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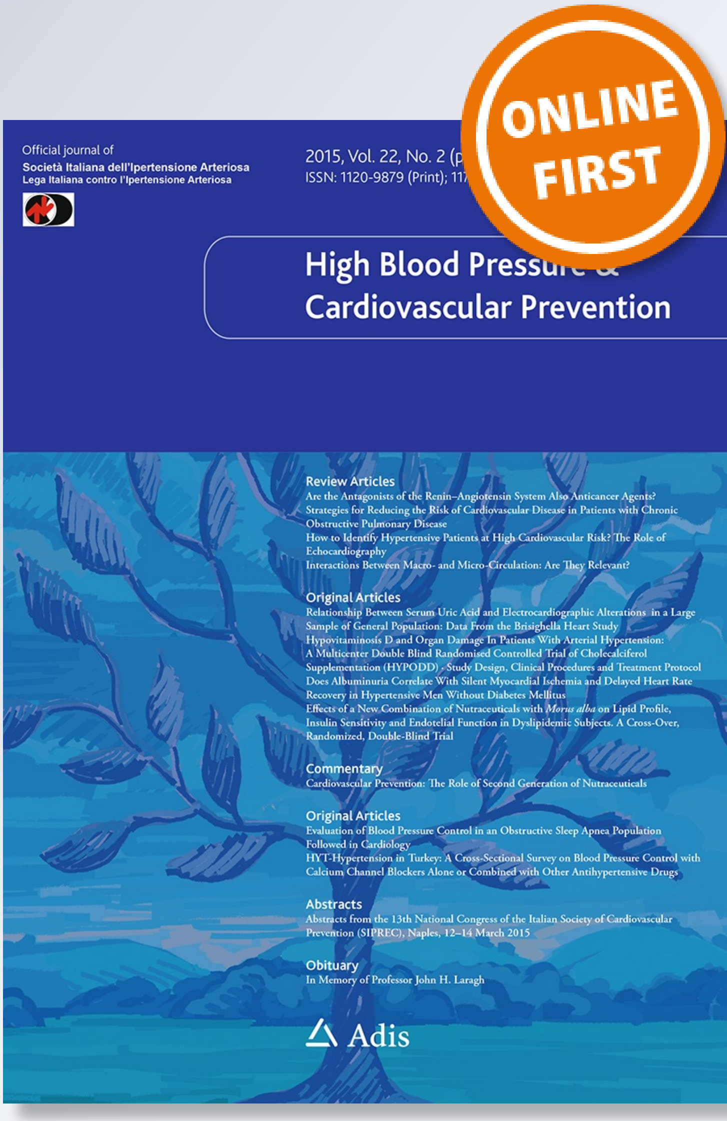

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ORIGINAL ARTICLE

Arterial Stiffness in a Rural Population of Argentina: Pilot Study

Alejandro Diaz[1] · Matías Tringler[1] · Cintia Galli[2] · Agustín Ramirez[2,3] · Edmundo Ignacio Cabrera Fischer[2]

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Abstract
Introduction Aortic stiffness evaluated through pulse wave velocity (PWV) measurement is nowadays accepted as a reliable parameter to estimate cardiovascular risk. However the data about arterial stiffness in South America come from urban populations.
Aims To determine the relationship between PWV changes and ageing and to identify the rate of change in each decade of life.
Methods PWV was measured in the carotid-femoral pathway in 400 inhabitants of Vela town and clinical parameters were recorded.
Results The prevalence of hypertension, dyslipidemia and diabetes was 33.5, 17.5, 5 % respectively. PWV was positively correlated with age (r: 0.817, $p = 0.01$) and was greater after the fifth decade of life (9.72 ± 2.2 vs. 5.87 ± 1.55 m/s; p: 0.001) than in youngers than 50 years, respectively. The risk (odds ratio) of having PWV >10 m/s was higher in hypertensives (OR: 50, $p = 0.001$), older than 50 years (OR: 44, $p = 0.001$), diabetics (OR 9.5, $p = 0.001$) and dyslipidemic patients (OR: 5, $p = 0.001$).
Conclusions This is the first study in a rural population of Argentina which determines the relationship of PWV with age and cardiovascular risk. PWV shows a slower growth in subjects less than 50 years compared to older adults with a strong relationship to the process of arterial aging, the development of hypertension and cardiovascular risk.

Keywords Pulse wave velocity · Arterial stiffness · Epidemiology · Rural population

1 Introduction

The non-invasive assessment of arterial stiffness is part of the comprehensive assessment of target organ damage in hypertension and global cardiovascular risk profile [1–3]. The measurement of carotid to femoral PWV is now considered a useful tool for arterial stiffness assessment in daily practice because of its easy use, low cost and high reproducibility [4].

Arterial stiffness is actually recognized as a surrogate end point for cardiovascular disease [4, 5] and the PWV is nowadays accepted as a prognostic predictor in patients with uncomplicated essential hypertension [2] as well as in the general population [5, 6]. In this way PWV was included in European Guidelines [1] as one factor, other than office BP, influencing prognosis and used for stratification of total cardiovascular (CV) risk [7].

Relating reference values of PWV, they come mostly from multicenter registries of Asia [8], USA [5, 9] and Europe [3]. However, several studies in Latin America and Hispanic populations showed significant differences in the size, structure and arterial stiffness of large and small arteries [10–14]. In addition there are very few population-based studies that evaluate arterial stiffness in populations of South America [15, 16].

Finally, up to date, there are no data about values of PWV in rural population studies in Argentina involving

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adolescents, young adults and older people. Moreover, considerations about those factors influencing PWV values, such aging [3], are also lacking. Therefore, it is a priority, for public health, to generate data to have objective criteria to determine the normality or abnormality of PWV to accurately assess vascular function.

On the basis of these considerations, the aims of this work were: (1) to determine the reference values of carotid femoral PWV in a rural population from Maria Ignacia Vela, (2) to determine the relationship of carotid femoral PWV with aging and the rate of change of this parameter in each decade of life in a community-based study and (3) to compare PWV values between normotensive and hypertensive subjects in this rural population.

2 Methods

This is a sub-study of the Vela Project that is an epidemiological door to door study aimed at determining the prevalence of cardiovascular risk factors in a well characterized rural population. The present is a descriptive, observational and cross-sectional population-based study carried out in healthy people from the rural town of Maria Ignacia Vela. Maria Ignacia Vela is a rural town located 50 km from Tandil, Argentina (37°40′00″S 59°15′00″W) at an altitude of 100 m above sea level. The latest population data from the National Institute of Statistics and Census reported 1822 inhabitants in 2001.

The Vela Project had two phases. The first one was focused on evaluating cardiovascular risk factors in the elementary, middle and high school populations [17, 18]. The second phase consisted of BP and cardiovascular risk assessment in the adult population. The probabilistic sampling used in the present study guarantees the representativeness of all elements of the population. The sample size was calculated according to the national prevalence of arterial hypertension [19] of 34 %. The calculation indicates that a representative sample should be at least 290 inhabitants.

The population studied was categorized according to the age decade (Group 1: 0–9 years, Group 2: 10–19 years, Group 3: 20–29 years, Group 4: 30–39 years, Group 5: 40–49 years, Group 6: 50–59 years, Group 7: 60–69 years old, Group 8: 80–89 years old, Group 9: 90–100 years old).

Blood pressure measurement: Three blood pressure (BP) measurements were made in two consecutive visits with a mercury sphygmomanometer, with the patient seated after, at least, 10 min at rest. An individual was defined as normotensive when blood pressure (BP) was lower than 140/90 mmHg. Subjects with BP values equal or higher than the previously mentioned values or under treatment for arterial hypertension were considered as hypertensive.

Pulse wave velocity measurement: PWV measurements were performed using a validated technique, which is routinely used by our group. The validation analysis was performed as requested by the Artery Society Guidelines [20]. Data acquisition and pulse wave velocity calculation were performed using hardware and software previously reported (Arteriometer V100, Oxytech, Buenos Aires, Argentina) [21, 22] to compare the data obtained with the mentioned technology against those calculated through blood flow waves (Echo-Doppler).

Briefly, we studied 21 subjects in which PWV was randomly measured on the same acquisition session through both, blood flow waves with Doppler technique [23] (Esaote My Lab 40, using a 7 MHz probe) and the Arteriometer technique (Arteriometer V100, Oxytech, Buenos Aires, Argentina) using tonometry to record the arterial pulse wave profile as previously reported [21, 22]. Shortly, two high fidelity strain gauge mechano-transducers (Motorola MPX 2050, Motorola Inc., Schaumburg, Il, USA) were positioned over the carotid and femoral arteries to obtain the blood pressure waves profiles. After that, the distance (m) between both points was measured and the time between both wave feet (s) was obtained. With this data, the distance was divided by the transit time of the both blood pressure waves giving the so-called PWV value (m/s).

With the echography technique, the study was performed with the subject in supine position, the carotid artery, 1–2 cm before the bifurcation, and the femoral artery, at inguinal level, were identified. After that, the identification of the Doppler flow was done and both signals were synchronized with the EKG R wave signal. Six registers from both vascular areas with the same carotid-femoral distance measured were used to calculate PWV and the mean value reported as the patient PWV [23].

Comparative analysis: The mean PWV value obtained with the Doppler technique was 9.01 ± 3.29 m/s while that calculated using the Arteriometer was 8.78 ± 3.05 m/s ($p = 0.81$), being the difference 0.23 m/s. As shown in Fig. 1, the correlation between both measurements showed a good agreement ($R^2 = 0.99$). This was in accordance with the validation previously published comparing the Vicorder reproducibility of carotid-femoral pulse wave velocity to the SphygmoCor measurements [24] and the data published by Calabia et al. reporting the agreement of PWV values obtained with the Complior method against the Doppler ultrasound method [23].

The Bland and Altman relationship was also calculated. As shown in Fig. 2 a good agreement was observed in values between 6 and 12 m/s. This was in accordance with the validation previously provided by Stone et al. Additionally, our data gave further information since it was

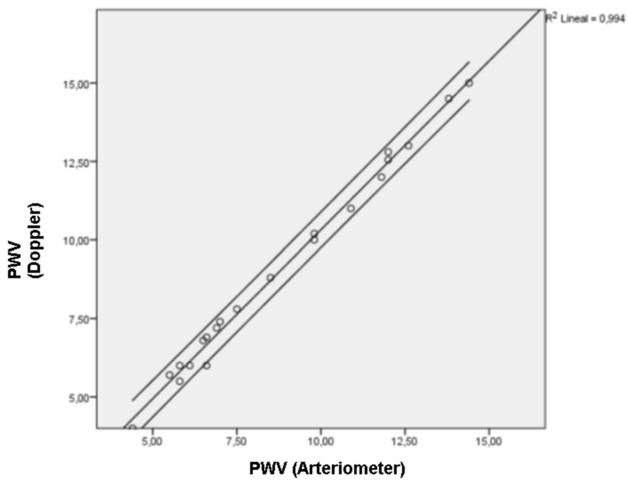


Fig. 1 Correlation between the pulse wave velocity (PWV) values measured by arteriometer method using two pulse arterial sensors and the ultrasonic pulse recording (Doppler)

shown that values ≤6 m/s, PWV were over-estimated while values higher than 12 m/s were under-estimated.

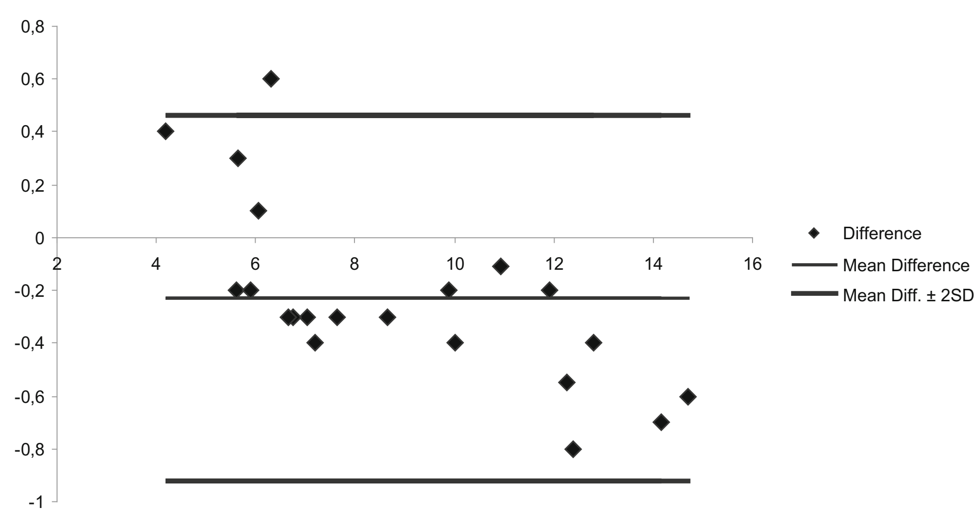
Measurement: Pulse wave velocity was obtained according to the technique previously published and developed in Favaloro University [21, 22]. Briefly, two high-fidelity silicon piezo-resistive pressure sensors (Motorola MPX 2050, Motorola Inc., Corporate 1303 E. Algonquin Road, Schaumburg, Illinois 60196, USA) were connected to an amplifier during data acquisition. The sensors were applied in two different sites of the same arterial pathway. The data was acquired in a computer with specific software that allowed obtaining the time delay between the two instantaneous arterial pulse waves obtained. The software works in a Windows environment performing an on-line digitized pressure wave acquisition that allows several PWV measurements along a single continuous record, which includes at least ten cardiac cycles. The same two physicians, one always obtaining the pressure waves and the other operating the computer, performed the data collection in order to avoid inter-operator differences. The operators monitored the quality of the pulse waves on-line and the acquisitions were repeated, if necessary. The software was able to calculate the PWV on-line, taking into account the distance measured between sensors. In this study, the sensors were positioned in (a) the carotid arteries and, (b) the femoral arteries, for carotid-femoral PWV evaluation.

Measurements were performed in a quiet room with stable room temperature after 10 min of rest with the patient in supine position. Measurements were obtained by duplicate [4]. The mean and the standard deviation of these measurements were always calculated and considered as the PWV value for each patient. In order to ensure a reliable measurement, special care was taken in monitoring that the standard deviation of measurements were less than 10 %.

According with international recommendations, the corrected PWV (PWVc) was calculated [3, 4] by multiplying by 0.8 [25], as suggested by the European recommendations [3, 4], and 10 m/s was considered as limit value for normal value.

Body mass index calculation: The weight was measured with the patients in the orthostatic position, with the arms extended along the body, barefoot and wearing light clothes. A stadiometer with a precision of 0.1 cm was used to measure height, with the participants standing barefoot. The body mass index (BMI) was calculated as weight in kilograms divided by the square of the height in square meters (kg/m^2). Normal weight was considered when BMI

Fig. 2 Bland-Altman plot showing the relationship between pulse wave velocity measurements obtained using two arterial pulse sensors and the ultrasonic pulse recording (Doppler)



was in-between 18.5 and 24.9 kg m^2. Overweight was defined when BMI was >25 and ≤29.9 and, finally, obesity was diagnosed when BMI was equal or higher than 30 kg/m^2.

Statistics: Measured and calculated values were expressed as mean value ± SD. A $p < 0.05$ was considered statistically significant. Differences among groups were tested by means of ANOVA and Bonferroni's post-test. The risk was analyzed by determining the multinomial logistic regression and calculating the odds ratio (OR). Comparative studies and quantification of risk were made with the Statistical Package for the Social Sciences 19.0 (Chicago, IL, USA).

Ethics approval: The protocol was approved by the "Ethics, teaching and Research Committee" from Santamarina Hospital, Tandil (resolution 0942/2010) and the postgraduate department of the Faculty of Medical Sciences, La Plata University, Buenos Aires, Argentina. Written consent describing the health examination and type of data collected was obtained from all participants.

Table 1 Population baseline characteristics according hypertensive and normotensive status

	Normotensive	Hypertensive	p
N	266 (66.5 %)	134 (33.5 %)	
Age (years)	32 ± 20.6	66 ± 13.7	0.001
BMI (kg/m^2)	23.3 ± 4	28.4 ± 5.4	0.001
SBP (mmHg)	116.9 ± 12	146.2 ± 21.8	0.001
DBP (mmHg)	67.1 ± 9.9	82.7 ± 13.4	0.001

BMI body mass index, *SBP* systolic blood pressure, *DBP* diastolic blood pressure

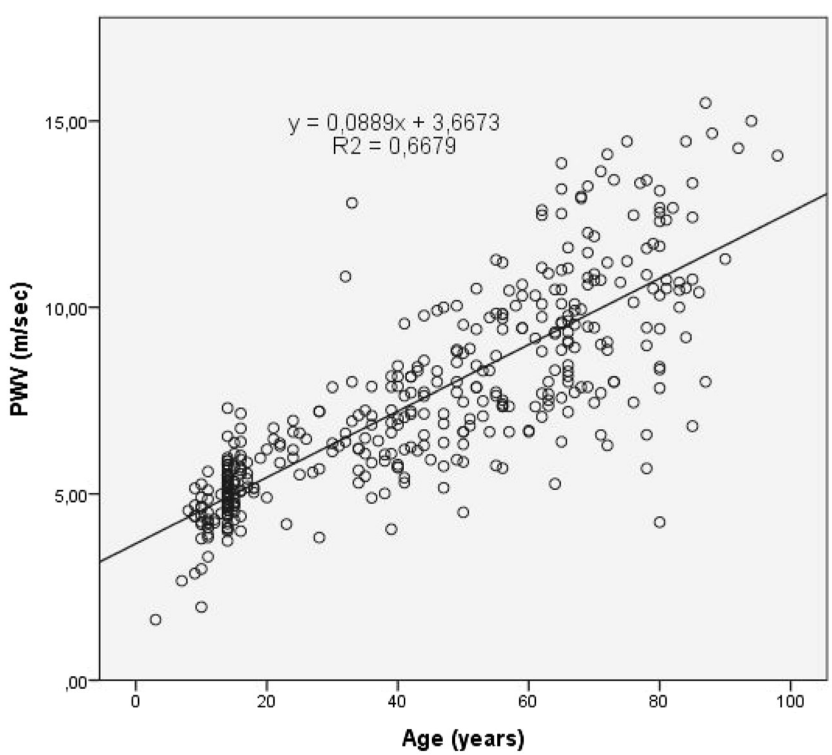
3 Results

In this study 400 individuals (208 males and 192 females) were evaluated. The mean age of the population was 43 ± 24.69 years (range 3–98) and the mean BMI was 25 ± 5.17 kg/m^2. The average height was 1.63 ± 0.12 m and weight of 67.5 ± 16.9 kg. The prevalence of hypertension, dyslipidemia and diabetes was 33.5, 17.5, and 5 % respectively. Only 5 % of the population had a family history of early cardiovascular disease and 44.7 % of the population used to perform regular physical activity.

As shown in Table 1 subjects with hypertension had higher mean age, BMI and, as expected, blood pressure than normotensive subjects. These differences remained significant independently of gender.

As it is shown in Fig. 3, carotid-femoral PWV showed a positive correlation with age (r: 0.817, $p < 0.01$). The slope of the fitted line showed a value of 0.089 m/s per year, representative of the PWV growth rate per year. Nevertheless, when the normotensive group was divided in decades of age (Fig. 2), the age dependent increase in

Fig. 3 Scatter graph showing the relationship between pulse wave velocity (PWV: in meters per second) and age in the study population (n = 400)



PWV was steeper in the groups over 50 years old. This was further evidenced by the fact that the mean PWV value in subjects under 50 years (5.87 m/s) was significantly lower than the observed in elders over 50 years (9.71 m/s; $p < 0.001$).

Figure 4 shows that PWV progressively increases with each decade of life; however this tendency is more pronounced after 50 years. In this way mean values of PWV showing significant differences between young subjects (≤50 years) and subjects >50 years (5.87 vs 9.72 m/s respectively, $p = 0.001$).

As gender and heart rates are related to PWV, linear regression analysis was performed in order to quantify the association among the measured and calculated variables. As it is shown in Table 2, the linear regression analysis including only females and considering the PWV as independent variable, showed a significant association between carotid-femoral PWV values and age ($p < 0.001$). Furthermore, a similar analysis showed a statistically significant relationship with heart rate ($p < 0.05$). On the contrary, no significant association was found with: systolic, diastolic and mean arterial pressure. When only males were included in the analysis, a significant association between carotid-femoral PWV values and age ($p < 0.001$) was found. No association was observed between PWV and heart rate ($p = 0.764$). Interestingly, in this cohort, PWV showed a significant association with systolic and mean arterial pressure ($p < 0.05$).

When considering the age related rate of PWV increase in subjects youngers than 50 years was 0.079 m/s per year, while in those over 50 years was 0.098 m/s per year (24 % increase in older than 50 years.

Finally, the risk of having hypertension after 50 years of age was 25.8 (OR 95 % CI: 14.4–46, $p = 0.001$). As shown in the multinomial regression analysis (Table 3) the population with an increased risk of having an abnormal high PWV were elderly patients with hypertension, diabetes and dyslipidemia ($p = 0.001$).

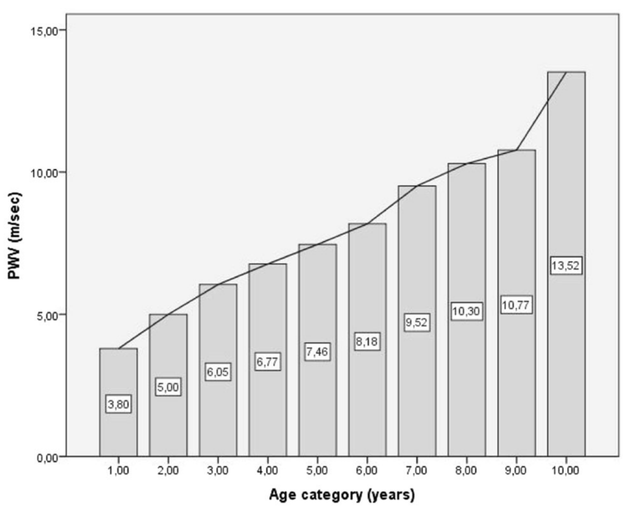


Fig. 4 Evolution of the mean values of the pulse wave velocity (PWV: in meters per second) in relation to age groups

Table 2 Linear regression analysis (n = 400), considering the carotid-femoral pulse wave velocity (m/s) as an independent variable

	Males (n = 208)		Females (n = 192)	
	R	Significance	R	Significance
Age	0.832	0.001	0.792	0.001
Heart rate	0.083	0.764	0.040	0.021
SAP (mmHg)	0.765	0.042	0.625	0.069
DAP (mmHg)	0.111	0.113	0.072	0.684
MAP (mmHg)	0.793	0.001	0.534	0.799

SAP, DAP and MAP are systolic, diastolic and mean arterial pressure; respectively. A $p < 0.05$ was considered statistically significant

4 Discussion

Several epidemiological studies argue that PWV is a direct measure of arterial stiffness and a marker of cardiovascular risk with predictive value over traditional risk factors [6, 26, 27]. That is because, in recent years, the determination of arterial stiffness, by measuring PWV, has been included in the screening of hypertensive patients for the evaluation of asymptomatic target organ damage [1, 2, 28].

Characteristically, arterial stiffness increases with age even in populations in which atherosclerosis is rare. Therefore it is necessary to differentiate the effects on PWV of disorders affecting the arterial wall other than the aging process [8].

In this way, Avolio and colleagues reported differences in arterial elasticity in urban and rural populations with different prevalence of hypertension and salt intake [29]. It has also been determined that the elasticity of the arterial wall is sensitive to changes such as those occurring in patients undergoing hemodialysis [21]. Recent studies of

Table 3 Risk evaluation to have an abnormal PWV value (>10 m/s)

	OR	CI 95 % (LL–UL)	p
Hypertension	50	20.9–121	0.001
Age >50 años	44.2	15.7–124	0.001
Diabetes	9.5	3.4–26.4	0.001
Dyslipidemia	5.0	2.8–5.8	0.001
FHCvD	1.9	0.72–5	0.181
Smoking	1.93	0.8–4.6	0.140

Calculated from odds ratio (OR) and multinomial logistic regression
LL lower limit, *UL* upper limit, *FHCvD* familiar history of CV disease

arterial elasticity of large vessels have shown differences between gender and ethnic groups [13, 14].

The present study was conducted in a rural population and represents the first phase of a cross-sectional study aiming to evaluate the prevalence of cardiovascular risk factors [17, 18].

The measurement of PWV is a validated method for the quantification of noninvasive arterial stiffness due to its simplicity, accuracy, reproducibility, and predictive value [28, 30].

The consensus on arterial stiffness of the European Society of Hypertension [4] set the measurement of carotid-femoral PWV as a standard method to quantify the arterial stiffness. This noninvasive method basically consists in sensing the arterial pulse profile in two different arterial sites of the same pathway. In addition, this consensus recommends a correction factor (0.8) of the measured distance considering the direct distance measured value times this factor with a new corrected PWV cut point set in 10 m/s. In this way, for the statistical analysis we used the corrected carotid-femoral distance (×0.8) which is the distance that has proven to be the best correlated with the "real" distance traveled by the pulse wave [25].

These results are coincident with previous reports, which stated that there is a strong association between PWV, age and hypertension [8, 29, 31]. Moreover, the contribution of other cardiovascular risk factors is smaller or not significant [31].

Our work is the first study in a rural population in Argentina which evaluates the relationship between PWV with age and cardiovascular risk. By this way, we think that it is possible to identify subjects at increased risk, based on the distribution of normal PWV values within each age group allowing an early detection of arterial damage or alteration.

To identify reference values in a population is an essential step before to incorporate PWV as a useful tool in the general clinic practice for early detection of target organ damage.

In South America, Farro et al. have established reference values for PWV from urban populations in Uruguay [15]. Furthermore, since there are ethnic, genetic and environmental differences in vascular structure and function in different populations it is not recommendable to translate these PWV values to other populations and/or countries [12, 16].

Definitions of the reference values in hypertensive subjects have clinical importance because it outline the existing temporal relationship between arterial stiffness and the development of hypertension. Recent data from the Framingham study showed that arterial stiffness precedes the development of hypertension and not "vice versa" [27]. This suggests that the real "silent murderer" could be arterial stiffness, so it is necessary to change the focus of attack and stimulate the development of interventions to modulate large-artery properties with the aim to have a major impact in the diagnosis and treatment of arterial hypertension and other pathologies associated with arterial wall damage.

In conclusion, this is the first study in a rural population of Argentina that evaluated the relationship of PWV with age and cardiovascular risk. The data provided, showed a slower PWV growth in subjects younger than 50 years compared to older. This behavior is related to the process of arterial aging, the development of hypertension and increased cardiovascular risk. This is a preliminary study to establish reference values of PWV for each age group and in the future this pilot study will contribute to establish normal values in this population considering the determinants of PWV.

Finally, we must mention that, at present, the only data on prognosis after PWV regression was obtained in a limited population of patients with end-stage renal failure. Therefore, the fact that PWV can predict mortality is a relevant argumentation favoring the assessment of PWV in the general practice. However, data from large studies in different populations are still needed.

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Conflict of interest There are no conflicts of interest to declare.

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