# Sex differences in arterial wave reflection and the role of exogenous and endogenous sex hormones: results of the Berlin Aging Study II

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**Background:** Arterial stiffness is tightly linked to hypertension. Sex differences in hypertension and arterial stiffness have already been established, yet the role of sex hormones is not precisely defined. This study examined age and sex differences of arterial wave reflection and associations with endogenous and exogenous sex hormones in women.

**Methods:** Pulse wave analysis was performed with an oscillometric device in 590 male and 400 female participants of the Berlin Aging Study II. Participants have been recruited from two age-strata, 22–35 years and 60–82 years. Data on exposures and potential confounders, including medication, have been collected at baseline visit.

**Results:** Aumentation index (AIx) and pulse wave velocity increased with age. Mean AIx was higher in women than in men. Multivariable regression analysis showed a positive association between use of oral contraceptive pills (OCPs) and AIx controlling for confounders (age, BMI, current smoking, central blood pressure), with a significantly higher mean AIx in OCP-users compared with nonusers (mean group difference: 4.41; 95% confidence interval 1.61–7.22). Per quartile decrease in estradiol level AIx increased by 1.72 (95% confidence interval 0.43–3.00). In OCP users endogenous estradiol was largely suppressed.

**Conclusion:** The findings suggest important sex differences in measures of arterial wave reflection, with a higher mean AIx observed in women compared with men. OCPs may promote the development of hypertension by increasing AIx. Suppressed endogenous estradiol levels may be responsible for this increased wave reflection due to increased vasotonus of the small and medium arteries.

**Keywords:** augmentation index, endothelial dysfunction, estradiol, HRT, oral contraceptives, prehypertension, sex and gender differences

**Abbreviations:** BP, blood pressure; CVD, cardiovascular disease; DHEA, dehydroepiandrosterone; HFpEF, heart failure with preserved ejection fraction; OCPs, oral contraceptive pills; PWA, pulse wave analysis; PWV, pulse wave velocity

## INTRODUCTION

Hypertension is a major public health issue, being the largest contributor to loss of global disability-adjusted life-years.

In both males and females, prevalence of hypertension and cardiovascular disease (CVD) increases with age. However, the development, pattern and end-organ damages of hypertension differ between the sexes. While the risk of young men (≤40 years) for hypertension is significantly higher than that of young women, with advancing age the risk approximates, and the relative risk of any CVD even gets reversed in older age.

It has been suggested that sex hormones are important in this context; however, the exact pathways are still poorly understood. A hallmark of hypertension is arterial stiffness. The relationship between arterial stiffness and blood pressure (BP) is currently understood as bidirectional. An increase in the vascular distension pressure causes an increase in arterial stiffness, and conversely, an increase in stiffness can lead to SBP elevation [1]. Commonly, stiffening occurs before hypertension becomes manifest, which makes stiffness an attractive 'target', for example in view of early recognition or for risk stratification [2].

Progressive stiffening of the arteries is an integral part of the normal ageing process [3]. Moreover, arterial stiffness is modulated by traditional risk factors, as there are sedentary lifestyle, smoking, obesity, high-cholesterol levels, glucose intolerance and nontraditional factors, such as renal

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impairment, rheumatoid disease and preeclampsia among others [4]. Indeed, there is some growing evidence that development and patterns of stiffness are differential in men and women and – just as hypertension and CVD – influenced by sex hormone status [5].

Pulse wave analysis (PWA) allows for the noninvasive assessment of the arterial status with the same ease as standard brachial BP measurement. The cuff-based oscillometric devices are now well validated [6,7] and the assessment of arterial stiffness is an established independent determinant of cardiovascular mortality in both sexes [8,9].

Whereas pulse wave velocity (PWV) is a direct measure of arterial stiffness, the augmentation index (AIx) – a measure of the augmentation of the antegrad systolic pulse wave by the reflected second wave – primarily reflects the vascular tone of the small and medium arteries and may be influenced by multiple factors for example sex hormones, the autonomic nervous system, timing of wave reflection, heart rate (HR), left ventricular function and height [10].

A permanent increase in arterial vascular stiffness is followed by an increase in central aortic BP [3], subsequently leading to organ damages such as myocardial diastolic dysfunction [11].

Sex and age differences in arterial stiffness and wave reflection have been ascribed mainly to the lower average height of women and sex differences in BMI. However, even in height-matched women and men it was shown that wave reflection occurred earlier during systole in women than it did in men [12]. This means that sex differences must exist beyond the known differences in average body height between men and women.

Sex hormone levels vary highly both interindividually and intraindividually and particularly between men and women. There are major alterations of sex hormone levels thoughout the life course, with more serious changes occurring in women (premenopause, perimenopause, postmenopause and pregnancy). In addition, in women administration of exogenous sex steroids in the form of oral contraceptive pills (OCPs) and postmenopausal hormone replacement therapy (HRT) is very common, and both represent major interferences with endogenous sex hormone levels.

Endogenous estrogen is considered largely protective for vascular function in women, and it has been suggested that, irrespective of the standard ageing process, the loss of estrogenic action in the arterial wall might play a particular, deleterious role, increasing arterial stiffness and reducing elasticity [13]. As to the impact of exogenously administered female sex hormones still there is not a complete and consistent picture. While both positive and negative effects on hypertension, CVD and arterial function have been shown in postmenopausal women, overall, postmenopausal HRT in women did not meet the expectations of a positive effect on cardiovascular mortality [14,15]. Likewise, there is evidence that OCP use may promote hypertension [15–17].

The aim of this study was to examine sex differences of arterial stiffness and wave reflection in a cohort of pre- and postmenopausal women and age matched men; and to estimate associations of using exogenous sex hormones [OCP and postmenopausal hormone therapy (HRT)] - with arterial pulse wave reflection (AIx) and PWV. We hypothesized that alterations of sex hormone levels may be associated with arterial stiffness as assessed by PWA.

# METHODS

## Study population

We performed a cross-sectional analysis of baseline data from the Berlin Aging Study II (BASE-II). BASE-II was launched to investigate factors associated with 'healthy' and 'unhealthy' aging and has been described previously in detail [18,19]. Briefly, BASE-II was recruited as a convenience sample from the greater Berlin metropolitan area by means of advertisements in local newspapers and the Berlin public transport system. In 2009–2014, 2172 participants were enrolled in the medical part of the study. Participants were aged 60–85 and 20–35 years, 75 and 25%, respectively, of total cohort. The group of young adults was conceived as an internal control group. All participants gave written informed consent and the study was approved by the Ethics Committee at Charité-Universitätsmedizin Berlin (EA2/029/09). PWA data were available from a subset of 990 participants (young group: $w = 115$, $m = 123$, old group: $w = 285$, $m = 467$) of BASE-II (Fig. 1).

## Pulse wave analysis

PWA was performed with the Mobil-O-Graph device (I.E.M. Germany, Stolberg, Germany) in a sitting position and relaxed athmosphere in a room as quiet as possible according to the international guidelines [20]. With an upper arm pressure sleeve oscillometric recording of the A. brachialis wave was analyzed. Peripheral BP measures were obtained and shape and amplitude of a central (aortic) BP curve were reconstructed via a mathematical transfer function. Subsequently measures of arterial stiffness, aortic PWV (aPWV) and AIx, were calculated [21]. For data quality assurance only measurements with good or very good quality were used based on the SD of pulse wave recording for a period of 8 s. Three consecutive measurements were performed and the average was used for further calculations.

PWV is generally accepted as the most simple, noninvasive, robust and reproducible method to assess arterial function [10]. It is defined as the time difference between the start of the first – forward – pulse wave and the beginning of the reflected wave. The distance used for

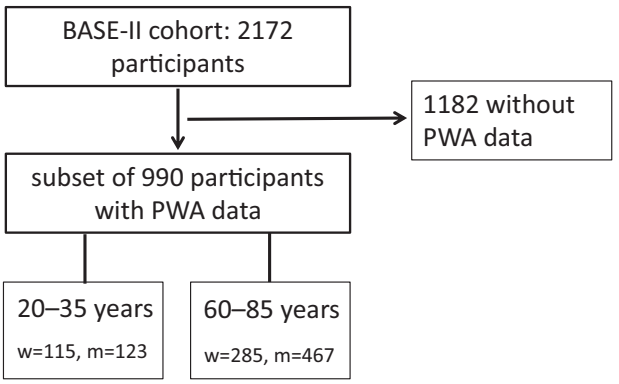


**FIGURE 1** Flow chart of sample selection.

the calculation from aortic arch to the aortic bifurcation depends on the body height and is divided by the reflection time/2.

The central AIx is defined as the difference between the first and the second systolic peak (P2 − P1) of the central arterial wave, expressed as a percentage of pulse pressure and based on the propagation time delay of the reflected wave [AIx% = ((P2 − P1)/PP) × 100]. AIx is a composite marker of wave reflections and arterial stiffness, which is determined by large artery stiffness, but also influenced by peripheral resistance (medium and small vessels), height, HR and sex.

Because AIx is influenced by HR, an index normalized for a HR of 75 bpm was used (AIx@75).

Peripheral vascular resistance (s mmHg/ml) was estimated by the Mobil-O-Graph.

## Covariates

All blood parameters were measured in a central certified laboratory using standardized protocols. Participants were asked to bring their medication plan and packets of all drugs used on a regular basis. Study staff took a comprehensive medication history (including indication, dosage, start and side effects). BMI was calculated as weight in kilograms divided by the square of the height in meters; waist and hip circumferences were measured in light clothes.

## Statistics

In descriptive statistics, values are expressed as percentages, mean ± SD or median and interquartile range. *t* Test or Kruskal–Wallis test were used to compare continuous variables, and the chi-squared test was used to compare proportions. Multivariable logistic and linear regression models were used to estimate adjusted associations of estradiol, OCP, HRT, and indices of arterial stiffness. Covariates in the final models were age, BMI, current smoking and central SBP. In one alternative logistic regression model, instead of BMI, height and waist circumference were included as covariates. Models were a priori stratified by age group (young, 20–35 years vs. old ≥60 years) and sex. The explanatory variables to be included in the multivariable regression models were identified both based on evidence that they acted as confounders of the association between the main exposure of interest and the outcome and did not lie on the causal pathway, and based on previous knowledge that they might act as a confounder. Estradiol levels were associated both with the outcome and the exposure of interest. However, since estradiol levels were assumed to be on the causal pathway between use of OCPs and AIx, we did not include OCP and estradiol together in one model. Effect modification by selected covariables was assessed by running stratified analyses and by including interaction terms (likelihood-ratio test). Pairwise correlation coefficients were assessed to identify potential multicollinearity. Statistical significance was set at *P* less than 0.05. We used StataCorp. 2017. Statistical Software: Release 15 (College Station, Texas, USA).

# RESULTS

Clinical characteristics of the sample are shown in Table 1. Since age is well known to be the major predictor of arterial stiffness, the sample was a priori stratified into a young group (22–35 years) and an old group (≥60 years) according to the inherent age distribution of the BASE-II cohort, and also stratified by sex for the purpose of assessing sex-differences.

On average AIx and aPWV increased with age. Furthermore, Table 1 shows significant age-specific sex differences of BP and measures of arterial stiffness. Among the young group of premenopausal women and men of the same age, women had significantly higher AIx values than men. Moreover, in the old group postmenopausal women showed significantly higher AIx values than men. In contrary women had consistently lower BP measures than men, both in the young (systolic and diastolic) and the old group (only diastolic).

Table 2 shows internal sex-specific and age-specific reference values, which were obtained from a subset of participants who did not use any regular medications. There was not a major discrepancy with the observations made in the total sample (Table 1).

Table 3 shows clinical and laboratory characteristics of premenopausal women stratified into an OCP user group

**TABLE 1. Clinical characteristics of the sample stratified by age group and sex**

| | Young group (22–35 years) | | Old group (60–82 years) | |
|---|---|---|---|---|
| | Men, $n = 123$ | Women, $n = 115$ | Men, $n = 467$ | Women, $n = 285$ |
| Age (years) | 29.5 ± 3.1 | 28.6 ± 3.1* | 68.7 ± 3.9 | 68.7 ± 3.3 |
| BMI (kg/m$^2$) | 23.9 ± 3.4 | 23.4 ± 3.3* | 27.3 ± 3.7 | 26.4 ± 4.8** |
| WC (cm) | 86.0 ± 9.5 | 79.3 ± 13.9** | 100.3 ± 10.1 | 91.5 ± 12.4** |
| Height (cm) | 181.1 ± 9.5 | 168.3 ± 6.5** | 175.6 ± 6.3 | 162.8 ± 6.0** |
| PWV (m/s) | 5.3 ± 0.4 | 5.1 ± 0.5** | 10.0 ± 0.9 | 10.0 ± 0.9 |
| AIx (%) | 9.7 ± 8.0 | 19.8 ± 8.3** | 20.4 ± 10.5 | 29.1 ± 10.1** |
| pSPB (mmHg) | 121.8 ± 10.9 | 114.9 ± 12.3** | 134.7 ± 16.4 | 133.9 ± 16.2 |
| pDBP (mmHg) | 76.7 ± 8.8 | 73.9 ± 9.2* | 86.3 ± 10.9 | 81.5 ± 11.1** |
| cSBP (mmHg) | 108.9 ± 10.5 | 104.7 ± 11.1* | 123.5 ± 15.7 | 123.3 ± 15.7 |
| PP (mmHg) | 43.8 ± 7.7 | 40.9 ± 8.6* | 47.8 ± 11.4 | 52.5 ± 11.5** |
| Current smoking, *n* (%) | 46 (37.40) | 39 (33.91) | 61 (13.06) | 34 (11.97) |

Data are presented as mean ± SD or numbers (percentages). AIx, augmentation index; PWV, pulse wave velocity; WC, waist circumference, pSBP/pDBP/cSBP: peripheral systolic/diastolic/central SBP, PP: pulse pressure; of 990 observations values were missing in current smoking (1), height (1), WC (2).
*$P < 0.05$.
**$P < 0.001$ (for comparison between women and men of the same age group).

**TABLE 2. Mean augmentation index and aortic pulse wave velocity of participants without any regular medication**

| | PWV (ms) | | AIx (%) | |
|---|---|---|---|---|
| | Men | Women | Men | Women |
| Young group, $n=114$ | 5.3 ± 0.4 | 5.1 ± 0.6 | 9.7 ± 9.0 | 17.9 ± 8.9 |
| Old group, $n=176$ | 10.1 ± 1.0 | 9.9 ± 1.1 | 21.8 ± 8.2 | 30.8 ± 8.1 |

AIx, augmentation index; PWV, pulse wave velocity; young males $n=81$, young females $n=33$, old males $n=123$, old females $n=53$, data are presented as mean ± SD.

**TABLE 3. Clinical and laboratory characteristics of premenopausal women according to use of oral contraceptive pills**

| | Use of oral contraceptive pills | | |
|---|---|---|---|
| | No, $n=64$ | Yes, $n=51$ | *P* value |
| Age (years) | 29.1 ± 3.2 | 28.1 ± 2.8 | 0.080 |
| PWV (ms) | 5.1 ± 0.6 | 5.1 ± 0.4 | 0.782 |
| AIx (%) | 17.6 ± 8.1 | 22.6 ± 7.6 | 0.001 |
| pSBP (mmHg) | 114.0 ± 12.9 | 116.1 ± 11.6 | 0.358 |
| cSBP (mmHg) | 103.8 ± 11.2 | 106.0 ± 11.1 | 0.287 |
| pDBP (mmHg) | 73.1 ± 9.1 | 74.9 ± 9.3 | 0.282 |
| Heart rate (bpm) | 72.1 ± 12.3 | 73.0 ± 9.3 | 0.613 |
| Height (cm) | 168.8 ± 6.9 | 178.6 ± 5.9 | 0.330 |
| WC (cm) | 80.1 ± 13.9 | 78.2 ± 14.0 | 0.461 |
| BMI (kg/m$^2$) | 23.5 ± 5.2 | 23.3 ± 5.5 | 0.886 |
| LDL-C (mg/dl) | 96.0 ± 29.7 | 92.9 ± 28.7 | 0.578 |
| Glucose (mg/dl) | 81.4 ± 7.0 | 79.1 ± 7.2 | 0.084 |
| Current smoking | 19 (26.7) | 20 (40) | 0.249 |
| Antihypertensive drugs | 1 (1.6) | 2 (4.0) | 0.420 |
| Estradiol (pmol/l) | 234.4 (123.0–487.6) | 22 (<18.4–58.0) | <0.001 |
| Testosterone (ng/ml) | 0.46 ± 0.24 | 0.31 ± 0.17 | <0.001 |
| DHEA (ng/ml) | 2257 ± 1107.7 | 1571.8 ± 680.0 | <0.001 |
| SHBG (nmol/l) | 68.3 ± 44.2 | 191.0 ± 105.2 | <0.001 |

AIx, augmentation index; cSBP, central SBP; DHEA, dehydroepiandrosteron; LDL-C, LDL cholesterol; pDBP, peripheral DBP; pSBP, peripheral SBP; PWV, pulse wave velocity; SHBG, steroid hormone binding globulin; WC, waist circumference; data are presented as mean ± SD, median (25–75th percentile), or numbers (percentages). Of 115 observations, values were missing in SHBG (6), DHEA (6), testosterone (6), estradiol (6), WC (1), height (1), weight (1) and BMI (1).

and an OCP nonuser group. There was strong evidence that the AIx was significantly higher in the OCP user group than in the nonuser group, whereas there was no difference in aPWV. The OCP user and non-OCP user groups were comparable as with potential confounding variables like age, height, weight, BMI or waist circumference, smoking or use of antihypertensive drugs. However, there was strong evidence of differences in median sex hormone levels with estradiol, testosterone and dehydroepiandrosteron (DHEA) being significantly lower and steroid hormone binding globulin (SHBG) being higher in the OCP-user group.

Regarding the group of postmenopausal women, there was no evidence of differences in measures of arterial stiffness, arterial wave reflecetion or BP between women with and without HRT (Supplementary Table 1, http://links.lww.com/HJH/B275). Yet median estradiol levels were significantly higher in women with HRT. Significantly, by trend, the overall cardio-metabolic profile appeared slightly more favourable in women with HRT (e.g. waist circumference 85.4 ± 8.9 vs. 91.9 ± 12.5 cm, $P=0.034$).

So far, the crude results suggested a strong positive association between using OCPs and AIx in premenopausal women, with a mean increase of AIx of 5.07 associated with using OCPs (coefficient = 5.07, 95% confidence interval (CI) 3.14–8.00, $P=0.001$; Table 4).

To adjust for potential confounding, we calculated multivariable regression models, stratified for sex and age group (Tables 4 and 5). There was reasonable evidence of a positive association between the use of OCPs and AIx after adjusting for important confounders (age, BMI, current smoking and central SBP). On average, OCP use was associated with an increase of 4.41 (95% CI 1.61–7.22, $P=0.002$) in AIx in premenopausal women. In an analogous manner, there was evidence of a negative association of measured estradiol levels and AIx. Per quartile decrease in estradiol level AIx increased by 1.72 (95% CI 0.43–3.00, $P=0.009$) (Table 4).

Similarly, in a logistic regression model, with high AIx as the outcome of interest (an AIx above the upper tertile, >24%, was defined as 'high AIx'), use of OCPs was associated with significantly increased odds of a high AIx [odds ratio (OR) 3.19; 95% CI 1.27–8.00, $P=0.0013$] controlling for BMI, central SBP, current smoking and age (Table 5).

Moreover, peripheral vascular resistance was increased in OCP users compared with nonusers (Supplementary Table 4, http://links.lww.com/HJH/B275).

There was no evidence of associations between OCP use or estradiol levels with aPWV (Supplementary Table 3a, http://links.lww.com/HJH/B275).

In the postmenopausal women group, neither estradiol levels nor the use of hormonal replacement therapy were independently associated with AIx or aPWV (Supplementary Tables 2 and 3b, http://links.lww.com/HJH/B275).

**TABLE 5. Association of oral contraceptive pill-use and high augmentation index (highest tertile)**

| Adjustment | Odds ratio | 95% CI | *P* |
|---|---|---|---|
| Non (crude) | 2.93 | 1.30–6.59 | 0.009 |
| Adjusted[a] (model 1) | 3.19 | 1.27–8.00 | 0.013 |
| Adjusted[b] (model 2) | 2.54 | 1.01–6.37 | 0.046 |

Logistic regression model, high AIx, highest tertile of AIx; CI, confidence interval.
[a]Adjusted for BMI, cSBP, current smoking, age.
[b]Adjusted for age, cSBP, current smoking, height, waist circumference.

**TABLE 4. Association of measured estradiol levels and oral contraceptive pill-use with augmentation index as continuous outcome**

| | Crude | | | Adjusted[a] | | |
|---|---|---|---|---|---|---|
| Exposure of interest | Coefficient | 95% CI | *P* value | Coefficient | 95% CI | *P* value |
| Estradiol (quartiles) | −1.69 | −3.06 to −0.33 | 0.08 | −1.72 | −3.00 to −0.43 | 0.009 |
| OCP use | 5.07 | 3.14–8.00 | 0.001 | 4.41 | 1.61–7.22 | 0.002 |

Linear regression model, CI, confidence interval; OCP, oral contraceptive pill.
[a]Adjusted for BMI, central SBP, current smoking, age.

## DISCUSSION

The principal finding of this study was that young premenopausal women taking OCPs had significantly higher AIx compared with those without use of OCPs. This indicates increased wave reflection due to increased vascular tone of the small and medium peripheral arterial vessels in women using OCPs. There was reasonable evidence that OCP use increased the odds of having a high AIx (above the upper tertile) by 3.2-times, controlling for important potential confounders (adjusted OR = 3.19, 95% CI: 1.27–8.00).

In premenopausal women taking OCPs endogenous estradiol (E2) levels were significantly suppressed when compared with the OCP nonuser group [median E2 (pmol/l) with OCPs: 22.0 (<18.4–58.0), without OCPs: 234.4 (123.0–487.6), $P < 0.001$]. Likewise, testosterone and DHEA levels were reduced, while SHBG was increased, with higher median levels measured in the OCP user group (Table 3). Multivariable linear regression analysis provided evidence of a negative association of estradiol E2 and AIx (coefficient = −1.72, 95% CI −3.00 to −0.43; $P = 0.009$; Table 4), whereas there was strong evidence that using OCPs on average was associated with an increase in AIx (coefficient = 5.07, 95% CI 3.14–8.00; $P = 0.001$).

To our knowledge, this is the first study to show associations of both the use of OCPs as well as measured estradiol levels with arterial wave reflection assessed by the AIx.

Findings of previous studies that have dealt with oral contraception, menstrual cycling and arterial stiffness have been highly inconsistent. This is likely to be due to different methods and devices used and parameters studied (for example aPWV, carotid-femoral PWV etc.) in the various studies.

For example, Yu *et al.* [22] showed that current OCP-use was associated with significantly increased aortic and peripheral SBP but not with increased arterial stiffness measured by aPWV. Another study recently demonstrated no differences in arterial stiffness (cf-PWV) between naturally menstrual cycling women and women who used monophasic OCPs [23]. Indeed, a recent subgroup analysis of the ENIGMA study found that use of the OCP was associated with increased large artery stiffness (aPWV) in young women [24]. No publication so far has dealt with contraception and AIx.

Another aspect that should be considered is the different timing and pattern of the age-associated increase of AIx and PWV: AIx is particularly alterable and tends to increase especially between the 2nd and 5th decade, which corresponds to the young group in this study, whereas PWV only starts to increase from the 5th decade onwards. According to this, in contrast to the situation in premenopausal women, we have found no evidence of associations of estradiol levels, or HRT with arterial wave reflection (AIx) in the postmenopausal women group in the current study.

PWV largely reflects structural arterial stiffness, mainly of the large arteries, whereas AIx is a combined measure, which is determined by large artery stiffness as well, but also influenced by peripheral resistance (medium and small vessels), height, HR and sex.

Given a high level of structural stiffness in higher age groups, it appears unlikely that rather discrete changes on a functional level (e.g. due to hormonal status) may be reflected by significant changes in AIx. In a similar vein the protective effect of estrogen is attenuated in vessels with advanced atherosclerosis [25]. In addition to the reduced compliance of vessels, absence, downregulation or dysfunction of the estrogen receptors on vascular and myocardial tissues is being discussed in this context.

The fact that there was no evidence of an association between HRT and measures of arterial stiffness and ware reflection in postmenopausal women is also in line with the failure of HRT to prevent cardiovascular endpoints in women starting HRT in late postmenopause (e.g. Women's Health Initiative trials) [26].

Consistent with other studies there was some suggestion that HRT was associated with favourable impact on cardiovascular risk factors like body fat distribution measured by a lower waist circumference (abdominal fat) compared with the HRT-nonusers (Supplementary Table 1, http://links.lww.com/HJH/B275) [27].

OCPs have been suspected of causing or promoting hypertension, given that several studies have shown an increase in BP associated with OCP use [16,22,28]. There are various pathways that have been suggested through which OCPs may impact cardiovascular disease, including activation of the renin–angiotensin–aldosterone system, inflammation, sympathic activation, glucose tolerance, amongst others [15].

So far, a link between exposure to OCPs, suppressed estradiol, arterial stiffness and hypertension has not been proposed. The present results suggest that oral contraception is associated with decreased endogenous sex steroids, in particular E2, and lower E2 is in turn associated with a higher AIx. It should be noted that OCPs used by premenopausal women in this study in 94% contained a combination of ethinylestradiol and progestogens. Only 6% of total 116 premenopausal women used pure progestin formulations for oral contraception.

There is a lot of previous evidence available on the effects of endogenous E2 (17ß-estradiol), and exogenous estradiol on endothelial cells and vascular smooth muscle cells. There is consensus that endogenous estrogen is rather protective for the vascular system with predominantly vasodilating effects in female sex [29]. It has been shown that disruption of ovulatory cycling, indicated by estrogen deficiency, or irregular menstrual cycling in premenopausal women is associated with an increased risk of coronary atherosclerosis and adverse cardiovascular events, respectively [30].

Moreover, the fact that loss of female sex hormones after menopause leads to a striking increase in the incidence of cardiovascular morbidity and mortality emphasizes this fact. There is good evidence that loss of estradiol during menopause reduces arterial distensibility, which may be mitigated by estradiol-substitution, but only during menopause transition [31].

Noteworthy, in addition to the AIx the peripheral vascular resistance, a hemodynamic parameter estimated by the Mobil-O-Graph, was increased in OCP-users compared with nonusers (Supplementary Table 4, http://links.lww.com/HJH/B275).

As another major result, the current study could confirm previously reported sex-differences of arterial wave reflection, both in young and in relatively healthy older adults. Sex differences in the timing of arterial wave reflection beyond differences in body height have been shown before [32], and it has been shown that the situation still worsens with the menopause [12,33].

The ENIGMA study group made the same observations and recently proposed a predominantly vascular phenotype of hypertension in women, which is characterized by elevated peripheral vascular resistance, aortic PWV, and AIx, contrasting with a cardiac phenotyope of hypertension in men. Importantly, they could show that this phenomenon, when observed in prehypertensive individuals was indeed associated with the development of manifest hypertension. This predominantly vascular phenotype of hypertension in women, that is women's higher arterial stiffness and wave reflection may contribute to their greater susceptibility to develop heart failure with preserved ejection fraction (HFpEF) [11,34]. In fact, there is evidence of an inverse relationship of arterial stiffness with parameters of diastolic function [35].

Based on our findings and the discussed results of others, we propose that the observed sex differences may be attributed to sex and sex hormone vascular interactions. The results suggest that women have genuinely higher wave reflection than age-matched men. During premenopause, this is counteracted by the effect of sex hormones, in particular estradiol. However, when the protective hormone effect drops out, with menopause, the unfavourable vascular phenotype may lead to hypertension with even more end organ damage, like diastolic dysfunction and HFpEF, being in fact more common in women than in men of the same age [32]. Furthermore, the use of OCPs may aggravate this phenomenon, withdrawing protective hormones during premenopause. Therefore, it is very likely that the higher AIx measured in this study population among young women using OCPs, will have an impact on future health outcomes.

Current ESC guidelines on hypertension (2018) estimate that about 5% of OCP-users develop a sustained increase in BP. Since it is important to identify women at risk of developing hypertension later in life early noninvasive PWA may be an ideal screening tool to be used at initiation and during follow-up of therapy with OCPs. Women with high AIx could be advised to use an alternative form of contraception, or at least more closely controlled.

Moreover, identification of different phenotypes of hypertension may allow better targeting of antihypertensive therapy according to the predominant underlying hemodynamic phenotype, for example peripheral vasodilators for female-type vascular phenotype [5].

### Strengths and weaknesses

The small number of study participants in certain subgroups limits the power of the study. Due to the cross-sectional design of this study we cannot make any assumptions as to causality. There may be residual unmeasured confounding, and confounding by indication cannot be ruled out. Further studies are needed with more participants and a prospective design.

The availability of PWA data and detailed medication data, and laboratory parameters, and medical history is strength of this study. On the contrary, progesterone – which was not measured in BASE-II – could not be considered in the analyses.

There is an ongoing discussion about the adequacy of the various methods for noninvasive assessment of aortic stiffness. Especially the estimation of aPWV with the Mobil-O-Graph, which was used in this study, and similar brachial cuff-based methods has been challenged [36]. Although there is no doubt that the Mobil-o-Graph provides reliable aPWV values, it is being criticized that the algorithm used yields estimates of aPWV which are mainly calculated from age and SBP, therefore do not provide additional prognostic information beyond that already supplied by these two risk factors [36]. For the current analysis this potential limitation is negligible. The main results relate to the AIx, which is not being challenged.

Part of this study was to show the sex and age distribution of AIx and aPWV in this population-based sample. In any case the Mobil-O-Graph is suited for that, and gives reliable results. Indeed, there was no evidence of differences e.g. between OCP or HRT users and nonusers. It would be interesting to see if there would be differences emerging if cf-PWV instead of aPWV was assessed. Anyway, the current study underlines that AIx is most useful in younger women and men without atherosclerotic alterations because it assesses the functional capacities of the vessel wall.

In conclusion, the findings suggest important sex differences in measures of arterial wave reflection. A higher mean AIx was observed in women compared with men. Furthermore, OCPs may promote the development of hypertension by interfering with endogenous sex hormone homeostasis in premenopausal women.

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Author contributions: U.S. and M.K. conceived of the study. U.S and M.K. analysed the data and developed the theory, M.K. performed the computations. U.S. and M.K. wrote the article. I.D. and E.S-T. provided the data and supervised the findings of this work together with V.R-Z. All authors discussed the results and contributed to the final article.

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### Conflicts of interest

There are no conflicts of interest.

## REFERENCES


1. Kotsis V, Stabouli S, Karafillis I, Nilsson P. Early vascular aging and the role of central blood pressure. *J Hypertens* 2011; 29:1847–1853.
2. Vlachopoulos C, Xaplanteris P, Aboyans V, Brodmann M, Cifkova R, Cosentino F, *et al*. The role of vascular biomarkers for primary and secondary prevention. A position paper from the European Society of Cardiology Working Group on peripheral circulation: Endorsed by the Association for Research into Arterial Structure and Physiology (ARTERY) Society. *Atherosclerosis* 2015; 241:507–532.
3. Celik T, Iyisoy A, Kursaklioglu H, Turhan H, Cagdas Yuksel U, Kilic S, *et al*. Impaired aortic elastic properties in young patients with prehypertension. *Blood Press Monit* 2006; 11:251–255.
4. Mikael LR, Paiva AMG, Gomes MM, Sousa ALL, Jardim P, Vitorino PVO, *et al*. Vascular aging and arterial stiffness. *Arq Bras Cardiol* 2017; 109:253–258.
5. Nardin C, Maki-Petaja KM, Miles KL, Yasmin. McDonnell BJ, Cockcroft JR, *et al*. Cardiovascular phenotype of elevated blood pressure differs markedly between young males and females: the enigma study. *Hypertension* 2018; 72:1277–1284.
6. Hametner B, Wassertheurer S, Kropf J, Mayer C, Eber B, Weber T. Oscillometric estimation of aortic pulse wave velocity: comparison with intra-aortic catheter measurements. *Blood Press Monit* 2013; 18:173–176.
7. Milan A, Zocaro G, Leone D, Tosello F, Buraioli I, Schiavone D, *et al*. Current assessment of pulse wave velocity: comprehensive review of validation studies. *J Hypertens* 2019; 37:1547–1557.
8. Ben-Shlomo Y, Spears M, Boustred C, May M, Anderson SG, Benjamin EJ, *et al*. Aortic pulse wave velocity improves cardiovascular event prediction: an individual participant meta-analysis of prospective observational data from 17,635 subjects. *J Am Coll Cardiol* 2014; 63:636–646.
9. Niiranen TJ, Kalesan B, Larson MG, Hamburg NM, Benjamin EJ, Mitchell GF, *et al*. Aortic-brachial arterial stiffness gradient and cardiovascular risk in the community: the Framingham heart study. *Hypertension* 2017; 69:1022–1028.
10. Papaioannou TG, Oikonomou E, Lazaros G, Christoforatou E, Vogiatzi G, Tsalamandris S, *et al*. The influence of resting heart rate on pulse wave velocity measurement is mediated by blood pressure and depends on aortic stiffness levels: insights from the Corinthia study. *Physiol Meas* 2019; 40:055005.
11. Seeland U, Brecht A, Nauman AT, Oertelt-Prigione S, Ruecke M, Knebel F, *et al*. Prevalence of arterial stiffness and the risk of myocardial diastolic dysfunction in women. *Biosci Rep* 2016; 36:1–9.
12. Kim JY, Park JB, Kim DS, Kim KS, Jeong JW, Park JC, *et al*. Gender difference in arterial stiffness in a multicenter cross-sectional study: the Korean Arterial Aging Study (KAAS). *Pulse (Basel)* 2014; 2:11–17.
13. Regnault V, Thomas F, Safar ME, Osborne-Pellegrin M, Khalil RA, Pannier B, *et al*. Sex difference in cardiovascular risk: role of pulse pressure amplification. *J Am Coll Cardiol* 2012; 59:1771–1777.
14. Manson JE, Aragaki AK, Rossouw JE, Anderson GL, Prentice RL, LaCroix AZ, *et al*. Menopausal hormone therapy and long-term all-cause and cause-specific mortality: the women's health initiative randomized trials. *JAMA* 2017; 318:927–938.
15. Shufelt CL, Bairey Merz CN. Contraceptive hormone use and cardiovascular disease. *J Am Coll Cardiol* 2009; 53:221–231.
16. Chasan-Taber L, Willett WC, Manson JE, Spiegelman D, Hunter DJ, Curhan G, *et al*. Prospective study of oral contraceptives and hypertension among women in the United States. *Circulation* 1996; 94:483–489.
17. Woods JW. Oral contraceptives and hypertension. *Hypertension* 1988; 11:I11–15.
18. Gerstorf D, Bertram L, Lindenberger U, Pawelec G, Demuth I, Steinhagen-Thiessen E, *et al*. Editorial: subjective perceptions of memory functioning in old age – nature, correlates, and developmental trajectories. *Gerontology* 2016; 62:311–315.
19. Bertram L, Bockenhoff A, Demuth I, Duzel S, Eckardt R, Li SC, *et al*. Cohort profile: the Berlin Aging Study II (BASE-II). *Int J Epidemiol* 2014; 43:703–712.
20. Van Bortel LM, Duprez D, Starmans-Kool MJ, Safar ME, Giannattasio C, Cockcroft J, *et al*. Clinical applications of arterial stiffness, Task Force III: recommendations for user procedures. *Am J Hypertens* 2002; 15:445–452.
21. Nunan D, Wassertheurer S, Lasserson D, Hametner B, Fleming S, Ward A, *et al*. Assessment of central haemomodynamics from a brachial cuff in a community setting. *BMC Cardiovasc Disord* 2012; 12:48.
22. Yu A, Giannone T, Scheffler P, Doonan RJ, Egiziano G, Gomez YH, *et al*. The effect of oral contraceptive pills and the natural menstrual cYCLe on arterial stiffness and hemodynamICs (CYCLIC). *J Hypertens* 2014; 32:100–107.
23. Priest SE, Shenouda N, MacDonald MJ. Effect of sex, menstrual cycle phase, and monophasic oral contraceptive pill use on local and central arterial stiffness in young adults. *Am J Physiol Heart Circ Physiol* 2018; 315:H357–H365.
24. Hickson SS, Miles KL, McDonnell BJ, Yasmin. Cockcroft JR, Wilkinson IB, *et al*. Use of the oral contraceptive pill is associated with increased large artery stiffness in young women: the ENIGMA study. *J Hypertens* 2011; 29:1155–1159.
25. Mendelsohn ME. Protective effects of estrogen on the cardiovascular system. *Am J Cardiol* 2002; 89:12E–17E; discussion 17E–18E.
26. Manson JE, Chlebowski RT, Stefanick ML, Aragaki AK, Rossouw JE, Prentice RL, *et al*. Menopausal hormone therapy and health outcomes during the intervention and extended poststopping phases of the Women's Health Initiative randomized trials. *JAMA* 2013; 310:1353–1368.
27. Chen Z, Bassford T, Green SB, Cauley JA, Jackson RD, LaCroix AZ, *et al*. Postmenopausal hormone therapy and body composition – a substudy of the estrogen plus progestin trial of the Women's Health Initiative. *Am J Clin Nutr* 2005; 82:651–656.
28. Harvey RE, Hart EC, Charkoudian N, Curry TB, Carter JR, Fu Q, *et al*. Oral contraceptive use, muscle sympathetic nerve activity, and systemic hemodynamics in young women. *Hypertension* 2015; 66:590–597.
29. Dai Z, Zhu HQ, Jiang DJ, Jiang JL, Deng HW, Li YJ. 17beta-estradiol preserves endothelial function by reduction of the endogenous nitric oxide synthase inhibitor level. *Int J Cardiol* 2004; 96:223–227.
30. Clarkson TB. Estrogen effects on arteries vary with stage of reproductive life and extent of subclinical atherosclerosis progression. *Menopause* 2018; 25:1262–1274.
31. Shufelt C, Elboudwarej O, Johnson BD, Mehta P, Bittner V, Braunstein G, *et al*. Carotid artery distensibility and hormone therapy and menopause: the Los Angeles Atherosclerosis Study. *Menopause* 2016; 23:150–157.
32. Gatzka CD, Kingwell BA, Cameron JD, Berry KL, Liang YL, Dewar EM, *et al*. Gender differences in the timing of arterial wave reflection beyond differences in body height. *J Hypertens* 2001; 19:2197–2203.
33. Costa-Hong VA, Muela HCS, Macedo TA, Sales ARK, Bortolotto LA. Gender differences of aortic wave reflection and influence of menopause on central blood pressure in patients with arterial hypertension. *BMC Cardiovasc Disord* 2018; 18:123.
34. Russo C, Jin Z, Palmieri V, Homma S, Rundek T, Elkind MS, *et al*. Arterial stiffness and wave reflection: sex differences and relationship with left ventricular diastolic function. *Hypertension* 2012; 60:362–368.
35. Wang ET, Ku IA, Shah SJ, Daviglus ML, Schreiner PJ, Konety SH, *et al*. Polycystic ovary syndrome is associated with higher left ventricular mass index: the CARDIA women's study. *J Clin Endocrinol Metab* 2012; 97:4656–4662.
36. Salvi P, Scalise F, Rovina M, Moretti F, Salvi L, Grillo A, *et al*. Noninvasive estimation of aortic stiffness through different approaches. *Hypertension* 2019; 74:117–129.