The role of Doppler evaluation of the uterine artery in girls around puberty

France Ziereisen, Claudine Heinrichs, Dominique Dufour, Marco Saeres, Ephraim Freddy Avni

Received: 1 January 2001
Accepted: 3 February 2001

Presented at the 37th Annual Congress ESPR, Lisbon, Portugal, May 2000
Awarded the Jacques Lefèbvre Award of the ESPR

F. Ziereisen (25) · D. Dufour · E. F. Avni
Department of Radiology, Hôpital Universitaire des Enfants Reine Fabiola, Avenue J J Crocq, 15, 1020 Brussels, Belgium
e-mail: sky95576@skynet.be
Tel.: + 32-24-77.32.20
Fax: + 32-24-78.54.39

C. Heinrichs
Department of Paediatrics, Hôpital Universitaire des Enfants Reine Fabiola, Brussels, Belgium

M. Saeres
IRIDIA Laboratory, Université Libre de Bruxelles, Brussels, Belgium

Abstract Background. Presently, the only sonographic parameters used to follow puberty in girls are size and morphology of the uterus and ovaries. Doppler of the uterine artery appears a useful complementary parameter to follow puberty. Objective. To determine the potential contribution of Doppler evaluation of the uterine artery in girls around puberty. Materials and methods. We investigated 61 healthy female volunteers aged 2–15 years (mean 10.3 years). In each girl we performed a standard pelvic transabdominal US examination, including measurement of the uterus and ovaries. Uterine arteries were visualized by colour Doppler and a pulsed signal was obtained in each patient. The blood flow velocity waveform was analysed and the pulsatility index (PI) was calculated. Growth of the uterus and ovaries was plotted against age, and the PI was compared to each of the other studied variables (age, size of uterus, volume of ovaries). Results. We observed a strong negative correlation between the PI of the uterine artery and the usually studied variables. We also observed a progressive modification of the Doppler signal pattern of the uterine artery during the establishment of puberty: the narrow systolic flow waves found in prepubertal girls were progressively replaced by a systolic-diastolic flow wave. Conclusions. The demonstration of diastolic flow can confirm the onset of puberty.

Introduction

Pelvic US is used to evaluate the morphological development and growth of the internal genitalia, especially in cases of precocious or delayed puberty. Several studies have been published which provide normal reference data for uterine and ovarian sizes and morphology from birth to puberty. Doppler study of uterine blood flow is systematically used during pelvic US examinations in women, but to date this has not been the case in children. The aim of this study was to evaluate uterine perfusion in healthy girls before, during and after puberty and to correlate uterine blood flow changes with growth of the internal genitalia. The potential utility of Doppler analysis of the uterine artery in the assessment of pubertal abnormalities is also discussed.

Materials and methods

Sixty-one healthy female volunteers aged 2–15 years (mean 10.3 years) were studied during a 6-month period. Ultrasound examination was performed according to the full-bladder technique, using a 5-MHz, real-time sector scanner (Aspen, Acuson, Moun-
tain View, Calif.). Uterine and ovarian diameters were measured and ovarian volumes (calculated using the simplified formula for an ellipsoid: \( \text{longitudinal diameter} \times \text{transverse diameter} \times \text{AP diameter} \times 0.52 \)) were calculated. After the standard US examination, a Doppler study was performed in each girl; the uterine arteries were localised by colour Doppler and a pulsed Doppler signal was obtained from one or both arteries in each patient. Considering that there is no significant difference between right and left uterine artery blood flow [1], measurements were obtained only in the more accessible artery. The blood flow velocity waveform was analysed, and in each case the pulsatility index (PI) \( \text{PI} = (A-B)/\text{mean} \), where \( A \) is the maximum (systolic) Doppler frequency shift, \( B \) is the minimum (diastolic) frequency shift and mean represents the average Doppler frequency shift was calculated. Growth of the uterus and ovaries was plotted against age. The PI was compared to each of the other studied variables (age, size of uterus, volume of ovaries). Pubertal development was assessed using the usual Tanner classification. The average duration of each ultrasound examination was 15 min.

We used SAS/IMSP statistical software for analysing the data. Statistical analysis was performed using the Spearman non-parametric test and stepwise regression. \( P < 0.01 \) was considered statistically significant.

Written informed consent was obtained from each participant (if the girl was old enough to understand the aim of the study), and from one parent. The study was submitted and accepted by the Ethics Committee of our institution.

During the study period we followed four girls with precocious puberty (two with McCune Albright syndrome and two with idiopathic precocious puberty) before and during treatment. We compared the internal genitalia (size and morphology), the clinical data (vaginal bleeding) and the PI of the uterine arteries.

Results

Uterine size and ovarian volume were relatively stable up to the age of 9 years. A progressive, age-related increase in ovarian volume and uterine size started after the age of 9 years (Figs. 1–7).

The correlation between PI and the other studied variables (age and measurements of uterus and ovaries) are expressed in Figs. 8–15. In all the graphs (Figs. 1–15), the fitted curve is a second-order polynomial.

Our next step was to perform statistical analysis. Firstly, we performed a Spearman rank-correlation between PI and the other variables. All the correlations were highly significant \((P < 0.001)\), ranking from \(-0.71\) for correlation between right ovary and PI, to \(-0.46\) for correlation between uterine body width/cervix width ratio and PI. In decreasing order, the three variables that correlated most with PI were right ovarian volume \((-0.71)\), uterine transverse diameter \((-0.70)\) and uterine length \((-0.68)\). We can therefore say that PI is highly correlated with right ovarian volume, uterine transverse diameter and uterine length, the other variables ranking from \(-0.64\) to \(-0.46\).

Next we performed a stepwise linear regression with PI as the dependent variable. The stepwise regression selected uterine transverse diameter \((P < 0.001)\) and right ovarian volume \((P = 0.002)\) as independent variables. No other variable was selected. By fitting the linear regression we obtained a \(r^2 \) of 0.51, but the plot of predicted PI in terms of observed PI indicated a non-linear relationship. Consequently, we refitted a stepwise regression, including the second-order terms as well. The selected model contained uterine transverse diameter, right ovarian volume (as for the previously fitted linear model) as well as (uterine transverse diameter \times right ovary volume) \((P = 0.003)\). No other variable was selected. This was indicated that there is some evidence for an interaction between uterine transverse diameter and right ovarian volume. By fitting the model we obtained a \(r^2 \) of 0.55. Figure 16 is a plot of observed PI in terms of predicted PI.

The pattern of Doppler signal of the uterine artery displayed three types of flow waves: type 1 – narrow systolic flow waves without diastolic flow, type 2 – systolic flow waves with interrupted Doppler signal during diastole, and type 3 – broad systolic flow waves with uninterrupted flow during the diastolic interval.

Type 1 (Fig. 17; narrow systolic flow waves without diastolic flow) was observed in 24 girls. The mean uterine length was 3.15 cm (2.2–4.19 cm), mean ratio of body width/cervix width (RBC) was 0.95 (0.62–1.5), mean right ovarian volume (ROV) was 1.53 ml (0.24–3 ml) and mean left ovarian volume (LOV) was 1.37 ml (0.27–3.7 ml). The mean PI was 6.27 (3.5–8).

Mean age of this group was 7 years, 9 months (2–12 years). Nineteen of these girls did not present any external sexual development; 3 girls were evaluated S2P2, 1 girl S2P1 and 1 girl S3P2.

Type 2 (Fig. 18; an interrupted diastolic flow wave) was present in 11 girls. The mean uterine length was 3.46 cm (2.45–5.51 cm), mean RBC was 1.014 (0.733–1.46), mean ROV was 2.88 ml (0.15–6.9 ml), LOV was 3.09 ml (0.72–3.82 ml) and the mean PI was 3.7 (2.5–5). Mean age of this group was 11 years (9–12 years 3 months). Five of these girls were evaluated S1P1, 2 S2P2, 2 S3P2, 1 S3P3 and 1 had no evaluation of external pubertal development.

In 26 girls we found type 3, uninterrupted diastolic flow waves (Fig. 19). The mean uterine length was 5.83 cm (3.41–7.86 cm), mean RBC was 1.35 (0.95–1.82), mean ROV was 5.64 ml (2.87–11.45 ml), mean LOV was 4.93 ml (1.74–12.8 ml) and the mean PI was 2.055 (1.1–2.96). Mean age of this group was 12 years 9 months (10 years 8 months to 15 years 5 months). Twelve of these girls were evaluated S5P5, 1 S4PA, 2 S3P4, 4 S3P3, 1 S2P3, 1 S3P3, 3 S2P2 and 2 were not evaluated.

None of the girls in the first and second groups had menses at the time of the investigation. Half (13/26) of the girls of the last group already had menses at the time of the study.

We recalled 13 girls (6/11 of the girls who presented the second type of Doppler signal pattern and 7/13 of
Figs. 1-7 Uterine length, uterine transverse diameter, uterine body AP diameter, uterine cervix AP diameter, ratio body/cervix, right ovarian volume and left ovarian volume in relation to age.
Figs.8–15 PL in relation to age, uterine length, uterine transverse diameter, uterine body AP diameter, uterine cervix AP diameter, ratio body/cervix, right ovarian volume and left ovarian volume.
the girls who presented the third type of Doppler signal pattern and who presented no menses at the time of the study). None of the girls in the second group had begun menstruation (16 months later in five cases and 12 months later in one case). Three girls in the last group have developed menses 5 days, 9 months and 12 months, respectively, after the initial US examination. Four girls of the same group still do not have menses 16 months after the initial US examination.

All four children with precocious puberty presented, before treatment, with a systolic-diastolic flow wave similar to those seen in type 3, with a low PI. After treatment we have observed a modification of the wave, with narrowing of the systolic flow wave and a null or interrupted diastole (type 1 or 2). In case of failure of the treatment there was modification of the flow wave with appearance of a diastolic component and a decrease of the PI.

Discussion

Puberty is the period of transition between childhood and adulthood. It proceeds through five stages from childhood to full maturity (P1 to P5), as described by Marshall and Tanner [2]. These stages reflect the progressive modifications of the breast, the external genitalia and sexual hair. Although secondary sex characteristics appear at a mean age of 10\(\frac{1}{2}\) years in girls, pubertal changes may occur as early as 9 years or as late as 12 years. Puberty is considered precocious if these changes are noted before 8 years of age and delayed when such changes have not occurred by 13 years of age. Most girls reach menarche around 12–13 years of age; however, menarche may be observed as early as 10 years or as late as 15 years in otherwise normal girls.

Oestrogen receptors [3] have been identified in the wall of human uterine arteries. Oestrogens produce a decrease in the vascular resistance of these arteries. Physiological effects of oestrogen on the vascular resistance of human uterine arteries have been studied in postmenopausal women [4, 5] and in women with ovulatory cycles [6]. In postmenopausal women or in women with inactive ovaries, the uterine artery Doppler shows narrow systolic flow waves without diastolic flow. After hormonal treatment, the systolic flow waves broaden with an interrupted flow signal during diastole. All studies have shown an increase of the vascular resistance of the uterine artery in postmenopausal women, and the vascular resistance could be restored to premenopausal levels by administration of oestrogen. Addition of progesterone to oestrogen treatment has no effect on the vascular resistance.

These studies have shown that Doppler examination is a valuable reflector of biological efficacy of oestrogen treatment in postmenopausal women, an imaging modality which is readily accessible and non-invasive. The same studies were performed in ovulatory cycles where blood flow in the uterine arteries is subject to variations; in ovulatory cycles the resistance to flow is lowest prior to ovulation and in the midluteal phase. In anovulatory cycles, the resistance to flow is higher than in ovulatory cycles and there is no cyclical change. Good perfusion of the uterus during the midluteal phase seems to be crucial for successful implant, at least in stimulated cycles, and a few authors [7] prefer not to transfer an embryo if PI is > 3 in the midluteal phase. Goswamy et al. [8] and Goswamy and Steptoe [9] have shown that poor uterine perfusion in the midluteal phase reduces the...
success of in vitro fertilization, and hormone therapy with oestrogen may improve the perfusion and pregnancy rates.

To date, in girls, the only sonographic parameters used to follow puberty are size and morphological appearance of the uterus and ovaries. Only a few studies [10, 11] have been published about Doppler of uterine or ovarian arteries around puberty. These studies analysed the PI in relation to Tanner breast stages, age and ovarian volume. The first study compared ovarian volume and PI in girls with precocious puberty and in control subjects. They found no significant correlation between PI and the volume of ovaries and concluded that a larger number of cases was needed to evaluate the validity of Doppler US in girls with precocious puberty. The second study compared PI with the breast Tanner stage and with age. They found a significant decline of the PI in the mid-pubertal period.

The size and morphology of internal genitalia in prepubertal and post-pubertal girls are well described in the literature [12–15]:

- Before puberty, the size and morphology of the uterus and ovaries are relatively stable between 2 and 9 years. The volume of each ovary is invariably < 2 ml with small follicles < 9 mm; the length of the uterus is < 4 cm, the transverse diameter of the uterus is < 1.5 cm, the uterine body width and cervical width are < 1 cm and the ratio body width/cervix width is < 1.2.
- From 9 years of age and above, there is an increase in size of the uterus and ovaries. The uterine body becomes wider than the uterine cervix, producing the adult pear-shaped uterus with a ratio body width/cervix width > 1.2; the uterine length ranges between 5 and 8 cm, 1.6–3 cm in maximum AP diameter, and 3.5 cm in width. The endometrial canal is seen as a thin echogenic interface, and the endometrium demonstrates cyclical changes every 28 days. In response to gonadotrophin stimulation at puberty, the ovaries acquire a more ovoid shape and migrate deeper into the pelvis. The postpubertal ovary measures 2.5–5 cm long, 1.5–3 cm wide, and 0.6–1.5 cm thick, with a volume of 1.8–5.7 ml (mean 4 ml).

Our study confirms the accepted values for size and volume of the internal genitalia as reported in the literature; it also introduces a new parameter that could be used to follow puberty, namely Doppler study of the uterine artery. If we analyse the Doppler signal pattern of the uterine artery in our studied population, we observe three types of pattern:

The first type demonstrates narrow systolic flow waves without diastolic flow. This group corresponds to the prepubertal stage. Mean uterine length is 3.15 cm and the mean RCC is 0.95 cm. Ovarian volume is less than 2 ml (mean ovarian volume 1.45 ml). Furthermore, in this group the PI is high, above 4, with a mean PI of 6.27. In this group, the majority of girls (19/24) do not present external sexual development and none of these girls have menses.

The second type demonstrates interrupted diastolic flow waves. The mean uterine length is 3.46 cm, the RCC is close to 1 (mean RCC 1.01) and the ovarian volume is close to 3. PI decreases and the mean PI is 3.7. In this group, we observed the onset of external sexual development. No girl in this group had menses at the time of the pelvic US evaluation and none of the six girls whom we recalled had begun menstruation 12 months (n = 1) and 16 months (n = 5) later.
The third type demonstrates uninterrupted diastolic flow. In this group, the uterus presents a pubertal morphology with a mean length of 5.83 cm, a RCC more than 2 (mean RCC 1.35) and the ovarian volume is greater than 4 ml. PI is low, less than 3. In this group, half of these girls (13/26) had begun menstruation at the time of the evaluation. Sixteen months later, we recalled 7 of 13 girls who had not been menstruating: 4 girls still had no menses 16 months later and 3 girls had begun menstruation between 5 days and 12 months after the initial pelvic ultrasound examination.

The resistive index (RI) and pulsatility index (PI) are most commonly used to provide a numerical description of Doppler flow signals. RI = \((A-B)/A\) and PI = \((A-B)/\text{mean}\), where \(A\) is the maximum (systolic) Doppler frequency shift, \(B\) is the minimum (diastolic) Doppler frequency shift and 'mean' represents the average Doppler frequency shift. As RI was invariably equal to 1 when diastolic flow is null and as we have no diastolic flow before puberty, we have favoured PI for Doppler analysis. PI (or RI) is calculated from ratios of systolic and diastolic Doppler shift frequency and is not influenced by the angle made between the Doppler beam and the long axis of the vessel.

During puberty, we observed a decrease of PI with a progressive modification of the shape of the uterine artery flow waves. First there was the appearance of an interrupted signal during the diastolic interval and later the diastolic signal became uninterrupted as seen in ovulatory cycles. The PI calculated in our patients presented a strong negative correlation with the other classical variables (age, size of uterus, volume of ovaries). PI therefore appears to be a useful complementary parameter to monitor pubertal changes and confirm correct oestrogenic impregnation.

In our study, the length of uterus did not appear to be a good parameter to evaluate puberty. In prepubertal girls (with Doppler pattern type 1), the average uterine length was 3.15 cm; 3.46 cm at the beginning of puberty (when Doppler pattern type 2 was observed) and 5.83 cm when an uninterrupted diastolic flow wave was present (Doppler pattern type 3). There was little difference between the uterine length at the prepubertal stage and at the beginning of puberty.

The RBC and mean ovarian volume (MOV) appear to be more useful. Before puberty, RBC is less than 1 and MOV is less than 2 ml. At the onset of puberty RBC is about 1 and MOV about 3 ml; and finally at the end of puberty RBC is more than 1.2 and MOV is more than 4 ml.

The average PI in prepubertal girls is 6.7. At the onset of puberty the PI decreases but stays above 3 (average PI of 3.7), and at the end of puberty the PI is less than 3. RBC, MOV and PI appear to be the most useful parameters to follow puberty in girls.

Sonographic analysis of the morphology of the internal genitalia is usually sufficient to confirm or exclude pubertal development. Doppler appears useful in doubtful cases, for example in cases of delayed puberty if the uterus presents intermediate size and morphology. In these cases, the existence of diastolic flow can confirm the onset of puberty.

Use of the PI and study of the uterine artery flow wave appear even more useful for the follow-up of girls with precocious puberty. The study by Jensen et al. [16] in girls with idiopathic central precocious puberty reveals that with adequate treatment (long-acting gonadotrophin-releasing hormone analogues), several girls conserved ovarian and uterine volumes greater than in age-matched control subjects.

In our small series, the modifications of PI appear more sensitive and of greater prognostic significance than modifications of uterine size or the development of an ovarian cyst. In two girls (aged 2 years and 7 years) who presented with McCune Albright syndrome, we have seen normalisation of the uterine arterial flow wave after treatment, despite the persistence of pubertal morphology of the uterus. One of these girls experienced vaginal bleeding with an ovarian cyst while being treated, and the Doppler was of pubertal type at that time and normalised after intensification of treatment. We have had a similar experience with two sisters who presented with idiopathic precocious puberty. Doppler study of the uterine arteries appears a useful parameter in the follow-up of these girls because modifications of PI precede modifications in size and morphology of the uterus and ovaries and allow treatment modification before appearance of clinical manifestations.

**Conclusion**

During prepubertal and pubertal periods we observe progressive modifications in size and morphology of the uterus and ovaries. Concomitantly, we see a change in the Doppler signal pattern of uterine artery flow – narrow systolic flow waves (as seen in postmenopausal women without hormone-replacement therapy) are found in prepubertal girls; they are progressively replaced by a systolic-diastolic flow wave with an uninterrupted signal during the diastolic interval at the end of the pubertal development. Statistical studies reveal a strong negative correlation between PI and all the other variables (age, size of uterus and ovaries). Doppler study of the uterine artery seems to be of particular value in the follow-up of girls with precocious puberty where modifications of the Doppler signal pattern of the uterine artery appear earlier than size and morphological changes of the internal genitalia.
References