ORIGINAL ARTICLE



Test-retest reliability of internal pudendal artery blood flow using color Doppler ultrasound in healthy women

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Abstract

Introduction and hypothesis The internal pudendal artery (IPA) is one of the main arteries supplying the pelvic floor muscles (PFMs) and vulvo-vaginal tissues. Its assessment with color Doppler ultrasound has been documented previously, but the reliability of IPA measurements has never been assessed. This study evaluates the test–retest reliability of IPA blood flow parameters measured by color Doppler ultrasound under two conditions: at rest and after a PFM contraction task.

Methods Twenty healthy women participated in this study. One observer performed two measurement sessions using a clinical ultrasound system with a curved-array probe on the participant's gluteal area. IPA measurements were repeated: at rest and after a PFM contraction task. Peak systolic velocity (PSV), time-averaged maximum velocity (TAMX), end-diastolic velocity (EDV), pulsatility index (PI), and resistance index (RI) were measured. Test–retest reliability was assessed using a paired *t* test, intraclass correlation coefficient (ICC), and Bland and Altman plots.

Results There was no significant difference for all IPA blood flow measurements between the two repeated sessions. At rest, reliability was excellent for PSV and TAMX and the variability between measurements, as per Bland and Altman plots, was small. After PFM contractions, reliability was excellent for PSV and TAMX and Fair to good for PI. The variability between measurements was small for PSV and acceptable for TAMX and PI. EDV and RI parameters did not perform as well. **Conclusion** The assessment of IPA blood flow with color Doppler ultrasound to evaluate vascular change in women is reliable.

 $\textbf{Keywords} \ \ Color \ Doppler \ ultrasound \ \cdot \ Imaging \ \cdot \ Internal \ pudendal \ artery \ \cdot \ Pelvic \ floor \ muscle \ exercises \ \cdot \ Women$

Introduction

The internal pudendal artery (IPA) is one of the main vessels supplying the pelvic floor muscles (PFMs) and vulvo-vaginal tissues. After entering the gluteal region, IPA curves around the ischial spine and enters the perineum through the lesser sciatic

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foramen [1]. Then, it travels through the pudendal canal and provides blood by its branches to the PFM, perineum, labia, vagina, clitoris, and rectum [1]. Its investigation with color Doppler ultrasound has previously been reported [2, 3]; however, to our knowledge, no study has assessed the test-retest reliability of the IPA blood flow measurements with color

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Doppler ultrasound. As there is no standardized measure of the vulvo-vaginal blood flow, this assessment could provide important information about blood flow parameters in women with lower, or challenged, perineal vascularity; specifically, in those from different populations (young and aging women), with different conditions (genitourinary syndrome of menopause; lactational atrophic vaginitis; pudendal nerve entrapment; diabetes; atherosclerotic disease) or before/after interventions (perineal radiotherapy or surgery) [4–10]. Observation of perineal vascular changes after an intervention aimed at improving perineal vascularity, such as hormonal or laser therapy, could also be addressed [11, 12]. Hence, measurement repeatability must first be determined before clinical use, either for diagnosis or assessment of treatment response.

Changes in skeletal muscle artery blood flow have previously been evaluated with color Doppler ultrasound at rest and after muscle contraction tasks [13, 14]. Those two conditions provide information about the artery's vascular function at rest and its hemodynamic response to muscle activation (contraction task). In healthy adults, a skeletal muscle contraction task alters vascular tone in active muscles' vessels within seconds of the onset of muscle work, allowing a rapid improvement of muscle perfusion and, therefore, an increase in blood flow in the arteries related to the muscles [15]. Hence, as the IPA is the main blood flow provider of the PFMs, it is anticipated that a maximal voluntary PFM contraction task could change IPA blood flow. The assessment of test-retest reliability of the IPA blood flow at rest and after a PFM contraction task is therefore important to determine the level of blood flow variability before and after activation of the PFM.

Thus, the purpose of this study is to assess the test–retest reliability of the IPA blood flow measurements in healthy women using color Doppler ultrasound under two conditions: at rest and after a PFM contraction task.

Materials and methods

A convenience sample of healthy adult women was recruited for this prospective test-retest cohort study using e-mail advertisements to a group interested in pelvic floor rehabilitation in the Montreal area. Women were included if they were aged 18 years and older and were not actively participating in a PFM training program during the study. Participants were excluded if they were pregnant, had vulvo-vaginal atrophy or dermatological disease of the vulva, had previously received radiotherapy for gynecological cancer or were taking antiestrogenic medication. Further, hormonal therapy or arterial hypertension medication dosage had to be stable for 6 months to ensure perineal blood flow stability.

The study received ethical approval from the Institutional Review Board of the Institut universitaire de gériatrie de Montréal (Montreal, Canada). Each volunteer provided written consent before participation.

One observer (JM) performed two repeated measurement sessions of the IPA blood flow at time 1 (T1) and at time 2 (T2). The assessments were made 1 month apart, at the same time of the day (2 h apart), and during the same phase of the women's menstrual cycle, to control for the effects of circadian and hormonal rhythm on the pelvic blood flow [16]. Caffeine, tobacco intake, and physical activity such as cardiovascular, strength training or any physical activity likely to modify blood flow were restricted 2 h before the assessments, in addition to sexual activity 24 h before the assessments (T1 and T2) as they have been shown to influence blood flow parameters [17-20]. To confirm compliance with those recommendations, the observer questioned the participants about each of them before the measurements. Furthermore, to avoid potential changes in blood flow between assessment sessions, participants were asked not to perform any PFM training between T1 and T2.



Fig. 1 a Probe position (*red circle*). b Doppler waveform of the internal pudendal artery blood flow with parameter signification. S peak systolic velocity, D end-diastolic velocity

Table 1Subject demographics

Parameters	Data		
Mean age \pm SD (years)	33.5±10.1		
Parity (%)			
Nulliparas	11 (58)		
Multiparas	8 (42)		
Menopausal status (%)			
Pre-menopausal	17 (89)		
Post-menopausal	2 (11)		
Hormonal therapy (%)			
None	6 (32)		
Hormonal contraception	12 (63)		
Systemic hormonal therapy	1 (5)		

For the measurement sessions, participants were taught, using vaginal digital palpation, how to perform a maximal pelvic floor muscle contraction without compensation (gluteal, adductor, and abdominal muscle contraction) in the supine lying position with the knees bent. Then, they were asked to rest in the prone position for 15 min in a noiseless room with constant heat and light to ensure standardized conditions. After this waiting time, using a clinical ultrasound system (Voluson E8; GE Healthcare), the observer placed a 2- to 7-MHz curved-array probe on the participant's right gluteal area according to Kovac's procedure [2]. The probe was first placed in the transverse plane to identify the ischium. Then, by scanning in a sagittal plane, the ischial spine was identified to locate the IPA. which courses medially (Fig. 1). The internal pudendal vessels (artery and vein) follow recognizable anatomical landmarks that are well described in the literature and that permit unequivocal identification of these vessels. Further, the internal pudendal artery can be confidently distinguished from the vein by its pulsatility demonstrated by Doppler waveforms. IPA blood flow measurements were repeated three times at rest and three times

Table 2	Blood	flow	measurements	at	rest
i able z	Blood	now	measurements	aı	res

after a PFM contraction task (five 10-s maximal contractions followed by ten 1-s maximal contractions). Although studies examining the acute effect of exercises on local blood flow report conflicting results [21], endurance and high-intensity interval exercises tasks appear to show the most significant change in artery blood flow [21-23]. Therefore, our PFM contraction task was chosen to target these parameters (endurance and highintensity interval exercises). At each measurement session, five blood flow parameters giving an overall picture of artery vascularization were collected for the two conditions using the pulsed Doppler mode: peak systolic velocity, time-averaged maximum velocity, end-diastolic velocity, pulsatility index, and resistance index. Peak systolic velocity value refers to the maximum velocity of blood flow during contraction of the heart ventricles. End-diastolic velocity value refers to the blood flow velocity at the end of the heart ventricles. Time-averaged maximum velocity value refers to the mean of maximum velocities averaged over a complete cardiac cycle (Fig. 1). The pulsatility index and resistance index are a calculated ratio reflecting peripheral resistance. The pulsatility index is defined as the difference between the peak systolic and minimum diastolic velocities divided by the mean velocity during the cardiac cycle. The resistance index is defined as the difference between the peak systolic velocity and end-diastolic velocity divided by the peak systolic velocity. Within a pulsed-wave Doppler recording, the clearest waveform was selected for analysis (Fig. 1).

A paired *t* test was used to detect statistical differences in IPA blood flow measurements between T1 and T2 for each the two conditions. Then, to assess the test–retest reliability of each blood flow parameter between T1 and T2, intraclass correlation coefficients (ICCs) and their 95% confidence intervals (CIs) were calculated using a two-way mixed model [24]. ICC values less than 0.4 were considered poor, 0.40 to 0.75 fair to good, and 0.75 to 1.00 excellent [25]. Finally, the Bland–Altman plots were used to analyze the degree of variability of parameters between T1 and T2 of each of the two

	Data				ICC	
	Mean T1 ± SD	Mean T2 ± SD	Mean difference (95% IC)	p value	ICC (95% CI)	p value
PSV (cm/s)	50.2 ± 13.2	49.3 ± 13.0	0.9 (-0.4 to 2.2)	0.178	0.99 ^a (0.97 to 1.00)	< 0.001
TAMX (cm/s)	9.2 ± 2.7	9.0 ± 2.6	0.1 (-1.2 to 1.5)	0.850	0.71 ^b (0.13 to 0.90)	0.014
EDV (cm/s)	1.0 ± 2.2	0.8 ± 2.1	0.2 (-0.6 to 1.1)	0.607	0.84 ^a (0.56 to 0.95)	0.001
PI	6.0 ± 2.5	5.7 ± 2.3	0.3 (-1.0 to 1.6)	0.605	0.64 ^b (-0.01 to 0.87)	0.027
RI	1.0 ± 0.1	1.0 ± 0.1	0.0 (-0.0 to 0.4)	0.738	0.67b (0.08 to 0.88)	0.018

PSV peak systolic velocity, TAMX timed-average maximum velocity, EDV end-diastolic velocity, PI pulsatile index, RI resistance index

^a Excellent

^b Fair to good

adequately visualized for the analysis (95% feasibility).

Quality was not sufficient for 1 out of 20 images owing to the inability to visualize the IPA because of the small size of the vessel. Eleven women were nulliparas, 8 were multiparas; 17

women were pre-menopausal, 2 women were post-menopausal; 12 were taking hormonal contraception and 1 was taking sys-

temic hormonal therapy. None had any unstable hormonal status

(menstrual cycle irregularities) or any unstable blood pressure

conditions. The mean difference and 95% limits of agreement between parameters were calculated [26].

Results

A total of 20 women aged 22–53 years (33.6 ± 9.9 years) were recruited. Among the data set, 19 out of 20 images were

Peak systolic velocity Time-averaged maximum velocity 10 9 Difference between T1 and T2 8 Difference between T1 and T2 +1.96 SD (5.9) 6 6-+1.96 SD 0 (5.2)4 С 0 3 C ര 8 00 0 0 2 0 Mean Mean ο Oh 0 (0.9) 0. (0.1) 0 0 -2 0 -3 -1.96 SD -1.96 SD -4 (-4.2)(5.0) -6 -6[.] 0 -8 -9 20 30 40 50 60 70 80 12 15 18 ģ 6 T1 and T2 mean T1 and T2 mean **End-diastolic velocity Pulsatility index** 0 8 6 1.96 SD Difference between T1 and T2 Difference between T1 and T2 6 (5.2) 0 4 4 +1.96 SD 0 0 (3.3)2 2 Mean C Mean 0 0 (0.3) 0 (0.2) 0 0 0 0 0 -2 0 -1.96 SD 0 -2 0 (-2.9)0 -1.96 SD -6 (-4.6) -8 ò 2 4 6 8 10 2 4 6 8 10 T1 and T2 mean T1 and T2 mean **Resistance index** .2 Difference between T1 and T2 0 +1.96 SD (0.1) 8 Mean 80° .0 (0.0) 0 -1.96 SD (-0.1) 0 -,2 ,80 ,85 ,9⁰ ,9[']5 1,00 1,05 T1 and T2 mean

Fig. 2 Measurements at rest. T1 first measurement session, T2 second measurement session

conditions. Table 1 summarizes the study subjects' demographics.

Rest condition

Table 2 summarizes the results of blood flow measurements at rest, including mean, mean difference, pairwise comparison, and ICC. At rest, there was no significant difference between means at T1 and T2 for all parameters (p > 0.05). Based on the ICC results, excellent reliability was obtained for peak systolic velocity (0.99 [95%CI 0.97–1.00]) and end-diastolic velocity parameters (0.84 [95% CI 0.56–0.95]). The reliability was fair to good for time-averaged maximum velocity (0.71 [95%CI 0.13–0.90]), pulsatility index (0.64 [95%CI –0.01–0.87]) and resistance index (0.67 [95% CI 0.08–088]).

Bland–Altman plots (Fig. 2) demonstrated minimal bias with the mean difference close to zero for all parameters. The 95% limits of agreement range for peak systolic velocity and time-averaged maximum velocity at rest were narrow considering the mean values, indicating a small variability between T1 and T2. However, for the end-diastolic velocity, pulsatility index and resistance index parameters, the limits of agreement on Bland–Altman plots were wide considering the mean values, thereby indicating larger measurement variability between T1 and T2.

PFM contraction task condition

Table 3 summarizes the results of blood flow measurements after the PFM contraction task, including mean, mean difference, pairwise comparison, and ICC. There was no significant difference between means at T1 and T2 for all parameters (p >0.05). Based on the ICC results, excellent reliability was obtained for peak systolic velocity (0.98 [95% CI 0.95 to 0.99]) and time-averaged maximum velocity parameters (0.76 [95% CI 0.32 to 0.92]). The reliability was fair to good for enddiastolic velocity (0.66 [95%CI -0.02 to 0.88]), pulsatility index (0.71 [95% CI 0.19 to 0.90]), and resistance index (0.55 [95%CI -0.26 to 0.84]).

Bland–Altman plots (Fig. 3) demonstrate minimal bias with the mean difference close to zero for all parameters after PFM contractions. The 95% limits of agreement range for peak systolic velocity was narrow considering the mean values, indicating a small measurement variability between T1 and T2. The 95% limits of agreement range for time-averaged velocity and pulsatility index was larger considering the mean value, indicating a still acceptable measurement variability between T1 and T2. Those limits of agreement were wide for end-diastolic velocity and resistance index measurements considering the mean values, indicating greater measurement variability between T1 and T2.

Discussion

To our knowledge, this is the first intra-observer study assessing the test-retest reliability of IPA blood flow measurements with color Doppler ultrasound. Of interest is that this assessment was achieved under two conditions that were used to evaluate vascular changes, which are at rest and after a PFM contraction task. At rest, the results showed an excellent reliability for peak systolic velocity and enddiastolic velocity parameters and a fair to good reliability for time-averaged maximum velocity, pulsatility index, and resistance index parameters according to the ICC values. Means of the difference between measurements were close to zero for all parameters. On Bland-Altman plots, the variability between measurement sessions (represented by the 95% limits of agreement) was small for peak systolic velocity and time-averaged maximum velocity parameters and wide for the end-diastolic velocity, pulsatility index, and resistance index parameters.

After a PFM contraction task, the results showed an excellent reliability for peak systolic velocity and time-averaged

 Table 3
 Blood flow measurements after a pelvic floor muscle contraction task

	Data				ICC		
	Mean $T1 \pm SD$	Mean $T2 \pm SD$	Mean difference (95% IC)	<i>p</i> value	ICC (95% CI)	p value	
PSV (cm/s)	51.2 ± 14.0	50.0 ± 14.1	1.2 (-0.7 to 3.1)	0.210	0.98 ^a (0.95 to 0.99)	< 0.001	
TAMX (cm/s)	10.9 ± 4.2	10.3 ± 3.8	0.0 (-2.1 to 2.1)	0.558	0.76 ^a (0.32 to 0.92)	0.005	
EDV (cm/s)	1.8 ± 3.3	2.0 ± 3.2	-0.2 (-1.9 to 1.6)	0.841	0.66 ^b (-0.02 to 0.88)	0.027	
PI	4.9 ± 1.6	4.7 ± 1.5	0.2 (-0.6 to 0.9)	0.671	0.71 ^b (0.19 to 0.90)	0.010	
RI	1.0 ± 0.1	0.95 ± 0.1	0.0 (-0.0 to 0.0)	0.556	0.55^{b} (-0.26 to 0.84)	0.064	

PSV peak systolic velocity, TAMX timed-average maximum velocity, EDV end-diastolic velocity, PI pulsatile index, RI resistance index

^a Excellent

^b Fair to good



Fig. 3 Measurements after a pelvic floor muscle (PFM) contraction task. T1 first measurement session, T2 second measurement session

maximum velocity parameters and a fair to good reliability for end-diastolic velocity, pulsatility index, and resistance index parameters according to the ICC values. Means of the difference between measurements were also close to zero for all parameters. On Bland–Altman plots, the variability between measurement sessions was small for peak systolic velocity parameters and larger but acceptable for time-averaged maximum velocity and pulsatility index parameters. However, this variability was wide for the end-diastolic velocity and resistance index parameters.

Moreover, when looking at raw data, the end-diastolic velocity parameter at rest and after a PFM contraction task was often equal to zero, causing the resistance index value to be equal to 1 (at rest: 11 out of 19 of participants at T1 [58%] and 14 out of 19 of participants at T2 [74%]; after PFM contractions: 10 out of 19 of participants at T1 [53%] and 11 out of 19 of participants at T2 [58%]). Therefore, the values related to these parameters (end-diastolic velocity and resistance index) are questionable for the analysis of changes in IPA blood flow.

As already mentioned, to our knowledge, there is no other publication in the current literature on the test–retest reliability of IPA blood flow measurements with color Doppler ultrasound. Two studies used a curved-array probe for a transcutaneous exploration of the IPA emplacement in the gluteal area to guide a pudendal nerve infiltration [2, 3]. The IPA was visible in 98% and 94% of the participants respectively [2, 3]. Thus, the feasibility of the IPA's assessment with the color Doppler ultrasound in those studies was similar to ours, which was 95%. Our study adds information about the reliability of multiple parameters used to assess IPA blood flow under the two conditions at rest and after a PFM contraction task.

The evaluation of the reliability of a measurement technique is important for assessing its stability and repeatability over time [27]. Afterward, researchers and clinicians can use it to assess evolution over time or changes following an intervention. As IPA blood flow has reliable parameters, it would be interesting to have normative data in healthy women. Those parameters could be used to observe vascular changes in different situations such as postpartum, post-menopausal status, aging or in conditions causing lower or challenged perineal vascularity. Peak systolic velocity, time-average maximum velocity and pulsatility index of the IPA blood flow could also be used to evaluate change in vascularity in studies using treatments aiming to improve perineal vascularity, such as hormonal therapy, laser therapy, and PFM training.

Pelvic floor muscle training refers to an exercise program with repeated PFM contractions. It is usually taught and supervised by a health professional, such as a physiotherapist and leads to PFM hypertrophy and improved strength [28]. Studies in skeletal muscles show that training increases blood flow in arteries related to the trained muscles at rest by improved vasodilation function, enlarged diameter, and decreased wall thickness of arteries in combination with improved capillary growth [29]. Skeletal muscle arteries also improve their capacity of adaptation to a muscle contraction task after muscle training, increasing their total blood volume to meet the muscle's needs [30]. As the IPA is the main artery providing blood flow to the PFMs, PFM training could improve IPA blood flow at rest and/or after a contraction task. Assessment of the changes in the IPA blood flow before and after PFM training could be interesting to confirm this hypothesis.

The present study has some limitations. First, this reliability study is closely linked to the population in which the measurements were made, composed of healthy younger women, aged between 22 and 53, mostly pre-menopausal. A similar reliability study of IPA blood flow measurements using color Doppler ultrasound should be undertaken with other populations such as men, older women and men, or in those with challenged vascular conditions. Second, to reach the same reliability results as this study, assessors need to have similar training concerning the measurement techniques. The assessor, a PhD fellow in physiotherapy, had 6 h of training with a radiologist (AT) and 25 h of practice before the beginning of the study. Finally, an inter-observer reliability study should be undertaken for research protocols that involve more than one assessor.

In conclusion, our findings suggest that peak systolic velocity and time-averaged maximum velocity might be reliable parameters of the IPA blood flow at rest, and that peak systolic velocity, time-averaged maximum velocity, and pulsatility index are reliable parameters after a PFM contraction task. The use of color Doppler ultrasound is promising for establishing normative data of the IPA blood flow and for assessing vascular changes in different situations causing lower or challenged perineal vascularity, or before and after treatment improving perineal vascularity.

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Compliance with ethical standards

Conflicts of interest None.

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