Accuracy of Doppler Assessment of the Uterine Arteries in Healthy Girls for the Diagnosis of Pubertal Onset

Amanda Cheuiche

Universidade Federal do Rio Grande do Sul <https://orcid.org/0000-0002-2157-3989>

Letícia Guimarães da Silveira

UFRGS: Universidade Federal do Rio Grande do Sul

Gustavo Monteiro Escott

UFRGS: Universidade Federal do Rio Grande do Sul

Iara Regina Siqueira Lucena

HCPA: Hospital de Clinicas de Porto Alegre

Márcia Puñales

Grupo Hospitalar Conceição: Grupo Hospitalar Conceicao

Fabíola Costenaro

Moinhos de Vento Hospital: Hospital Moinhos de Vento

Cristiane Kopacek

UFCSPA: Universidade Federal de Ciencias da Saude de Porto Alegre

Leila Cristina Pedroso de Paula

HCPA: Hospital de Clinicas de Porto Alegre

Sandra Pinho Silveiro (✉ ssilveiro@hcpa.edu.br)

UFRGS: Universidade Federal do Rio Grande do Sul

Research Article

Keywords: pulsatility index, puberty, ultrasonography, Doppler

Posted Date: July 13th, 2021

DOI: <https://doi.org/10.21203/rs.3.rs-669604/v1>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License.

[Read Full License](#)

Version of Record: A version of this preprint was published at Endocrine on November 30th, 2021. See the published version at <https://doi.org/10.1007/s12020-021-02948-y>.

Abstract

Purpose: To evaluate the accuracy of the uterine artery pulsatility index (PI) for the diagnosis of pubertal onset in girls.

Methods: Cross-sectional study of girls with normal pubertal development. Puberty was diagnosed by the presence of Tanner breast development score ≥ 2 . All girls underwent pelvic ultrasound and Doppler imaging of the uterine arteries. We evaluated the uterine artery PI and uterine, endometrial, and ovarian measurements. We used ROC curves with cutoffs determined by Youden index for data analysis.

Results: We included 169 girls aged 5-16 years who underwent 202 pelvic ultrasound examinations. Prepubertal girls had a significantly higher mean PI (mean, 6.70; SD, 2.15) than girls in initial puberty (mean, 4.14; SD, 1.55) and in late puberty (mean, 2.81; SD, 1.05) ($P < 0.001$ for all comparisons), which reflects a progressive increase in blood flow to the uterus with the progression of puberty. ROC curve analysis showed that the PI was able to identify the onset of puberty with a mean area under the curve of 0.838 (SD, 0.04) ($P < 0.001$), and the PI cutoff point of 5.05 had a sensitivity of 77%, specificity of 85%, positive predictive value (PPV) of 92%, and accuracy of 79%. The combination of PI < 5.05 plus uterine volume $> 3.75 \text{ cm}^3$ had a sensitivity of 73%, specificity of 95%, PPV of 97%, and accuracy of 79% to detect initial puberty.

Conclusions: The significant reduction in the PI during pubertal development combined with increasing uterine volume can be a valuable, highly specific, noninvasive tool to confirm the onset of puberty.

Introduction

In the pediatric scenario, pelvic ultrasound is a rapid, low-cost, noninvasive method for the assessment of uterine development, ovarian volume, and for the detection of ovarian cysts and tumors [1]. The size and morphology of the uterus and ovaries are relatively stable during childhood, but during puberty the uterine fundus progressively increases in size and becomes wider than the cervix, thus assuming the typical pear shape observed in adult women [2–4]. The endometrial lining is then visible and varies according to the phase of the menstrual cycle [3]. Ovarian size also increases, and multiple cysts can be seen during each menstrual cycle, representing stimulated follicles [1].

Doppler ultrasound allows the assessment of utero-ovarian blood flow and flow impedance measurement in this vascular tree. The pulsatility index (PI), defined as the difference between peak systolic flow and end-diastolic flow divided by the mean flow velocity, reflects the impedance to blood flow in the vessel distal to the sampling point [5]. Considering that the proximal blood flow remains constant, an increase in distal impedance will result in an increase in PI; likewise, a decrease in distal impedance will result in a decrease in PI, which could reflect a progressive increase in blood flow to the uterus during puberty.

Few studies have evaluated flow velocity in the uterine arteries of healthy women at different ages and pubertal stages [5–9]. Most of these studies observed an increase in PI in the prepubertal stage and a

decrease during puberty [5, 7, 9]. One small study, however, found no significant differences in the assessment of vascular flow among girls at different pubertal stages [8].

The aim of this study was to evaluate the accuracy of the uterine artery PI for the diagnosis of pubertal onset in healthy girls, in comparison or in combination with the usual parameters of uterine and ovarian volumes.

Patients And Methods

Study population

We conducted a cross-sectional study of healthy girls with normal pubertal development who were examined by a pediatric endocrinologist. The examination included evaluation of the pubertal stage according to the Tanner classification and pelvic ultrasound with Doppler imaging of the uterine arteries.

The study protocol was approved by the Hospital de Clínicas de Porto Alegre Research Ethics Committee (GPPG number 2019 – 0468). The Standards for Reporting Diagnostic Accuracy Studies (STARD) guidelines were followed according to the latest protocol update [10].

Healthy girls were eligible if they had thelarche and pubarche after 8 years of age and no history or evidence of any uncontrolled systemic disease. The onset of puberty was diagnosed by the presence of Tanner breast development score ≥ 2 , as assessed by visual inspection and breast palpation. The Tanner stages for breast development are classified as follows: 1 (no glandular tissue), 2 (breast bud appearance), 3 (further enlargement of breast and areola), 4 (areola and papilla forming a secondary mound above the level of the breast), and 5 (mature stage, with projection of papilla only, due to recession of the areola) [11]. We divided the girls into 3 stages: prepuberty (Tanner breast development score 1), initial puberty (Tanner breast development scores 2 and 3), and late puberty (Tanner breast development scores 4 and 5). The exclusion criterion was the use of estrogen or progestogen for any purpose.

We recorded the participants' clinical data, including age of menarche, pubarche, and thelarche. We measured standing height using a stadiometer to the nearest 0.1 cm, and weight using a digital scale with 10 g of precision. We calculated body mass index (BMI) as weight (kg)/height (m²). We calculated the girls' target height according to the following equation [12]: $([\text{father's height cm} - 13 \text{ cm}] + \text{mother's height cm})/2$.

Ultrasound and Doppler examination

The same experienced pediatric radiologist performed transabdominal pelvic ultrasound with full bladder technique in all participants with an ultrasound transducer (Aplio 400, Toshiba; Aplio 300, Toshiba) using a convex (2–5 MHz) or microconvex (5–8 MHz) probe, according to the size of the participant. We used a 50-Hz filter to remove low-frequency signals produced by motion artifacts and adjusted the angle of insonation to obtain maximum color intensity, and 3 to 5 consecutive heartbeats were analyzed.

We recorded uterine and ovarian volumes and endometrial thickness. Uterine and ovarian volumes were calculated according to the following formula for ellipsoid volume: volume (cm³) = longitudinal diameter (cm) × transverse diameter (cm) × anteroposterior diameter (cm) × 0.5233. We obtained the Doppler signal from the right and left uterine arteries and calculated the uterine artery PI. The average value of both arteries was recorded, since no significant difference was observed between the right and left arteries. Each examination took an average of 15 minutes, plus 10 minutes for the delivery of the printed result.

We included an independent sample of 4 girls at different pubertal stages to assess intraobserver variability. The same operator performed 3 PI measurements during the ultrasound examination. We analyzed the coefficient of variation and intraclass correlation coefficient.

Statistical analysis

Results were presented as mean (SD) or median (range) unless otherwise stated. We assessed the normality of data distribution by using the Kolmogorov-Smirnov test. We used Student's *t* test or analysis of variance (ANOVA) to analyze parametric continuous variables, and one-way ANOVA or the Kruskal-Wallis test for comparisons involving different pubertal stages. We assessed correlations between PI and uterine and ovarian parameters with Spearman's correlation coefficient. Receiver operating characteristic (ROC) curves were used to determine the ability of PI and other ultrasound parameters to discriminate between initial puberty and prepuberty and the Youden index (sensitivity + specificity – 1) to determine the optimal cutoff values [13]. We compared the area under the curve (AUC) with the bootstrap test for 2 correlated ROC curves [14] and calculated sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), positive likelihood ratio (LR+), negative likelihood ratio (LR–), and accuracy for the cutoff points found.

We calculated the sample size to estimate the sensitivity and specificity of PI for the diagnosis of pubertal onset using the online version of the PSS Health tool [15]. Considering a 95% confidence level, desired confidence interval width of 20% using the Wilson method, outcome prevalence of 40%, and expected sensitivity of 83% and specificity of 94% as reported by Paesano et al. [9], a total sample size of 133 participants was deemed necessary.

We performed all analyses in SPSS, version 20, and R statistical software, version 3.6.2 (The R Project for Statistical Computing). We set the significance level at 5%.

Results

We included 169 girls aged 5 to 16 years (mean, 11.3; SD, 1.8) who underwent 202 pelvic ultrasound examinations with Doppler assessment (Fig. 1). Twenty-seven girls underwent pelvic ultrasound in more than 1 of the 3 pubertal stages.

The study flowchart is presented in Fig. 2. Mean ages of thelarche, pubarche, and menarche were 10.1 (SD, 1.2), 10.2 (SD, 1.2), and 12.1 (SD, 1.2) years, respectively. Table 1 summarizes the clinical

characteristics of the participants.

Table 1

Clinical characteristics and ultrasound measurements of the participants according to pubertal stage.

Characteristic	Prepuberty (N = 39)	Initial puberty (N = 92)	Late puberty (N = 71)	P*
Clinical characteristics				
Age at pelvic US (years)	9.3 ± 1.3	11.0 ± 1.4	12.7 ± 1.4	< 0.001
Age of thelarche (years)	-	10.2 ± 1.2	10.0 ± 0.9	0.077
Age of pubarche (years)	10.2 ± 1.3	10.3 ± 1.4	10.2 ± 0.9	0.836
Age of menarche (years)	-	-	12.1 ± 1.2	0.196
Age of maternal menarche (years)	12.4 ± 2.7	12.9 ± 1.4	12.7 ± 1.6	0.440
Prematurity, %	6 (15)	13 (14)	10 (14)	0.400
Weight (kg)	32.0 ± 5.0	39.8 ± 9.0	48.9 ± 12.1	NA
Height (cm)	133.6 ± 7.2	143.8 ± 8.9	154.0 ± 7.7	NA
Height-for-age z-score	-0.12 (-1.14; 0.56)	-0.06 (-0.87; 0.58)	-0.24 (-0.43; 0.48)	0.524
BMI (kg/m ²)	17.9 ± 2.4	19.2 ± 3.5	20.6 ± 4.5	NA
BMI z-score	0.60 (-0.50; 1.30)	0.61 (-0.50; 1.20)	0.64 (-0.60 to 1.20)	0.935
Overweight	11 (28)	26 (28)	17 (24)	0.212
Obesity	0	3 (3,2)	2 (2,8)	0.134
Target height (cm)	162.0 ± 4.4	162.7 ± 5.7	161.0 ± 6.0	0.206
Ultrasound measurements				
Pulsatility index	6.70 ± 2.15	4.14 ± 1.55	2.81 ± 1.05	< 0.001
Uterine volume (cm ³)	2.60 (2.2; 3.6)	6.30 (4.4; 12.45)	34.3 (22.8; 47.6)	< 0.001
Uterine longitudinal diameter (cm)	3.35 ± 0.47	4.30 ± 0.83	6.24 ± 1.0	< 0.001

Data are presented as mean ± SD, median (IQR) or number of cases (%).

BMI: body mass index; NA: not applicable; US: ultrasound.

*P for comparison between the three groups.

Characteristic	Prepuberty (N = 39)	Initial puberty (N = 92)	Late puberty (N = 71)	P*
Endometrial thickness (mm)	0 (0; 1.3)	2.2 (1.2; 3.4)	6.0 (3.8; 9.1)	< 0.001
Right ovarian volume (cm ³)	1.64 ± 0.79	3.55 ± 1.98	7.16 ± 4.54	< 0.001
Left ovarian volume (cm ³)	1.62 ± 0.85	3.30 ± 1.82	6.03 ± 4.40	< 0.001
Mean ovarian volume (cm ³)	1.64 ± 0.77	3.43 ± 1.77	6.55 ± 3.76	< 0.001
Data are presented as mean ± SD, median (IQR) or number of cases (%).				
BMI: body mass index; NA: not applicable; US: ultrasound.				
*P for comparison between the three groups.				

Figure 3 shows the mean PI according to Tanner stage (panel A) and to study group (panel B). No differences were observed in mean PI measurements between Tanner 2 and 3 ($P = 0.939$) or between Tanner 4 and 5 ($P = 0.833$), and, therefore, the girls were grouped in 3 categories: prepubertal, initial and late puberty. Prepubertal girls had a significantly higher mean PI (mean, 6.70; SD, 2.15) than girls in initial puberty (mean, 4.14; SD, 1.55) and in late puberty (mean, 2.81; SD, 1.05) ($P < 0.001$ for all comparisons).

Table 1 shows the other ultrasound measurements of the participants according to pubertal stage. The uterine volume and longitudinal diameter, endometrial thickness, and ovarian volumes of prepubertal girls were significantly different from those of girls in initial and late puberty. Additionally, these measurements were also different between girls in initial puberty and those in late puberty.

ROC curve analysis showed that the PI was able to identify the onset of puberty with a mean AUC of 0.838 (SD, 0.04) ($P < 0.001$), which was comparable to the AUC of other ultrasound parameters (Fig. 4).

Table 2 shows the AUC, cutoff, sensitivity, specificity, PPV, NPV, LR+, LR-, and accuracy of PI and other pelvic ultrasound parameters. The PI cutoff point of 5.05 had a sensitivity of 77%, specificity of 85%, PPV of 92%, NPV of 61%, and accuracy of 79% to detect initial puberty. The combination of $PI < 5.05$ plus uterine volume $> 3.75 \text{ cm}^3$ increased the specificity (95%) and PPV (97%). The addition of ovarian volume $> 2.15 \text{ cm}^3$ decreased the sensitivity and did not increase the specificity or PPV.

Table 2

Diagnostic value of pulsatility index and ultrasound measurements for the diagnosis of pubertal onset.

Parameter	AUC	Cutoff	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	LR+	LR-	Accuracy (%)
Pulsatility index	0.838	5.05	77	85	92	61	5.13	0.27	79
Uterine volume	0.897	3.75 cm ³	81	79	90	65	3.86	0.24	80
Uterine longitudinal diameter	0.837	3.55 cm	84	68	86	63	2.63	0.24	79
Right ovarian volume	0.811	2.15 cm ³	78	77	89	60	3.39	0.29	78
Left ovarian volume	0.802	2.15 cm ³	76	77	89	58	3.30	0.31	76
Endometrial thickness	0.809	1.35 mm	75	79	90	57	3.57	0.32	76
PI and UV combined	0.866	PI 5.05; UV 3.75 cm ³	73	95	97	60	14.6	0.28	79
PI, UV, and OV combined	0.886	PI 5.05; UV 3.75 cm ³ ; OV 2.15 cm ³	66	95	97	54	13	0.36	75
AUC: area under the curve; LR+: positive likelihood ratio; LR-: negative likelihood ratio; OV: ovarian volume; PI: pulsatility index; PPV: positive predictive value; NPV: negative predictive value; UV: uterine volume.									

We found a strong negative correlation of PI with uterine volume ($r_s = -0.72$, $P < 0.001$) and a moderate negative correlation with endometrial thickness ($r_s = -0.68$, $P < 0.001$), uterine longitudinal diameter ($r_s = -0.68$, $P < 0.001$), and right ($r_s = -0.60$, $P < 0.0001$) and left ($r_s = -0.59$, $P < 0.001$) ovarian volumes.

The intraclass correlation coefficient for intraobserver agreement of the PI was 0.967 (95% confidence interval, 0.782–0.998; $P < 0.001$), indicating excellent reliability. The coefficient of variation was 12%.

Discussion

We found that girls who had already started puberty had a significantly lower Doppler uterine artery PI than prepubertal girls. The PI cutoff point of 5.05 had the highest accuracy in detecting initial puberty. The combination of PI below 5.05 plus uterine volume above 3.75 cm³ had the highest PPV (97%). Additionally, we identified significant negative correlations between PI and uterine volume, endometrial thickness, uterine longitudinal diameter, and ovarian volumes.

To the best of our knowledge, the present study is the first in Latin America to evaluate the contribution of Doppler assessment of the uterine arteries to the detection of signs of pubertal onset. Due to well-recognized distinct ethnic influences on pubertal development, it is of utmost importance to establish a pattern for each different population. A Danish study found a significant increase in the uterine artery flow velocity during puberty, with a decline in vascular resistance and PI in mid-puberty [5]. In contrast to our results, a significant decrease in the PI was found only after Tanner stage 3. Finally, an increase in the PI was detected in Tanner-5 girls and in adult women, possibly indicating the completion of angiogenesis [5]. Changes in the Doppler signal pattern of the uterine arteries was also identified in a Belgian study of girls aged 2 to 15 years [7]. Narrow systolic flow waves and a mean PI of 6.27 were observed in prepubertal girls, interrupted diastolic flow waves and a mean PI of 3.7 in girls in the beginning of external sexual development, and an uninterrupted signal during the diastolic interval at the end of puberty (mean PI of 2.0). Similar to our results, a negative correlation between PI and uterine and ovarian parameters was also observed [7]. The main mechanism proposed for the changes in vascular flow pattern is the existence of estrogen receptors on uterine arterial walls, which promotes a reduction in vascular resistance in the presence of hormonal stimulation [16, 17]. In contrast to the findings above, a study of girls from Tehran found no significant difference in uterine vascular flow between those without pubertal signs, those with signs of puberty but without menarche, and those after the beginning of menstruation [8]. However, the sample size was only 60 girls. A recent Italian study identified that a PI cutoff of 4.6 could differentiate pubertal from prepubertal girls with an accuracy of 87% and, when combined with a longitudinal uterine diameter of 35 mm, the accuracy increased to 91% [9]. Unlike our study, which used only the clinical parameter (Tanner breast stage) as a reference test for the diagnosis of the onset of puberty, a combination of clinical (Tanner stage), laboratory (luteinizing hormone peak after gonadotropin-releasing hormone [GnRH] stimulation) and ultrasound (longitudinal uterine diameter) criteria were used for the diagnosis of pubertal activation. Hormonal evaluation increased the reliability of the reference standard, but the criteria used led to the inclusion of girls with Tanner 2 and 3 in the prepubertal group [9]. A previous Italian study considered a PI below 2.5 indicative of a rapidly growing uterus in a sample of girls with signs of sexual precocity, with a 98.5% agreement with the GnRH stimulation test [18]. However, it did not include a control group of girls with normal pubertal development.

Endometrial thickness seems to be less studied than other ultrasound parameters. An Israeli study found a low sensitivity (57%) and a specificity of 100% for the presence of endometrial echo in differentiating central precocious puberty from isolated premature thelarche [19]. In contrast, a study of Chinese girls identified endometrial thickness as the best parameter (AUC of 0.933 with a cutoff point of 2.6 mm) to differentiate healthy prepubertal girls from those with central precocious puberty in the age group of 8 to

10 years [20]. In our study, the sensitivity of endometrial thickness was slightly lower than that of other ultrasound parameters, but with a specificity comparable to that of uterine volume.

The cutoff point for ovarian volume to identify the onset of puberty varies widely in the literature. A previous Brazilian study found a cutoff of 1.0 cm³ for ovarian volume, for which we found a higher value of 2.15 cm³ in the present study [21]. However, cutoffs of up to 4.4 cm³ have been reported [22]. The right and left ovaries had similar volumes in our study population, as previously described [21, 23, 24].

The limitation of our study was the unavailability of hormonal laboratory evaluation for confirmation of the onset of puberty. The strengths of our study include the large number of girls, exceeding the necessary calculated sample size, and the fact that all pelvic ultrasound examinations were performed by the same experienced pediatric radiologist using the same machine. Additionally, we strictly followed the recommended steps to analyze a diagnostic test, including STARD guidelines [10] and rigorous statistical analyses.

In conclusion, we confirm a significant reduction in the PI during pubertal development in our ethnic background, which reflects a progressive increase in blood flow to the uterus. Alone or in combination with uterine and ovarian volumes, the PI can be a valuable, highly specific, noninvasive tool to confirm the onset of puberty in the evaluation of pubertal disorders and possibly to assess the response to treatment in this set.

Declarations

Funding Sources: This work was supported by grants from Fundação de Amparo à Pesquisa do Estado do Rio Grande do Sul (FAPERGS), Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), Programa de Apoio a Núcleos de Excelência (PRONEX process number 16/2551-0000-476-5), Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES), Fundo de Incentivo à Pesquisa e Eventos (FIPE) Hospital de Clínicas de Porto Alegre.

Declaration of Interest: The authors have no conflicts of interest to disclose.

Availability of data and material: Data and material are available from the authors upon request.

Ethics approval: The study protocol was approved by the Hospital de Clínicas de Porto Alegre Research Ethics Committee (GPPG number 2019-0468).

Consent to participate: The researchers signed a term of commitment for the use of data collected from medical records. They also agreed that this information would be used solely and exclusively for the execution of this project. The pediatric endocrinologists have signed a document authorizing the use of data from their patients' medical records. Written informed consent was waived by the Institutional Review Board because of the retrospective nature of the study.

Acknowledgements: Hospital de Clínicas de Porto Alegre Research and Events Support Fund and Universidade Federal do Rio Grande do Sul Medical Science Program: Endocrinology.

References

1. Ziereisen F, Guissard G, Damry N, Avni E.F. Sonographic imaging of the paediatric female pelvis. *Eur. Radiol.* **15**,1296–309 (2005). DOI: 10.1007/s00330-005-2648-6.
2. de Vries L., Phillip M. Role of pelvic ultrasound in girls with precocious puberty. *Horm Res Paediatr* **75**,148–52 (2011). DOI: 10.1159/000323361.
3. Garel L., Dubois J., Grignon A., Filiatrault D., Van Vliet G. US of the pediatric female pelvis: a clinical perspective. *Radiographics* **21**, 1393–407 (2001). DOI: 10.1148/radiographics.21.6.g01nv041393.
4. Asăvoaie C., Fufezan O., Coșarcă M. Ovarian and uterine ultrasonography in pediatric patients. Pictorial essay. *Med. Ultrason.* **16**,160–7 (2014). DOI: 10.11152/mu.201.3.2066.162.ca1of2.
5. Mosfeldt Laursen E., Holm K., Brocks V., Jarden M., Müller J. Doppler assessment of flow velocity in the uterine artery during pubertal maturation. *Ultrasound Obstet. Gynecol.* **8**, 341–345 (1996). DOI: 10.1046/j.1469-0705.1996.08050341.x.
6. Long M.G., Boulton J.E., Hanson M.E., Begent R.H. Doppler time velocity waveform studies of the uterine artery and uterus. *Br. J. Obstet. Gynaecol.* **96**, 588–93 (1989). DOI: 10.1111/j.1471-0528.1989.tb03261.x.
7. Ziereisen F., Heinrichs C., Dufour D., Saerens M., Avni E.F. The role of Doppler evaluation of the uterine artery in girls around puberty. *Pediatr. Radiol.* **31**, 712–719 (2001). DOI: 10.1007/s002470100463.
8. Golestani R., Sheikhatvan M., Behpour A.M., Mehdizadeh M., Hajiashrafi A.A. Relationship between uterine and ovarian arterial blood flow measured by Doppler sonography at different stages of puberty. *Taiwan J. Obstet. Gynecol.* **47**, 62–65 (2008). DOI: 10.1016/S1028-4559(08)60056-X.
9. Paesano P.L., Colantoni C., Mora S. et al. Validation of an Accurate and Noninvasive Tool to Exclude Female Precocious Puberty: Pelvic Ultrasound With Uterine Artery Pulsatility Index. *AJR Am. J. Roentgenol.* **213**, 451–457 (2019). DOI: 10.2214/AJR.18.19875.
10. Cohen J.F., Korevaar D.A., Altman D.G. et al. STARD 2015 guidelines for reporting diagnostic accuracy studies: explanation and elaboration. *BMJ Open* **6**, e012799 (2016) DOI: 10.1136/bmjopen-2016-012799
11. Marshall W.A., Tanner J.M. Variations in pattern of pubertal changes in girls. *Arch. Dis. Child.* **44**, 291–303 (1969). DOI: 10.1136/adc.44.235.291.
12. Tanner J.M., Goldstein H., Whitehouse R.H. Standards for children's height at ages 2-9 years allowing for heights of parents. *Arch. Dis. Child.* **45**, 755–762 (1970). DOI: 10.1136/adc.45.244.755.
13. Youden W.J. Index for rating diagnostic tests. *Cancer* **3**, 32–35 (1950). DOI: 10.1002/1097-0142(1950)3:1<32::aid-cnrc2820030106>3.0.co;2-3.
14. Venkatraman E.S., Begg C.B. A distribution-free procedure for comparing receiver operating characteristic curves from a paired experiment. *Biometrika* **83**, 835–848 (1996).

15. Power and Sample Size for Health Researchers [Internet] https://hcupa-unidade-bioestatistica.shinyapps.io/PSS_Health/
16. Perrot-Applanat M., Groyer-Picard M.T., Garcia E., Lorenzo F., Milgrom E. Immunocytochemical demonstration of estrogen and progesterone receptors in muscle cells of uterine arteries in rabbits and humans. *Endocrinology* **123**, 1511–1519 (1988). DOI: 10.1210/endo-123-3-1511.
17. de Ziegler D., Bessis R., Frydman R. Vascular resistance of uterine arteries: physiological effects of estradiol and progesterone. *Fertil. Steril.* **55**, 775–779 (1991).
18. Battaglia C., Mancini F., Regnani G., Persico N., Iughetti L., De Aloysio D. Pelvic ultrasound and color Doppler findings in different isosexual precocities. *Ultrasound Obstet. Gynecol.* **22**, 277–283 (2003). DOI: 10.1002/uog.154.
19. de Vries L., Horev G., Schwartz M., Phillip M. Ultrasonographic and clinical parameters for early differentiation between precocious puberty and premature thelarche. *Eur. J. Endocrinol.* **154**, 891–898 (2006). DOI: 10.1530/eje.1.02151.
20. Wen X., Wen D., Zhang H., Zhang H., Yang Y. Observational study pelvic ultrasound a useful tool in the diagnosis and differentiation of precocious puberty in Chinese girls. *Medicine* **97**, e0092 (2018). DOI: 10.1097/MD.00000000000010092
21. Herter L.D., Golendziner E., Flores J.A. et al. Ovarian and uterine findings in pelvic sonography: comparison between prepubertal girls, girls with isolated thelarche, and girls with central precocious puberty. *J. Ultrasound Med.* **21**, 1237–1246 (2002). DOI: 10.7863/jum.2002.21.11.1237.
22. Badouraki M., Christoforidis A., Economou I., Dimitriadis A.S., Katzos G. Evaluation of pelvic ultrasonography in the diagnosis and differentiation of various forms of sexual precocity in girls. *Ultrasound Obstet. Gynecol.* **32**, 819–827 (2008). DOI: 10.1002/uog.6148.
23. Buzi F., Pilotta A., Dordoni D., Lombardi A., Zaglio S., Adlard P. Pelvic ultrasonography in normal girls and in girls with pubertal precocity. *Acta. Paediatr.* **87**, 1138–1145 (1998). DOI: 10.1080/080352598750031121.
24. Haber H.P., Wollmann H.A., Ranke M.B. Pelvic ultrasonography: early differentiation between isolated premature thelarche and central precocious puberty. *Eur. J. Pediatr.* **154**, 182–186 (1995). DOI: 10.1007/BF01954267.

Figures

A) Prepubertal girl (PI 9.8)

B) Pubertal girl (PI 2.15)

Figure 1

Color Doppler ultrasound of the uterine arteries. A) Prepubertal girl. B) Pubertal girl. PI, pulsatility index. Courtesy of Iara Lucena, MD.

Figure 2

Study flowchart. PI: pulsatility index; US: ultrasound

Figure 3

Mean PI according to Tanner stage (panel A) and study group (panel B). * $P < 0.001$ for comparison with stages 2, 3, 4, and 5. ** $P < 0.001$ for comparison with stages 4 and 5. *** $P < 0.001$ for comparison with stages 2 and 3. PI: pulsatility index.

Figure 4

Comparison between the AUC of ROC curves of PI, uterine volume, uterine longitudinal diameter, endometrial thickness, and right and left ovarian volumes. Diagonal line is the regression line. AUC: area under the curve; ROC: receiver operating characteristic; PI: pulsatility index.