assumption in Beulen et al. is that c is constant in the investigated pressure range, yielding to a linear P–D^2 relationship. While such assumption is reasonable for the flexible tubes used for the validation of their method, arteries exhibit a nonlinear, approximately exponential relationship (Fung, 1967) and c is pressure-dependent (Spronck et al., 2015). Application of their method on our data underestimated P_s (Figure S3 https://figshare.com/s/4aab7f7fd026d8fbb761). On the contrary, we assumed that c determined by the lnDU-loop in early systole describes the slope of the P–D^2 relationship in the proximity of P_d but allows the estimation of γ that, together with local D_d and P_d (here assumed equal to P_d in the brachial artery), defines the exponential relationship between P and D^2. Hence, our method relaxes the assumption that c is pressure independent. As a result, the $_nFF$ closely matched that calculated on the tonometer pressure acquisition (Table 1) and $_nP_s$, on average, did not underestimate but agreed well with $_tP_s$.

4.1 | Limitations

As all the noninvasive methods for the estimation of pressure, whether based on transfer functions or local arterial waveforms, the accuracy of our method strongly depends on the fidelity of measurement of peripheral P, with brachial cuff measurement typically under and overestimating P_s and P_d, respectively (Picone et al., 2017). Our method requires only P_d and, hence, is less affected by any potential inaccuracies pertaining cuff measurements. However, the inaccuracy of the measurements of brachial pressure likely affected the comparison between the two techniques, since tonometer waveforms were calibrated using both P_s and P_d.

The accuracy of determining the local pressure waveform proposed in this work strongly depends on the accuracy of the estimation of wave speed c which appears squared in the formulas (Equation 9). Here, we used the lnDU-loop method to noninvasively estimate c from the relationship between D and U in early systole. It was suggested previously that the accuracy of the loop methods is affected by the proximity to the reflection site and magnitude of the reflected waves (Borlotti et al., 2014; Segers et al., 2014) and that the lnDU-loop method might underestimate c at the level of the CCA (Willemet et al., 2016). This underestimation was not observed here; c estimated with the lnDU-loop and D^2P-loop (Alastruey, 2011), which is not affected by reflections, did not differ significantly (Giudici et al., 2021). Nevertheless, the method proposed here can be applied on any noninvasive estimate of c, provided that the correct P_c is known.

5 | CONCLUSIONS

This proof of concept study introduces a method for estimating pressure using local hemodynamic parameters recorded noninvasively. The proposed technique was tested on the common carotid artery where reference pressures for comparison could also be acquired noninvasively using tonometry. The promising result presented suggests that pressure can be estimated noninvasively at any arterial location where blood velocity and diameter waveforms can be acquired via ultrasound, making the measurement of central aortic pressure a real possibility. Doing so will characterize ventricular afterload more precisely, including potentially cardiovascular risk. Further work is warranted to test the effectiveness of the new method against invasively measured pressure and in estimating central aortic pressure.

DISCLOSURES
JKC is a former president of the Artery Society. MK is responsible for clinical studies at Esaote SpA (Genova, Italy).

AUTHORS CONTRIBUTION
AG contributed to the conceptualization, data analysis, manuscript drafting, and editing. AG and AWK developed the analytical method. AWK, CP, JKC, MK, and IBW contributed to the conceptualization, manuscript editing, and project supervision. CM contributed to the data acquisition and management.

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REFERENCES

Abdullateef, S., Mariscal-Harana, J., & Khir, A. W. (2020). Impact of tapering of arterial vessels on blood pressure, pulse wave velocity, and wave intensity analysis using one-dimensional computational model. *International Journal for Numerical Methods in Biomedical Engineering*, *18*, e3312. https://doi.org/10.1002/cnm.3312

Alastruey, J. (2011). Numerical assessment of time-domain methods for the estimation of local arterial pulse wave speed. *Journal of Biomechanics*, *44*, 885–891. https://doi.org/10.1016/j.jbiomech.2010.12.002

Beulen, B. W. A. M. M., Bijnens, N., Koutsouridis, G. G., Brands, P. J., Rutten, M. C. M., & Van de Vosse, F. N. (2011). Towards non-invasive blood pressure assessment in arteries by using ultrasound. *Ultrasound in Medicine and Biology*, *37*, 788–797.

Bland, M. J., & Altman, D. G. (1986). Statistical methods for assessing agreement between two methods of clinical measurement. *Lancet*, *327*, 307–310. https://doi.org/10.1016/S0140-6736(86)90837-8

Borlaug, B. A., Olson, T. P., Mohamed, S. A., Melenovsky, V., Sorrell, V. L., Noonan, K., Lin, G., & Redfield, M. M. (2014). A randomized pilot study of aortic waveform guided therapy in chronic heart failure. *Journal of the American Heart Association*, *3*(4), e001214. https://doi.org/10.1161/JAHA.113.000745

Borlotti, A., Li, Y., Parker, K. H., & Khir, A. W. (2014). Experimental evaluation of local wave speed in the presence of reflected waves. *Journal of Biomechanics*, *47*, 87–95. https://doi.org/10.1016/j.jbiomech.2013.10.007

Bramwell, J. C., Hill, A. V., & McSwiney, B. A. (1923). The velocity of the pulse wave in man in relation to age as measured by the hot-wire sphygmograph. *Heart*, *10*, 233–255.

Chen, C. H., Ting, C. T., Nussbacher, A., Nevo, E., Kass, D. A., Pak, P., Wang, S. P., Chang, M. S., & Yin, F. C. P. (1996). Validation of carotid artery tonometry as a means of estimating augmentation index of ascending aortic pressure. *Hypertension*, *27*, 168–175. https://doi.org/10.1161/01.HYP.27.2.168

Cheng, H. M., Chuang, S. Y., Sung, S. H., Yu, W. C., Pearson, A., Lakatta, E. G., Pan, W. H., & Chen, C. H. (2013). Derivation and validation of diagnostic thresholds for central blood pressure measurements based on long-term cardiovascular risks. *Journal of the American College of Cardiology*, *62*, 1780–1787. https://doi.org/10.1016/j.jacc.2013.06.029

Chirinos, J. A., Segers, P., Gillebert, T. C., De Buyzere, M. L., Van Daele, C. M., Khan, Z. A., Khawar, U., De Bacquer, D., & Rietzschel, E. R. (2013). Central pulse pressure and its hemodynamic determinants in middle-aged adults with impaired fasting glucose and diabetes: The Asklepios study. *Diabetes Care*, *36*, 2359–2365.

Costello, B. T., Schultz, M. G., Black, J. A., & Sharman, J. E. (2015). Evaluation of a brachial cuff and suprasystolic waveform algorithm method to noninvasively derive central blood pressure. *American Journal of Hypertension*, *28*, 480–486. https://doi.org/10.1093/ajh/hpu163

Davies, J. E., Whinnett, Z. I., Francis, D. P., Willson, K., Foale, R. A., Malik, I. S., Hughes, A. D., Parker, K. H., & Mayet, J. (2005). Use of simultaneous pressure and velocity measurements to estimate arterial wave speed at a single site in humans. *American Journal of Physiology-Heart and Circulatory Physiology*, *290*, H878–H885. https://doi.org/10.1152/ajpheart.00751.2005

Ding, F. H., Fan, W. X., Zhang, R. Y., Zhang, Q., Li, Y., & Wang, J. G. (2011). Validation of the noninvasive assessment of central blood pressure by the sphygmocor and omron devices against the invasive catheter measurement. *American Journal of Hypertension*, *24*, 1306–1311. https://doi.org/10.1038/ajh.2011.145

Feng, J., & Khir, A. W. (2010). Determination of wave speed and wave separation in the arteries using diameter and velocity. *Journal of Biomechanics*, *43*, 455–462. https://doi.org/10.1016/j.jbiomech.2009.09.046

Fung, Y. C. (1967). Elasticity of soft tissues in simple elongation. *American Journal of Physiology*, *213*, 1532–1544. https://doi.org/10.1152/ajplegacy.1967.213.6.1532

Gavish, B., & Izzo, J. L. (2016). Arterial stiffness: Going a step beyond. *American Journal of Hypertension*, *29*, 1223–1233.

Ghasemi, Z., Kim, C. S., Ginsberg, E., Gupta, A., & Hahn, J. O. (2017). Model-based blind system identification approach to estimation of central aortic blood pressure waveform from noninvasive diametric circulatory signals. *Journal of Dynamic Systems, Measurement, and Control*, *139*, 1–10. https://doi.org/10.1115/1.4035451

Giannattasio, C., Salvi, P., Valbusa, F., Kearney-Schwartz, A., Capra, A., Amigoni, M., Failla, M., Boffi, L., Madotto, F., Benetos, A., & Mancia, G. (2008). Simultaneous measurement of beat-to-beat carotid diameter and pressure changes to assess arterial mechanical properties. *Hypertension*, *52*, 896–902. https://doi.org/10.1161/HYPERTENSIONAHA.108.116509

Giudici, A., Palombo, C., Kozakova, M., Morizzo, C., Losso, L., Nannipieri, M., Berta, R., Hughes, A. D., Cruickshank, J. K., & Khir, A. W. (2020). Weight loss after bariatric surgery significantly improves carotid and cardiac function in apparently healthy people with morbid obesity. *Obesity Surgery*, *30*, 3776–3783. https://doi.org/10.1007/s11695-020-04686-y

Giudici, A., Palombo, C., Kozakova, M., Morizzo, C., Penno, G., Jamagidze, G., Della Latta, D., Chiappino, D., Cruickshank, J. K., & Khir, A. W. (2021). Noninvasive carotid pressure-diameter loops to identify viscoelastic properties in ageing, hypertension and type 2 diabetes. *Journal of Hypertension*, in press. https://doi.org/10.1097/HJH.0000000000002918

Grillo, A., Salvati, P., Furlanis, G., Baldi, C., Rovina, M., Salvi, L., Faini, A., Bilo, G., Fabris, B., Carretta, R., Benetos, A., & Parati, G. (2020). Mean arterial pressure estimated by brachial pulse wave analysis and comparison with currently used algorithms. *Journal of Hypertension*, *38*, 2161–2168. https://doi.org/10.1097/HJH.0000000000002564

Khir, A. W., O'Brien, A., Gibbs, J. S. R., & Parker, K. H. (2001). Determination of wave speed and wave separation in the arteries. *Journal of Biomechanics*, *34*, 1145–1155. https://doi.org/10.1016/S0021-9290(01)00076-8

Khir, A. W., & Parker, K. H. (2005). Wave intensity in the ascending aorta: Effects of arterial occlusion. *Journal of Biomechanics*, *38*, 647–655.

Laugesen, E., Rossen, N. B., Peters, C. D., Mæng, M., Ebbehøj, E., Knudsen, S. T., Hansen, K. W., Bøtker, H. E., & Poulsen, P. L.

(2014). Assessment of central blood pressure in patients with type 2 diabetes: A comparison between sphygmocor and invasively measured values. *American Journal of Hypertension*, *27*, 169–176.

Li, Y., & Khir, A. W. (2011). Experimental validation of non-invasive and fluid density independent methods for the determination of local wave speed and arrival time of reflected wave. *Journal of Biomechanics*, *44*, 1393–1399. https://doi.org/10.1016/j.jbiomech.2010.12.019

McEniery, C. M., Cockcroft, J. R., Roman, M. J., Franklin, S. S., & Wilkinson, I. B. (2014). Central blood pressure: Current evidence and clinical importance. *European Heart Journal*, *35*, 1719–1725.

Meinders, J. M., & Hoeks, A. P. G. (2004). Simultaneous assessment of diameter and pressure waveforms in the carotid artery. *Ultrasound in Medicine and Biology*, *30*, 147–154. https://doi.org/10.1016/j.ultrasmedbio.2003.10.014

Millasseau, S. C., Patel, S. J., Redwood, S. R., Ritter, J. M., & Chowienczyk, P. J. (2003). Pressure wave reflection assessed from the peripheral pulse: Is a transfer function necessary? *Hypertension*, *41*, 1016–1020. https://doi.org/10.1161/01.HYP.0000057574.64076.A5

Park, C. M., Hughes, A. D., Henein, M. Y., & Khir, A. W. (2020). Mechanisms of aortic flow deceleration and the effect of wave reflection on left ventricular function. *Frontiers in Physiology*, *11*, 578701. https://doi.org/10.3389/fphys.2020.578701

Pauca, A. L., O'Rourke, M. F., & Kon, N. D. (2001). Prospective evaluation of a method for estimating ascending aortic pressure from the radial artery pressure waveform. *Hypertension*, *38*, 932–937. https://doi.org/10.1161/hy1001.096106

Picone, D. S., Schultz, M. G., Otahal, P., Aakhus, S., Al-Jumaily, A. M., Black, J. A., Bos, W. J., Chambers, J. B., Chen, C. H., Cheng, H. M., Cremer, A., Davies, J. E., Dwyer, N., Gould, B. A., Hughes, A. D., Lacy, P. S., Laugesen, E., Liang, F., Melamed, R., ... Sharman, J. E. (2017). Accuracy of cuff-measured blood pressure: Systematic reviews and meta-analyses. *Journal of the American College of Cardiology*, *70*, 572–586.

Rabben, S. I., Stergiopulos, N., Hellevik, L. R., Smiseth, O. A., Slørdahl, S., Urheim, S., & Angelsen, B. (2004). An ultrasound-based method for determining pulse wave velocity in superficial arteries. *Journal of Biomechanics*, *37*, 1615–1622. https://doi.org/10.1016/j.jbiomech.2003.12.031

Reference Values for Arterial Measurements Collaboration. (2014). Establishing reference values for central blood pressure and its amplification in a general healthy population and according to cardiovascular risk factors. *European Heart Journal*, *35*, 3122–3133. https://doi.org/10.1093/eurheartj/ehu293

Roman, M. J., Devereux, R. B., Kizer, J. R., Lee, E. T., Galloway, J. M., Ali, T., Umans, J. G., & Howard, B. V. (2007). Central pressure more strongly relates to vascular disease and outcome than does brachial pressure: The strong heart study. *Hypertension*, *50*, 197–203.

Segers, P., Mahieu, D., Kips, J., Rietzschel, E., De Buyzere, M., De Bacquer, D., Bekaert, S., De Backer, G., Gillebert, T., Verdonck, P., & Van Bortel, L. (2009). Amplification of the pressure pulse in the upper limb in healthy, middle-aged men and women. *Hypertension*, *54*, 414–420. https://doi.org/10.1161/HYPERTENSIONAHA.109.133009

Segers, P., Rietzschel, E., Heireman, S., Debuyzere, M., Gillebert, T., Verdonck, P., & Van Bortel, L. (2005). Carotid tonometry versus synthesized aorta pressure waves for the estimation of central systolic blood pressure and augmentation index. *American Journal of Hypertension*, *18*, 1168–1173. https://doi.org/10.1016/j.amjhyper.2005.04.005

Segers, P., Swillens, A., Taelman, L., & Vierendeels, J. (2014). Wave reflection leads to over- and underestimation of local wave speed by the PU- and QA-loop methods: Theoretical basis and solution to the problem. *Physiological Measurement*, *35*, 847–861.

Sharman, J. E., Avolio, A. P., Baulmann, J., Benetos, A., Blacher, J., Blizzard, C. L., Boutouyrie, P., Chen, C.-H., Chowienczyk, P., Cockcroft, J. R., Cruickshank, J. K., Ferreira, I., Ghiadoni, L., Hughes, A., Jankowski, P., Laurent, S., McDonnell, B. J., McEniery, C., Millasseau, S. C., ... Vlachopoulos, C. (2017). Validation of non-invasive central blood pressure devices: ARTERY Society task force consensus statement on protocol standardization. *European Heart Journal*, *38*, 2805–2812.

Sharman, J. E., Marwick, T. H., Gilroy, D., Otahal, P., Abhayaratna, W. P., & Stowasser, M. (2013). Randomized trial of guiding hypertension management using central aortic blood pressure compared with best-practice care principal findings of the BP GUIDE study. *Hypertension*, *62*, 1138–1145. https://doi.org/10.1161/HYPERTENSIONAHA.113.02001

Spronck, B., Avolio, A. P., Tan, I., Butlin, M., Reesink, K. D., & Delhaas, T. (2017). Arterial stiffness index beta and cardio-ankle vascular index inherently depend on blood pressure but can be readily corrected. *Journal of Hypertension*, *35*, 98–104. https://doi.org/10.1097/HJH.0000000000001132

Spronck, B., Heusinkveld, M., Vanmolkot, F., Roodt, J. O., Hermeling, E., Delhaas, T., Kroon, A., & Reesink, K. (2015). Pressure-dependence of arterial stiffness: Potential clinical implications. *Journal of Hypertension*, *33*, 330–338.

Sugawara, M., Niki, K., Ohte, N., Okada, T., & Harada, A. (2009). Clinical usefulness of wave intensity analysis. *Medical & Biological Engineering & Computing*, *47*, 197–206. https://doi.org/10.1007/s11517-008-0388-x

Vennin, S., Mayer, A., Li, Y., Fok, H., Clapp, B., Alastruey, J., & Chowienczyk, P. (2015). Noninvasive calculation of the aortic blood pressure waveform from the flow velocity waveform: A proof of concept. *American Journal of Physiology. Heart and Circulatory Physiology*, *309*, H969–H976.

Wang, K., Cheng, H., Chuang, S., & Spurgeon, H. A. (2011). Central or peripheral systolic or pulse pressure: Which best relates to target-organs and future. *Journal of Hypertension*, *27*, 461–467.

Wang, K. L., Cheng, H. M., Sung, S. H., Chuang, S. Y., Li, C. H., Spurgeon, H. A., Ting, C. T., Najjar, S. S., Lakatta, E. G., Yin, F. C. P., Chou, P., & Chen, C. H. (2010). Wave reflection and arterial stiffness in the prediction of 15-year all-cause and cardiovascular mortalities: A community-based study. *Hypertension*, *55*, 799–805.

Willemet, M., Vennin, S., & Alastruey, J. (2016). Computational assessment of hemodynamics-based diagnostic tools using a database of virtual subjects: Application to three case studies. *Journal of Biomechanics*, *49*, 3908–3914.

Williams, B., Lacy, P. S., Thom, S. M., Cruickshank, K., Stanton, A., Collier, D., Hughes, A. D., Thurston, H., & O'Rourke, M. (2006). Differential impact of blood pressure-lowering drugs on central aortic pressure and clinical outcomes: Principal results of the Conduit Artery Function Evaluation (CAFE) study. *Circulation*, *113*, 1213–1225.

Williams, B., Mancia, G., Spiering, W., Agabiti Rosei, E., Azizi, M., Burnier, M., Clement, D. L., Coca, A., de Simone, G., Dominiczak, A., Kahan, T., Mahfoud, F., Redon, J., Ruilope, L., Zanchetti, A., Kerins, M., Kjeldsen, S. E., Kreutz, R., Laurent, S., ... Desormais, I. (2018). 2018 ESC/ESH Guidelines for the management of arterial hypertension. *European Heart Journal*, *39*, 3021–3104.

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