# THE USE OF ENDOVAGINAL SONOGRAPHY AND DOPPLER ULTRASOUND IN THE DETECTION OF ENDOMETRIAL CARCINOMA IN WOMEN PRESENTING WITH POSTMENOPAUSAL BLEEDING

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## Abstract

**Purpose**

To evaluate the role of endovaginal sonography (EVS) and Doppler ultrasound in detecting endometrial carcinoma in women presenting with postmenopausal bleeding.

**Materials & Methods**

We prospectively evaluated 421 women with EVS over a 5-year period. Of these 31 (7.4%) were diagnosed with endometrial carcinoma at histopathology. For each patient, biometric and morphologic parameters, as well as Doppler indices of the endometrium were obtained.

**Results**

Applying a combination of biometric and morphologic criteria, EVS diagnosed malignancy with a sensitivity of 77% (95% CI: 59% - 90%) and a specificity of 84% (80% - 87%). Using only biometric criteria (endometrial thickness > 2mm indicating malignancy), EVS achieved a sensitivity of 100% (91% - 100%) and a specificity of 24% (20% - 29%), whereas the corresponding sensitivity and specificity for endometrial thickness > 5mm was 74% (55% - 88%) and 59% (54% - 64%), respectively. The most predictive Doppler index was peak venous velocity (95% CI for odds ratio: 1.08 – 1.40).

**Conclusion**

Using a combination of biometric and morphologic sonographic criteria achieves the best accuracy in diagnosing patients with endometrial carcinoma, however at the cost of a decreased sensitivity.

## Résumé

**But**

Évaluer le rôle de l'échographie endovaginale (EEV) et du doppler afin de détecter la présence d'adénocarcinome de l'endomètre dans un contexte clinique de saignement postménauposé.

**Matériel et méthode**

Nous avons suivi de façon prospective 421 femmes par EEV et ce sur une période de 5 ans. De ces patientes, 31(74%) avaient un diagnostic de carcinome endométrial à l'histopathologie. Des mesures biométriques, morphologiques ainsi qu'une étude doppler ont été obtenues pour chacune de ces patientes.

**Résultats**

En appliquant les critères biométriques et morphologiques, l'EEV a démontré une sensibilité de 77% (95% IC: 59% - 90%) et une spécificité de 84% (80% - 87%) pour diagnostiquer la néoplasie. En utilisant seulement les critères biométriques (épaisseur de l'endomètre > 2 mm indiquant une néoplasie), l'EEV atteint une sensibilité de 100% (91% - 100%) une spécificité de 24% (20 - 29%), alors que la sensibilité et spécificité d'un critère d'épaisseur de l'endomètre >5 mm deviennentt respectivement 74%(55% - 88%) et 59%(54% - 64%). La vélocité veineuse maximale s'est avérée être le critère doppler le plus prédictif (95% IC odds ratio 1.08-1.40).

**Conclusion**

L'utisation d'une combinaison de critères biométriques et morphologiques en échographie permet d'obtenir des résultats optimaux pour diagnostiquer les patientes ayant un carcinome de l'endomètre, cependant en acceptant une perte au niveau de la sensibilité.

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## 1 - INTRODUCTION

Abnormal vaginal bleeding is a common problem accounting for up to 20% of office visits to a general gynecologist (1). Although no significant endometrial pathology will be found in the majority of patients, approximately 10% of patients presenting with *postmenopausal* bleeding will be diagnosed with endometrial carcinoma (2). Carcinoma of the endometrium is the most common invasive malignancy of the female genital tract, reaching a peak incidence between 55 and 65 years of age. Intermenstrual and postmenopausal bleeding is the initial symptom in 75-90% of patients (3-5). Early diagnosis and treatment is important since the depth of myometrial invasion is considered the factor most responsible for the extreme variation in the 5-year survival of patients with Stage I disease: from 40 to 60 % in the most invasive cases to 90 - 100% in cases with little or no myometrial involvement (6-9). Traditionally, postmenopausal women with abnormal vaginal bleeding were referred for a diagnostic curettage (D & C) under general anaesthesia if a definite cause was not evident using clinical evaluation. However, this procedure is invasive and includes complications such as infection, bleeding and uterine perforation (10). D & Cs can be inaccurate in patients with distortion of the endometrial canal due to leiomyomata, uterine anomalies and cervical stenosis (10). In 60% of patients, less than half of the endometrial cavity surface is actually sampled (11). More recently, office-based endometrial sampling procedures have gained widespread acceptance. The convenience to the patient and physician, as well as the cost containment of these procedures has been firmly established in the literature (12). However, outpatient endometrial sampling techniques are somewhat less accurate than the D & C performed under general anaesthesia (13-16). Furthermore, the role of endometrial sampling in screening for endometrial carcinoma, has not been firmly established. A less invasive technique that provides an accurate diagnosis of endometrial carcinoma in women presenting with postmenopausal bleeding would certainly be of clinical value. Furthermore, if this method proved to be efficacious in detecting the disease at an early stage, a screening program could be implemented for asymptomatic postmenopausal women. Sonography has recently been advocated as a noninvasive test for evaluating postmenopausal women presenting with abnormal vaginal bleeding (17,18). Whether the role of

sonography lies in identifying patients that would benefit most from undergoing endometrial sampling, or is reserved for patients who have undergone unsuccessful sampling procedures, remains controversial (19-23).

The use of transabdominal sonography in the detection of endometrial carcinoma has been well documented (24-27). Chambers (26) and Nsari (27) reported sensitivities of 67% and 91% respectively using transabdominal sonography to detect endometrial carcinoma. Particular emphasis has been placed on the alteration of the normal endometrial stripe as an indicator of endometrial pathology (24,25,27-29). However, obesity, retroflexion and multiple leiomyomas of the uterus can make assessment of the endometrial stripe using transabdominal sonography technically difficult. With the advent of endovaginal sonography (EVS) these technical limitations have largely been overcome. Furthermore, the greater resolution afforded with the higher frequency endovaginal probe can improve the detection of endometrial carcinoma (30-32). A prospective comparison of endovaginal and transabdominal sonography by Coleman et al. (33) reported that endovaginal scans yielded new information in 60% of cases and allowed better visualization of pelvic structures in 22% of cases. The clinical diagnosis was altered on the basis of endovaginal sonographic findings in 24% of patients and confirmed with certainty in 72% of patients. Therefore, we feel that any study reporting on the accuracy of early endometrial carcinoma detection using sonography, should include an endovaginal examination (31,34). Although EVS has been demonstrated to have a high sensitivity for detecting *early* endometrial carcinoma when certain morphological criteria are applied, no features unique to malignant disease have been identified (32,35,36). In particular, most studies evaluating the role of EVS in patients presenting with postmenopausal bleeding have emphasized the thickness of the endometrial lining as a study endpoint (5,37-41). However, morphological criteria at real-time ultrasound, such as heterogeneous endometrial echotexture, increased endometrial echogenicity, and poor definition of the endo-myometrial junction, can improve the specificity of diagnosing endometrial carcinoma (41-43). Furthermore, the advent of Doppler ultrasound offers the potential advantage of characterizing tissue using functional, in addition to structural criteria. Since approximately 80-90% of all curettage procedures performed for

postmenopausal bleeding result in benign diagnoses (2,10,24,39,44), unnecessary endometrial sampling may be avoided if the real-time findings of EVS combined with Doppler ultrasound can be used to identify endometria with a high likelihood of disease.

A low impedance signal pattern on Doppler ultrasound, is postulated to originate when high velocity flow enters a vessel with little or no vascular resistance. This was demonstrated by Taylor et al. who obtained histologic correlation in eight neoplasms with low impedance flow (45). Microscopic examination in all these cases demonstrated the presence of large, thin-walled, sinusoidal spaces that lacked muscular support and therefore corresponded well to the theoretical vascular spaces. Changes in impedance to blood flow in an organ may therefore be an early obligatory event in the evolution of a malignant lesion. It is in this regard that the addition of Doppler ultrasound offers the greatest potential for detection and characterization of endometrial carcinoma. Recent advances in Doppler techniques have allowed detailed documentation of normal flow velocity waveforms in the uterine and ovarian arteries (46-51). Whereas extensive work on tumour vascularity of breast carcinomas, liver neoplasms and gestational trophoblastic disease has been undertaken (45,52-55), studies reporting on the role of Doppler ultrasound in the diagnosis of endometrial carcinoma have reported conflicting results (56-61).

In order to evaluate the role of EVS and Doppler ultrasound in detecting endometrial carcinoma, we *prospectively* examined 557 women presenting with postmenopausal bleeding over a five-year period. Of these, 421 met our final inclusion criteria. Patients were included irrespective of whether they were receiving hormonal replacement therapy. Sonographic assessment of the uterus included measurements of endometrial thickness, a detailed description of endometrial morphology, as well as colour and pulsed Doppler analysis. In addition, for each patient, the sonologist was asked to classify the appearance of the endometrium as benign, malignant or indeterminate. Findings at sonography were correlated with histological examination of the endometrium where available. Specifically our *objectives* were as follows:

1) To estimate the sensitivity and specificity of EVS, using real-time parameters of endometrial thickness and morphology, to diagnose endometrial carcinoma.
2) To estimate the predictive value of Doppler ultrasound in diagnosing endometrial carcinoma over that achieved by using real-time sonographic features alone.
3) To determine optimal threshold values for endometrial thickness, and Doppler ultrasound parameters where appropriate, to distinguish benign from malignant endometrial pathology.
4) To estimate the sensitivity and specificity of using endometrial thickness, versus a combination of real-time sonographic features, in diagnosing endometrial carcinoma.

The ensuing chapters of this thesis are subdivided into the following sections. Chapter 2 contains a literature review of 1) the accuracy of various endometrial sampling techniques in detecting endometrial carcinoma and other endometrial pathology, and 2) the role of EVS and Doppler ultrasound in differentiating benign from malignant endometria. Chapter 3 describes the study methodology. Chapter 4 presents the results of the study and includes details of the statistical analysis. Chapter 5 presents a discussion of the study results, including discrepancies with the literature, potential clinical applications and study limitations.

## 2 - LITERATURE REVIEW

### 2.1 Endometrial histology

Although dilatation and curettage (D & C) is generally considered the "standard of reference" for obtaining the necessary diagnostic intrauterine pathology, support for this assertion in the literature is lacking. The sensitivity and specificity of D & C are difficult to assess because few large series confirm the histology with a subsequent hysterectomy specimen. In a series of 512 patients in whom the uteri were removed immediately after the D & C, endometrial lesions were missed in up to 10% of cases including: 38 endometrial polyps, 4 submucosal fibroids, 2 endocervical polyps, 2 placental polyps and 1 undisturbed pregnancy (10). For diagnosing endometrial hyperplasia or

carcinoma, false negative rates ranging from 2% to 6% have been reported (10,11,62,63). In a study of 50 consecutive patients who underwent D & C immediately prior to hysterectomy, Stock and Kanbour found that in 60% of patients less than 1/2 of the endometrial surface was sampled and in 16% less than 1/4 of the surface was actually sampled (11). In addition, as emphasized by Word et al. (10) in a review of over 6,000 D & Cs, this procedure is invasive and may be associated with complications such as infection, bleeding and uterine perforation. In this series, the overall risk of uterine perforation was determined to be 1 in 99, and increased to 1 in 38 for postmenopausal women.

In 1982, a critical review of the role of diagnostic D & Cs was published after an analysis of 33 reports consisting of 13,598 D & Cs and 5,851 endometrial sampling procedures (62). The number of procedures yielding specimens adequate for histologic interpretation varied from 77 - 94% for D & Cs, and 85 - 99% for endometrial sampling techniques. In assessing safety, the D & C was associated with a higher complication rate than the endometrial sampling technique. Furthermore, the D & C had a low mortality rate, which was not reported with endometrial sampling. Office-based endometrial sampling procedures, such as Pipelle and Vabra sampling have gained widespread acceptance, since the convenience to the patient, lower complication rate, and cost containment of these procedures have been firmly established in the literature (12). However, outpatient endometrial sampling techniques are somewhat less accurate than the D & C performed under general anaesthesia, particularly in the setting of endometrial polyps, submucous myomas and thin endometria (13-16,62-65).

*Table I* below summarizes the sensitivity of various endometrial sampling procedures, in the detection of endometrial carcinoma and/or endometrial hyperplasia, published during the last few decades. When attempting to assess the diagnostic accuracy of endometrial sampling a number of difficulties arise, since few large series confirm the histology with a subsequent hysterectomy specimen. First, the true incidence of lesions in the uterus cannot be determined when D & C is used as the standard of reference, since the diagnostic accuracy of D & C is not precisely known. Second, the more complete the first procedure is for obtaining samples of the endometrium, the less likely a second procedure

will provide tissue leading to the same diagnosis (66). Thus, the first procedure artifactually decreases the diagnostic accuracy of the second. The studies listed in *Table 1* vary considerably in their methodology making meaningful comparisons difficult. For example, some studies evaluated a general gynecologic patient population presenting with a variety of symptoms suggestive of endometrial pathology, while others restricted their sample to a subpopulation at risk for endometrial carcinoma, or to patients already proven to have endometrial carcinoma. The standard(s) of reference used are also variable. Nevertheless, a number of useful generalizations can be drawn from these studies. In 12 of the 15 studies listed, the sensitivity of endometrial sampling for detecting carcinoma is 85% or greater, with the two largest series reporting sensitivities of 94% and 96% respectively. The sensitivity for diagnosing endometrial hyperplasia is more modest ranging from 58 - 86%. None of the series on endometrial sampling reported a false positive diagnosis for endometrial carcinoma.

***Table I. Sensitivity of Endometrial Biopsy in Diagnosing Uterine Cancer / Hyperplasia***

| *Author* | *Year* | *No. Patients* | *Positive Bx. (%) Carcinoma* | *Positive Bx. (%) Hyperplasia* | *Gold STD* |
|---|---|---|---|---|---|
| Palmer [0] | 1950 | 301 | 86/95 (91%) | ----- | D & C, biopsy |
| Jordan[+] | 1956 | 128 | 23/25 (92%) | ----- | Hyster. |
| McGuire[++] | 1962 | 136 | 61/72 (85%) | ----- | D&C, Hyster. |
| Slaughter[++] | 1962 | 406 | 52/68 (76%) | ----- | D&C, Hyster. 1-yr. F/U for neg. biopsies |
| Hofmeister[+] | 1974 | 20,677 | 176/187 (94%) | ----- | D&C, Hyster. |
| Vuopala[+] | 1977 | 722 | 56/65 (86%) | 27/60 (45%) | D&C, Hyster. |
| Lutz[++] | 1977 | 103 | 25/27 (93%) | ----- | D&C, Hyster. |
| Greenwood[+] | 1979 | 228 | 17/21 (81%) | 17/25 (68%) | Hyster. |
| Ferenczy[+] | 1979 | 73 | 7/7 (100%) | 19/22 (86%) | D&C, Hyster. |
| Grimes[+] | 1982 | 1,123 | 42/44 (96%) | ----- | D&C, Hyster. |
| Stovall [+] | 1989 | 240 | 5/7 (71%) | 15/26 (58%) | Hyster. |
| Stovall [+++] | 1991 | 40 | 39/40 (97.5) | ----- | Hyster. |
| Goldchmit[+] | 1993 | 176 | 3/3 (100%) | 11/18 (61%) | D&C |
| Zorlu[+++] | 1994 | 26 | 24/26 (95%) | ----- | Hyster. |
| Van Bosch[++] | 1995 | 126 | 6/6 (100%) | N/A (64%) | Hyster. Hysteroscopic biopsy |

[+] General gynecologic patient population
[++] Subpopulation at risk for endometrial carcinoma
[+++] Patients with proven endometrial carcinoma
[0] Study population not described
N/A = Not available

Advantages of office-based endometrial sampling procedures in comparison to D & Cs include convenience to the patient, a low complication rate, and a relatively low cost. However, outpatient endometrial sampling techniques may result in specimens *inadequate* for histologic interpretation in up

to 15% of cases (62). In addition, office biopsies may be technically impossible in 10% of patients due to cervical stenosis (67). Although the accuracy of endometrial sampling procedures appears to be comparable to D & C for diagnosing endometrial carcinoma, a lower accuracy is achieved in the setting of endometrial hyperplasia, polyps or submucosal leiomyomas (13-16,62-65). This is clinically relevant, because endometrial polyps or submucosal myomas have been reported in up to 90% of patients with recurrent postmenopausal bleeding (68). The detection of pedunculated benign conditions in the uterine cavity is a limitation of all blind sampling procedures, including D & C. Hysteroscopy, which allows direct visualization of the endometrial cavity, is superior to D & C in making an accurate diagnosis of endometrial polyps and submucosal myomas (69,70). Hysteroscopy is, however, an invasive method that carries a small but real risk of perioperative complications. Furthermore, the technique cannot be applied in all women with postmenopausal bleeding. Alternative methods of screening and/or assessing endometrial pathology in postmenopausal women would certainly be of clinical value. Several authors have recommended combining endometrial sampling and endovaginal sonography (EVS) in the diagnosis of endometrial disease in postmenopausal women (14,20,21,71).

## 2.2 Real-time endovaginal sonography

### *2.2.1 Measurements of endometrial thickness*

The advent of high resolution transvaginal probes has revolutionized the ability to visualize the endometrium sonographically and to detect endometrial pathology (30-33). The use of endovaginal sonographic measurements of endometrial thickness, as a predictor of disease in postmenopausal women with bleeding, has recently been well established in the literature (39). Over the past 10 years, numerous articles have been published with differing recommendations regarding the optimal cut-off values for endometrial thickness, above which disease is more likely. Many of these studies, however, have investigated small patient populations with a low prevalence of disease (18,38,72-79). In these series, fewer than 10 patients harbouring endometrial malignancy were studied, making it difficult to draw meaningful conclusions regarding optimal cut-off values.

More recently, larger trials have been conducted to define an endometrial thickness below which no pathology is found, in the hopes of using this measurement as a screening tool in postmenopausal women with abnormal uterine bleeding (5,17,19,22,40,80,81). Cut-off values ranging from 4 - 10 mm (double layer endometrial thickness) have been proposed. In all studies, the mean endometrial thickness for patients with hyperplasia or carcinoma was greater than for those without pathology.

In the study by Granberg et al. (5) of 1991, it was found that in 205 postmenopausal women with bleeding, an endometrial thickness of < 9mm indicated absence of endometrial carcinoma at D & C. All 18 patients with proven endometrial carcinoma at histology had an endometrial thickness of 18.2mm ± 6.2mm (mean ± SD), while patients with a diagnosis of endometrial atrophy (n = 157) or hyperplasia (n = 13) had measurements of 3.4mm ± 1.2mm and 9.7mm ± 2.5mm, respectively. Endometrial thickness measurements used in this study included the contents of a distended uterine cavity, in addition to the anterior and posterior endometrial layers. This technique overestimates the endometrial thickness in benign conditions with fluid retention due to cervical stenosis. Furthermore, no distinction is possible between conditions that result in localized or diffuse endometrial thickening, such as endometrial hyperplasia and carcinoma, versus endometrial polyps. Nevertheless, the purpose of this study was primarily to differentiate a benign from a pathological endometrial process without attempting to diagnose specific histological subtypes. In this study, an endometrial thickness of ≥ 5mm resulted in a positive predictive value of 87% for detecting the presence of endometrial pathology. Using this cut-off value, 70% of the curettage procedures could have been avoided without misclassifying a single case of endometrial carcinoma. Similarly Guner et al. (19) studied 289 postmenopausal women of whom 192 presented with abnormal vaginal bleeding, and concluded that curettage is not necessary to exclude carcinoma if the endometrial thickness measures less than 5mm as assessed by EVS. Using this cut-off value, EVS was able to diagnose the presence of endometrial carcinoma with a sensitivity of 100%, a specificity of 49%, a positive predictive value of 13%, and a negative predictive value of 100%. Corresponding values for detecting all endometrial pathology were as follows: sensitivity 100%, specificity

77%, positive predictive value 75%, and negative predictive value of 100%. In the group of patients that presented with postmenopausal bleeding, there was a significant linear correlation between the probability of finding endometrial pathology and increasing endometrial thickness (linear regression $R = 0.91$, $p < 0.03$).

Haller et al. (80) in 1996, compared the accuracy of detecting endometrial pathology using endometrial thickness measurements with EVS versus hysteroscopic findings in women with postmenopausal bleeding. A total of 81 women not receiving hormonal replacement therapy were entered into the study protocol. The final diagnosis from histological specimen at D & C included: endometrial atrophy (n = 12), irregular proliferative changes (n = 21), polyps (n = 16), hyperplasia (n = 16), and endometrial carcinoma (n = 16). Using an endometrial thickness of ≥ 5mm to indicate a positive test, EVS diagnosed the presence of endometrial carcinoma with a sensitivity of 100%, specificity of 26%, PPV of 25%, and NPV of 100%. All patients with a diagnosis of endometrial carcinoma at D & C had an endometrial thickness of ≥ 8mm. Using the same criteria to detect all endometrial pathology, EVS achieved a sensitivity of 96%, specificity of 46%, PPV of 72%, and NPV of 88%. In the same patient population, hysteroscopy detected endometrial carcinoma with a sensitivity of 50% and a specificity of 100%; and detected all endometrial pathology with a sensitivity of 96% and a specificity of 94%. Karlsson et al. (17) measured the endometrial thickness using sonography in 105 women presenting with postmenopausal bleeding, one day prior to the scheduled D & C. These authors included in their endometrial measurements, fluid or polyps that distend the uterine cavity. An endometrial thickness of ≤ 5mm as measured by endovaginal scanning was considered benign, and > 5mm as pathological. At histopathology, 58 (55%) endometria were considered benign and 47 (45%) pathological, including 16 carcinomas. EVS detected the presence of endometrial pathology with a sensitivity of 97%, specificity of 81%, PPV of 72%, and NPV of 98%. Corresponding values for endometrial carcinoma using the same sonographic criteria were sensitivity 94%, specificity 64%, PPV 32%, and NPV 98%, respectively. These authors concluded that EVS is a valuable diagnostic

instrument for detecting pathological conditions of the uterine mucosa with a sensitivity comparable to endometrial sampling or D & C.

From January 1988 to December 1992, Kurjak et al. (82) screened 5,013 asymptomatic women with EVS, who were 40 years of age or older. Patients on hormonal replacement therapy were not entered into the study protocol. A positive test for malignancy on real-time EVS, included an endometrial thickness of 10mm or more. Thirty-four (0.68%) cases of endometrial abnormalities were detected with EVS and were operated on. Of these, six (0.12%) were endometrial carcinoma, 18 (0.36%) were endometrial hyperplasia, and 10 (0.12%) were benign endometrial polyps. EVS correctly classified all 6 malignant and all 28 benign conditions.

Malinova et al. (81) evaluated the usefulness of sonographic measurements of endometrial thickness combined with a progesterone challenge test, to detect endometrial pathology in postmenopausal women. Only women who had been postmenopausal for at least 2 years prior to sonography, and were not receiving hormone replacement therapy, were included in the study protocol. Of the 284 patients that met the inclusion criteria 130 women were asymptomatic, while 154 presented with postmenopausal bleeding. Both groups were further subdivided into patients with normal findings at EVS (endometrial thickness $\leq$ 5mm), and those with abnormal findings (thickness $\geq$ 6mm). In the group of asymptomatic women, there was a negative correlation between endometrial thickness and number of years since menopause ($r = -0.39$). In the 42 women who presented with postmenopausal bleeding and normal sonographic findings, the final diagnosis at D & C included endometrial atrophy in 41 patients and endometrial polyp in one patient. In the 112 symptomatic women with abnormal findings at EVS, the final diagnosis was as follows: atrophy ($n = 5$), polyp ($n = 27$), hyperplasia ($n = 11$), and carcinoma ($n = 69$). Therefore, in patients presenting with postmenopausal bleeding, EVS diagnosed the presence of endometrial carcinoma with a sensitivity of 100%, a specificity of 49%, a PPV of 62%, and a NPV of 100%. The lowest measurement for endometrial thickness in patients with proven endometrial carcinoma was 7mm. This high prevalence (69/154 or 45%) of endometrial carcinoma among women presenting with postmenopausal bleeding likely indicates a selection bias, and increases the PPV

of the test. When using the same cut-off value as a screening test for endometrial pathology, the calculated sensitivity and specificity were 99% and 89% respectively.

Pertl et. al in 1996 (22), studied 169 postmenopausal women with EVS prior to a diagnostic curettage or hysterectomy. Indications for curettage included postmenopausal bleeding (n = 150), or an endometrial thickness ≥ 6mm (n = 19). Thirty-five patients were on hormonal replacement therapy. Of the 169 patients entered into the study protocol, 91 had normal findings at histopathology, 40 patients had endometrial hyperplasia, 17 had polyps, and in 21 cases endometrial carcinoma was found. Applying a threshold of ≥ 6mm to indicate a positive test, EVS diagnosed endometrial pathology with a sensitivity of 90%, a specificity of 31%, a PPV of 53% and a NPV of 78%. Corresponding values for diagnosing endometrial carcinoma using the same sonographic criteria were sensitivity 96%, specificity 24%, PPV 17% and NPV 97%. There was one false negative diagnosis of endometrial carcinoma in a patient with an endometrial thickness of 5mm. By using the same threshold value as a criterion for entry into the study, and as a determinant for outcome, the authors are introducing an important selection bias. However, only 19/169 or 11% of patients enrolled into the study were included on the basis of endometrial thickness. Nevertheless, clinical follow-up on the group of patients not presenting with post-menopausal bleeding and whose endometrium measured < 6mm with EVS would have been important to obtain.

The largest study published to date on the evaluation of endometrial thickness in women presenting with postmenopausal bleeding is a multicenter study known as the "Nordic Trial" where 1,168 women were prospectively evaluated with EVS (40). Of the 1,168 women entered into the study protocol, 351 (30%) were receiving hormonal replacement therapy. In this study, an endometrial abnormality was defined as the presence of endometrial polyps (n = 140), hyperplasia (n = 112), or carcinoma (n = 114). The sensitivity and specificity for endometrial disease was determined using receiver operating characteristic (ROC) curve analysis, for cut-off levels of endometrial thickness ranging from 1 - 72 mm. The sensitivity and specificity for identifying endometrial pathology varied greatly depending on the cut-off value used. In the Nordic trial, EVS

achieved a sensitivity and specificity of 100% and 51% (95% CI: 48%-54%) respectively, for diagnosing endometrial carcinoma when a cut-off value of ≥ 5mm was applied. If a cut-off value of ≥ 6 mm had been used, the specificity would have improved to 59% (95% CI: 56%-62%) at the expense of missing two carcinomas. When evaluating for all endometrial pathology, a cut-off value of ≥ 5 mm resulted in a sensitivity of 96%, a specificity of 68%, a PPV of 61%, and a NPV of 97%. The corresponding figures at a cut-off limit of ≥ 6mm were 94%, 78%, 69%, and 96%, respectively. These authors concluded that the risk of finding pathological alterations of the endometrium at D & C when the endometrium measured ≤ 4 mm on EVS was 3.6%, with a 95% confidence limit of 5.5%. Therefore, in women with postmenopausal bleeding and an endometrial thickness of ≤ 4mm, routine histological sampling does not appear justified.

The role of EVS in detecting endometrial cancer and other endometrial abnormalities in postmenopausal women with vaginal bleeding is well summarized in the following meta-analysis of English-language and non-English-language articles published between 1966 and 1996 (83). The meta-analysis comprised 35 studies and includes 5,892 women. Using a threshold value of > 5 mm to define abnormal endometrial thickening, 96% (95% CI: 94%-98%) of women with cancer had an abnormal EVS, whereas 92% (95% CI: 90%-93%) of women with endometrial pathology had an abnormal test result. Corresponding specificities were 61% (95% CI: 59%-63%) and 81% (95% CI: 79%-83%), respectively. The false-negative rate of 8% for EVS compares favourably with those achieved using office-based endometrial biopsy devices. EVS was equally accurate at identifying women with endometrial disease, regardless of whether or not they were receiving hormonal replacement therapy. For a postmenopausal woman with vaginal bleeding and a 10% pretest probability of endometrial cancer, the posttest probability decreases to 1%, given a negative EVS. These authors concluded that EVS is highly sensitive for detecting endometrial carcinoma, and can identify patients at low risk for endometrial disease obviating the need for endometrial sampling in this subgroup of patients. However, women on hormonal replacement therapy had a significantly higher false positive rate (specificity 77%, 95% CI: 75%-79%), compared to patients not taking hormones (specificity 92%, 95% CI: 90%-94%). These results are not

surprising, since endometrial thickness is known to increase after the initiation of hormone replacement therapy. The degree of increase in endometrial thickness, however, will vary depending on the type of hormonal regimen used; and is most marked with the ingestion of sequential estrogen/progesterone, followed by unopposed estrogen and is least affected by continuous combined estrogen/progesterone regimens (72). For this reason, many authors have excluded patients on hormonal replacement therapy, in studies evaluating endometrial thickness in women with postmenopausal bleeding. Others have advocated a higher threshold value for endometrial thickness in postmenopausal women on hormonal replacement therapy compared to controls (8 mm versus 5 mm) (79,84).

*2.2.2 Morphological assessment of the endometrium*

Although the cut-off values described in the literature vary considerably, endometrial thickness is often used as the sole criterion in the sonographic assessment of the endometrium in postmenopausal women. As evidenced from the preceding studies, proposed cut-off values for detecting endometrial carcinoma result in a high sensitivity but a relatively low specificity. Most authors recommend using a low cut-off value such as 4 or 5 mm, which maintains the sensitivity but sacrifices specificity. This results in many unnecessary curettage procedures being performed in order not to miss a carcinoma. By increasing the threshold value, the specificity will improve, however at the cost of increasing the number of false negative examinations. To address this issue, a number of investigators have recently studied morphological features of the endometrium, in addition to measuring endometrial thickness with EVS.

The use of morphological criteria for evaluating the endometrium is often rejected on the premise that the method is too subjective, and depends to a greater extent on the examiner's experience. However, morphological analysis and pattern recognition forms the basis of most radiological image interpretation. Designing studies where multiple reviewers with varying degrees of expertise perform the sonographic examinations will improve the generalizability of these results. In addition, although measurements of endometrial thickness are

generally considered an objective criterion, this is unclear in practice since the recommended cut-off values in the literature vary from 4mm to 10mm. This is not surprising when one considers that the unit of measurement is in the millimeter range, where even small deviations in the measured thickness will significantly affect the test performance. Furthermore, Duda et al. (85) have shown that differences in measurements of endometrial thickness on EVS between observers approach statistical significance, and that the variability between observers depends largely on the years of experience. Despite these limitations, endometrial biometry has been established as a useful parameter in the sonographic evaluation of the postmenopausal endometrium. The use of morphological, in addition to biometric criteria, adds little time to the sonographic examination, and offers the potential to improve the predictive value of this test.

Weigel et al. (41) in 1995 emphasized this point by publishing an article entitled: "Measuring the thickness - is that all we have to do for sonographic assessment of endometrium in postmenopausal women?" This group of investigators prospectively examined 200 patients in order to ascertain the value of using morphological features on gray-scale ultrasound imaging in patients with an endometrial thickness in the indeterminate range for pathology (3mm to 10 mm). Several morphologic criteria of the endometrium relative to the myometrium were identified: 1) homogenous low echo, 2) homogeneous high echo, 3) heterogeneous low echo, and 4) heterogeneous high echo. In addition, the presence or absence of a central echo between the two endometrial surfaces was documented. Homogeneity, low-level echogenicity, and a sonographically depictable central echo between symmetrical endometrial layers indicated the absence of disease; whereas, heterogeneity and increased echogenicity were hallmarks of pathologic changes. This group concluded that combining metric and morphologic parameters improved not only the predictability of pathologic findings, but also the overall accuracy of the sonographic evaluation of the endometrium in postmenopausal women.

Brandner et al. (86) evaluated 221 postmenopausal women with EVS, including 139 (63%) who presented with abnormal vaginal bleeding. This group of investigators used various morphological criteria, as well as endometrial

thickness, to classify patients as having endometrial atrophy, proliferative endometria, endometrial hyperplasia/polyps or endometrial carcinoma. At histopathology, 32 patients with endometrial carcinoma were diagnosed. EVS correctly diagnosed the presence of disease in 30 patients; there were 8 false positive and 2 false negative diagnoses. Therefore, EVS detected the presence of endometrial carcinoma with a sensitivity of 93%, a specificity of 96%, a positive predictive value of 79%, and an overall accuracy of 96%. These authors concluded that EVS using morphological criteria represents a valid, non-invasive method for diagnosing endometrial pathology.

Hulka et al. (42) retrospectively reviewed the sonograms of 73 women aged 45 years or older, who had abnormal endometria from January 1992 through to January 1993 as per a search of the departmental computer data base. Histological correlation was available in 68 patients, who constituted the final study group. This group of investigators found that the mean endometrial thickness in the eight patients with carcinoma (mean 29.7 mm, range 18.5 - 63.0 mm), was greater than in patients with other histologic diagnoses, aside from one patient with a secretory endometrium (39 mm). However, patients with endometrial carcinoma in this study demonstrated a wide range of thickness that overlapped with those of benign conditions. Therefore, these authors concluded that analyzing the sonographic appearance of the endometrium remains necessary for differential diagnosis. They found that most endometrial carcinomas had a heterogeneous appearance, while benign endometria tended to be uniformly echogenic, with cystic spaces commonly encountered in polyps.

Emanuel et al. (87) examined 260 patients referred for evaluation of abnormal uterine bleeding, of whom 47 were postmenopausal. Using a combination of morphologic and biometric criteria, all 7 cases of endometrial carcinoma were correctly diagnosed with EVS. For detecting all intrauterine abnormalities in patients with abnormal vaginal bleeding EVS demonstrated a sensitivity of 96% and a specificity of 89%. With a pretest probability (prevalence) of 42%, this resulted in posttest probabilities of 3% in the case of a normal sonogram and 87% for an abnormal sonogram. The corresponding likelihood ratios were 0.04 and 9.09, respectively. These authors concluded that EVS appears to be an effective procedure to exclude endometrial and intrauterine

abnormalities. EVS could be implemented as a routine first-step in patients with abnormal uterine bleeding, and identify those requiring further evaluation in case of an abnormal or inconclusive sonogram.

Other investigators, however, remain skeptical as to the role of EVS in the evaluation of patients presenting with abnormal vaginal bleeding. Hänggi et al. (23) studied 203 consecutive women with EVS prior to a scheduled diagnostic D & C or hysterectomy, of whom 91 presented with symptoms of postmenopausal bleeding. The prevalence of endometrial carcinoma was 26% (n = 24) in this subgroup of patients. Criteria for malignancy on EVS included an endometrial thickness of greater than 5 mm, areas of decreased echogenicity or heterogeneity, and poor definition of the endo-myometrial junction. Applying these sonographic criteria, endometrial carcinoma was diagnosed with a sensitivity of 85%, specificity of 78%, PPV of 52%, and NPV of 95%. Of the patients with proven endometrial carcinoma at curettage, 3/24 (13%) had an endometrial thickness of less than 5mm on EVS. These authors concluded that dilatation and curettage is necessary in the evaluation of women presenting with postmenopausal bleeding.

*2.2.3 Colour and pulsed Doppler sonography*

In addition to using endometrial thickness and morphology, more recently a number of investigators have used colour and pulsed Doppler indices, in an attempt to better characterize endometrial pathology (88). The Doppler examination is based on the Doppler effect first described by Christian Johann Doppler in 1842 (89). Although Doppler enunciated his principle in 1942, he confused its interpretation and used it incorrectly to explain the colour of binary stars (90). The acoustical Doppler effect was first demonstrated in 1845 by Buys Ballot using a trumpeter riding on a steam locomotive (90). As the train moved towards a group of observers on the platform the sound of the trumpet increased in pitch, while the reverse occurred when the train moved away from the platform. This difference between the received and transmitted frequencies when sampling a moving target forms the basis of the Doppler effect, referred to as the Doppler frequency shift.

Colour and pulse gated (duplex) Doppler sonography involves the detection of phase, amplitude, and frequency shift of blood flow within vessels. With duplex sonography, a graphic representation of Doppler information (spectral display) from a single point on a real-time section is provided. With colour Doppler imaging, flow information from an entire sonographic section is encoded in colour and superimposed on the gray scale image. An actual real-time image of the flow is obtained, and direction, mean velocity, as well as areas of turbulence can be identified and quantified using colour encoded velocity maps. Colour Doppler is capable of routinely demonstrating very small intraparenchymal vessels in the uterus and ovaries, in addition to major pelvic arteries and veins. These small intraparenchymal vessels are difficult to sample consistently with pulsed Doppler, because they are not directly visualized on gray scale imaging alone. Although colour Doppler greatly facilitates the depiction of vascular structures in the pelvis, pulsed Doppler remains essential, as it defines changes in vascular impedance and provides information about flow characteristics within different organs. A low impedance signal pattern on Doppler ultrasound occurs, when high velocity flow enters a vessel with little or no vascular resistance (45). Sinusoidal spaces that lack muscular support and therefore form a low-resistance bed, can be seen on microscopic examination of malignant tumour growth (45). Changes in impedance to blood flow in an organ may therefore be an early event in the development of a malignant lesion. It is in this regard that the addition of Doppler ultrasound may facilitate the detection and characterization of endometrial carcinoma.

Several methods have been proposed to quantify the characteristics of pelvic blood flow (88). Velocity measurements alone (peak systolic velocity, end diastolic velocity) have been used primarily for quantifying vascular stenoses, and have not been applied extensively for tissue characterization of endometrial pathology. Ratios between systolic and diastolic flow are ideally suited for tissue characterization, since they provide an estimate of vascular impedance, are angle-independent, and unitless. Commonly used ratios are the pulsatility index (PI), resistive index (RI), and the S/D ratio. The PI is defined as the peak systolic velocity (PSV) minus the end diastolic velocity (EDV) divided by the mean velocity (91). The RI is calculated as the PSV minus the EDV divided by the PSV

(92). Low values for PI and RI indicate increased diastolic flow and decreased vascular impedance. Other investigators have relied on the S/D ratio, a simple measurement of PSV divided by EDV (93). There are theoretical advantages to using PI to measure vascular impedance, since it takes into account the entire frequency spectrum of the cardiac cycle; nevertheless, the superiority of one ratio over another has not been clearly established.

Several investigators have measured Doppler indices in an attempt to differentiate benign from malignant endometria, in patients presenting with post-menopausal bleeding. However, opinions differ as to the role of Doppler ultrasound in this clinical setting. Bourne et al. (56) studied uterine artery blood flow in 54 postmenopausal women, 17 of whom were found to have endometrial carcinoma at dilatation and curettage. The mean PI of patients with endometrial carcinoma was 0.89 while the PI of patients without endometrial carcinoma was 4.25. Retrospectively applying a cutoff PI value of 2.00, would result in a true positive rate of 99.0% and a false positive rate of 2.6%. In contrast, measurements of endometrial thickness in the same group of patients (using a threshold value of 5 mm) yielded a true positive rate of 99%, and a false positive rate of 41%. These authors concluded that transvaginal pulsed Doppler ultrasound, particularly with colour flow imaging had great potential in detecting endometrial carcinoma in postmenopausal women. Two potential biases of this study design merit further discussion. First, it is important to note that 8 of 17 patients with endometrial carcinoma in this study presented with advanced disease. It has been shown that abnormal Doppler findings are more likely to be encountered in advanced carcinomas with larger tumour volumes (57). In addition, the authors were not blinded to the presence of endometrial carcinoma at the time of the ultrasound evaluation.

This same group of investigators also examined uterine arterial flow in 138 postmenopausal women (17 of whom had endometrial carcinoma), using pulsed and colour Doppler imaging (57). These authors concluded that the PI was more accurate than endometrial thickness in differentiating benign from malignant endometria. Using an arbitrary threshold value of 10 mm for endometrial thickness, the rate of cancer detection was 14/17 (82%, 95% CI: 54%-96%). The false-positive rate for asymptomatic women not receiving exogenous

hormones was 4/84 (5%, CI: 1%-12%), while for women on hormonal replacement therapy it was 6/35 (17%, CI: 7%-34%). Corresponding rates for PI using an arbitrary threshold value of 1.50 were as follows: detection rate for endometrial cancer 17/17 (100%, CI: 81%-100%), false positive rate for women not receiving hormonal replacement therapy 1/85 (1%), false positive rate for women on a hormonal regimen 4/35 (11%).

The increase in the false positive fraction for women receiving exogenous hormones is not unexpected, given the lower impedance to pelvic blood flow, in this group of patients (84,94-96). Additional factors resulting in reduced values of uterine artery RI and PI, include increased heart rate, antihypertensive medication, residual ovarian function and uterine leiomyomas (58,97,98). Conversely, values of RI and PI tend to increase with the number of years after menopause (97). A study by Hata et al. (58) using Doppler ultrasound to assess tumour vascularity in gynecologic disorders, demonstrated abnormal flow in 7 of 7 endometrial carcinomas. Abnormal flow was arbitrarily defined as a RI less than 0.7. Similar low impedance flow was demonstrated in 8 of 31 patients (25.8%) with uterine leiomyomas. However, these were reliably differentiated from endometrial carcinomas using morphologic criteria alone. Leiomyomas coexisting with endometrial carcinoma can be seen in up to 29% of cases (3,99).

A potential pitfall may arise in cases of early endometrial carcinoma where morphologic change is not yet evident. Low impedance flow in this instance could falsely be attributed to the presence of a coexisting uterine leiomyoma. Doppler interrogation of endometrial or intratumoural blood flow rather than uterine arterial flow may limit false positive results from concomitant pelvic pathology. In addition, it has been shown that indices of intratumoural blood flow are more sensitive markers of endometrial cancer, than indices obtained from uterine arterial blood flow (56,57,100,101).

Kurjak et al. (59), examined 750 postmenopausal women with transvaginal colour Doppler sonography one day prior to a scheduled hysterectomy for a variety of pelvic pathology. 32 of 35 patients (91%) with proven endometrial carcinoma demonstrated intratumoural or peritumoural blood flow. Endometrial arterial blood flow was absent in normal, atrophic and 92% of hyperplastic endometria. The mean RI obtained from patients with

endometrial carcinoma was 0.42, while for patients with endometrial hyperplasia the mean RI was 0.65 ($p < 0.05$). The same group of investigators examined 5013 asymptomatic women over a 5-year period from January 1988 until December 1992 (82). Entry criteria for the study included 1) women aged 40 years or older, 2) women not receiving hormonal replacement therapy, and 3) no complaints of pelvic symptomatology. A positive finding for malignancy at EVS was regarded as an endometrial thickness of 10 mm or greater, and/or abnormal blood flow with a low resistive index ($\leq 0.42$). Thirty-four (0.68%) cases of endometrial abnormalities were detected and operated on. Of these, there were six patients with endometrial carcinoma, 18 patients with endometrial hyperplasia, and 20 patients with benign endometrial polyps. EVS successfully differentiated malignant from benign endometria in all cases, on the basis of the pulsed Doppler indices.

Aleem et al. (102), studied 42 postmenopausal women prior to dilatation and curettage with endovaginal colour and pulsed Doppler. These investigators found an overlap in measurements of endometrial thickness in patients with endometrial carcinoma and in patients with hyperplasia. Using a cut-off value of $\geq 8$ mm, endometrial carcinoma was differentiated from all benign uterine conditions with a sensitivity of 100%, and a specificity of 60%. Using Doppler ultrasound, vascular visualization rates for endometrial carcinoma were 43% for endometrial vessels and 92% for myometrial vessels, whereas the corresponding rates for endometrial hyperplasia were 12% and 43%, respectively. These authors concluded that the presence of endometrial and myometrial feeder vessels, with low vascular impedance to blood flow and dense vascular arrangement, is suggestive of malignant endometrial conditions. Similar conclusions were drawn by Merce et al (101), who studied 45 patients with metrorrhagia, of whom 21 had abnormal findings at histopathology. They found that measurements of intramyometrial RI were highly accurate in predicting endometrial pathology. Using the mean intramyometrial RI of the control group ($0.79 \pm 0.16$ SD) as a threshold value, the presence of endometrial pathology was predicted with a sensitivity of 84%, a specificity of 86%, a PPV of 84% and a NPV of 86%. The RI of uterine arterial flow was less accurate, with a sensitivity of 81%, and a significant decrease in specificity of 50%.

Other investigators, however, found endometrial thickness to be a better predictor of endometrial pathology than any of the Doppler indices evaluated to date. Sladkevicius et al. (60), examined 138 consecutive women scheduled for curettage because of postmenopausal bleeding with EVS including colour and pulsed Doppler techniques. Receiver-operator characteristic curves showed endometrial thickness to be a better discriminator between benign and malignant endometria than any Doppler variable; 14 mm was the optimal threshold value, the sensitivity being 88% (95% CI: 66%-97%) and the specificity 81% (95% CI: 75%-89%). The best Doppler variable for differentiating between benign and malignant endometria was the presence of colour flow within the endometrium, the sensitivity being 87% (95% CI: 67%-97%), and the specificity 66% (95% CI: 57%-75%). The PI of subendometrial and intraendometrial flow showed considerable overlap between benign and malignant endometria. Differences in sensitivity of Doppler systems to blood flow, as well as in the type of patient population studied may in part account for the discrepancy among published results. Conoscenti et al. (61), studied 149 women with postmenopausal bleeding using EVS and Doppler ultrasound to evaluate the accuracy of one or more sonographic parameters in predicting endometrial pathology. Pathological sonographic criteria included endometrial thickness $\geq$ 8 mm, abnormal endometrial echotexture, and RI of endometrial blood flow $\leq$ 0.4. Using these criteria to distinguish pathological from normal endometrium, EVS showed a sensitivity of 69% (95% CI: 58%-80%), a specificity of 83% (95% CI: 75%-91%), a PPV of 74% (95% CI: 63%-85%), and a NPV of 72% (95% CI: 63%-81%). Considering endometrial thickness as a single parameter, the most sensitive cut-off value for defining normality was < 4mm. This resulted in a sensitivity of 95% (95% CI: 90%-100%), a specificity of 49% (95% CI: 39%-59%), a PPV of 57% (95% CI: 47%-67%), and a NPV of 94% (95% CI: 87%-100%). These authors concluded that measurements of endometrial thickness are preferable to using a combination of sonographic criteria in the evaluation of women with postmenopausal bleeding. Other investigators have also emphasized the limited value of Doppler ultrasound in differentiating benign from malignant endometria (103,104).

*2.2.4. Summary of role of EVS in detecting endometrial pathology*

The available body of literature suggests that EVS, using measurements of endometrial thickness, has a high sensitivity for detecting endometrial carcinoma. Most authors recommend using a low cut-off value such as 4 or 5 mm, which maintains a high sensitivity but sacrifices specificity. This results in many unnecessary curettage procedures being performed in order not to miss a carcinoma. In addition, the number of false positive diagnoses increases with the use of hormonal replacement therapy. Most studies reporting on the role of EVS in detecting endometrial pathology have excluded patients on a hormonal regimen. To better reflect the population at large, we have elected to study all women presenting with postmenopausal bleeding, irrespective of exogenous hormone intake. In addition, although a number of investigators have recently suggested that a combination of biometric and morphologic criteria improves the specificity of EVS in diagnosing endometrial carcinoma, to our knowledge, no large scale study has addressed this issue (23,40,41,83,86,87).

Finally, the role of Doppler ultrasound remains controversial. Earlier studies reported high accuracy rates using Doppler indices of uterine arterial blood flow to differentiate malignant from benign endometria. These results have not been corroborated by other groups of investigators. Differences in patient selection, study design, and Doppler equipment used, may account in part, for the reported discrepancies. We have attempted to study a large consecutive population of postmenopausal women presenting with abnormal vaginal bleeding using standard, commercially available Doppler ultrasound machines.

## 3 - STUDY DESIGN

This chapter begins with a description of the patient population, including a discussion of the selection criteria, as well as sample size calculations. Following this, the equipment and examination technique used is described. Details on data collection, including patient demographics, real-time EVS and Doppler examination are then provided. The standard of reference used to differentiate benign from malignant endometria, and normal from pathological

endometria is defined. Finally, details on the statistical methods used to analyze this dataset are provided.

## 3. 1 Patient Population

### *3.1.1 Inclusion criteria*

The patient population consisted of postmenopausal women referred to EVS for evaluation of abnormal vaginal bleeding from September 1992 until May 1997. The catchment area consisted of clinics and private offices of gynecologists affiliated with the McGill University Health Center. Only patients who were being evaluated for the first time with this complaint were entered into our study protocol. Women were considered postmenopausal if they had not spontaneously menstruated for at least one year. Hormonal assays were not routinely performed to document the onset of menopause. We have chosen to study these particular women because they represent a well-defined group with a relatively high incidence of endometrial carcinoma (105). Although we are aware of the inherent potential for bias in this selection, we felt the additional cost of studying a broader group of patients could not be justified. In addition, since up to 90% of patients with endometrial carcinoma present with abnormal vaginal bleeding, efforts at detecting endometrial carcinoma in a clinical setting, are by and large directed at symptomatic patients (3,4,5). Nevertheless, the results of this study may not be applicable to an asymptomatic population with a lower prevalence of disease. In particular, the positive predictive value of any test would be considerably decreased.

Patients who met our entry criteria were included in the final analysis of results under the following circumstances: 1) the EVS was judged adequate for evaluating the endometrium, 2) a uterine malignancy other than endometrial carcinoma was not present at histopathology, 3) an adequate sample of endometrial tissue was obtained for histological analysis or appropriate clinical follow-up was available (see section 3.3.4 on *standard of reference*).

### *3.1.2 Sample size calculation*

Sample size calculations were based on the estimated difference between two binomials of the variables endometrial thickness and endometrial resistive

index (RI), assuming a desired confidence interval width of 0.2. Given a predicted probability of disease of 0.2 in a group of patients with endometrial thickness ≤ 5 mm, and a predicted probability of 0.7 in a group of patients with endometrial thickness > 5 mm, the required sample size would be 142 patients per group. On the other hand, given a predicted probability of disease of 0.6 in a group of patients with an endometrial RI ≤ 0.5, and a predicted probability of 0.4 in a group of patients with an endometrial RI > 0.5, results in a required sample size of 184 patients per group. Therefore, assuming an approximately 30% exclusion rate (184 x 2 = 368 ~ 550 x 0.70), we aimed to enroll 550 patients into our study protocol.

There are no specific guidelines as to the number of observations recommended per predictor in logistic regression modeling. However, a frequently quoted guideline in linear regression is that there must be at least 10 observations for a given predictor in the model with the largest number of covariates (107). Given the potential of 25 to 30 covariates in our data set, a study population of at least 300 would be needed to meet the above guideline. However, each subject in a logistic regression model provides only a binary outcome, and thereby contributes less information (on average) compared to that provided by each subject in a linear regression on a continuous variable. Therefore, higher ratios of subjects to number of predictor variables might be advisable for logistic regression modeling.

### *3.1.3 Ethical considerations*

This study was approved by the Montreal General Hospital Research and Ethics Committee, and all participating patients gave informed consent.

## 3. 2 Equipment and Examination Technique

Two experienced sonographers performed the endovaginal sonographic examinations. All sonographic examinations were verified and interpreted prospectively during the course of the real-time examination by three radiologists experienced in body imaging (CR, MA, AA). The sonographers and radiologists were aware of the patients' age, menstrual and obstetric history, use

of hormones, general medical and surgical history, other forms of medication, family history and presenting symptoms. The radiologists, however, were blinded to the results of other imaging tests and previous endometrial histology where applicable.

Endovaginal sonographic examinations were performed in the standard fashion using commercially available sonographic equipment (Acuson 128; Acuson, Mountainview, California) (106). The endovaginal sonographic probe produced a 5.0 MHz beam for imaging and a 3.5 MHz pulsed colour Doppler system for blood flow analysis. Prior to use, the end of the probe was covered with a coupling gel, then inserted into a condom and finally recoated with sterile gel before insertion into the vagina.

Transmitter power, image processing and pulse length was held constant. Gain was adjusted for each individual so as to produce consistent and optimal depiction of the uterine zones. Images were obtained at 0.5 cm. intervals in the sagittal and transverse planes relative to the uterus, with special attention given in all cases to the appearance of the endometrium. Systematic examination of the cervix and adnexa was also undertaken.

Colour Doppler was used to provide an overview of the vascularity, obviating in part, a time consuming search of a large area with a gated, pulsed Doppler device. The intensity of the colour produced is proportional to the Doppler frequency shift. By convention, blood flowing towards the transducer was portrayed in red, while flow away from the transducer was modulated in blue. The spatial peak temporal average intensity of the probe was maintained at a minimum of 94mW/cm2 and the wall filter at 100MHz. Colour flow images of the uterine arteries were sampled lateral to the internal cervical os in the longitudinal plane to ensure a consistent anatomic location. In each instance, the angle of insonation was adjusted to obtain maximum colour intensity. A range gate was then placed across the vessel and the cursor of the pulsed Doppler manipulated to obtain maximum waveform amplitude and clarity. Angle correction was used for the pulsed Doppler in all cases of uterine artery sampling.

Endometrial or tumour blood flow was also initially localized using colour flow imaging. Areas demonstrating colour flow were sampled with

pulsed Doppler. The Doppler angle was assumed to be 0 degrees in all cases using duplex Doppler imaging and also in cases where accurate correction was not possible with color flow. In instances where colour Doppler was unsuccessful, the region of interest was interrogated using pulsed Doppler with a wide gate (5-8mm) and maximal gain. When a signal was detected, the sample volume was reduced in the range of 1.5 - 3 mm and the angle of the transducer manually adjusted until the maximum amplitude and frequency shift were obtained. Theoretically, the increased sensitivity of pulsed Doppler may allow detection of tumour vascularity where none is seen using colour flow imaging.

Colour and pulsed Doppler interrogation was limited to 10 mins. to ensure the feasibility of applying this technique in daily clinical practice. Details of real-time sonographic and Doppler findings were recorded on video-tapes, with select images recorded on hard-copy film or digital format.

### 3.3 Data Collection

*3.3.1 Patient demographics*

Immediately prior to the sonographic examination each patient was interviewed and the following demographic data were recorded: 1) patient age, 2) menstrual history (age of menarche, age of menopause), 3) use of hormones (duration and type of agent), 4) general medical and surgical history, 5) other forms of medication, 6) family history of malignancy, and 7) pertinent presenting symptoms.

*3.3.2 Real time examination*

The thickness of the endometrium was measured on the scanner display using digital calipers. For each patient, the double-layer endometrial thickness was recorded by measuring the distance between the ventral and dorsal endo-myometrial interface. Any intraluminal fluid or mass distending the endometrial cavity was subtracted from measurements of endometrial thickness. In addition to endometrial biometry, data on endometrial morphology was obtained as follows: 1) presence or absence of an endometrial mass, 2) endometrial borders: well-defined, focally or diffusely poorly defined (if the poor definition was felt to be secondary to co-existing adenomyosis this was duly noted), 3) endometrial

echogenicity: echogenic, hypoechoic, heterogeneous, 4) cysts or calcification within the endometrial complex/mass, 5) presence of adenopathy or ascites, 6) real-time EVS diagnosis: benign, malignant, indeterminate, 7) real-time EVS diagnosis: nonpathological vs. pathological.

A diagnosis of *endometrial malignancy* was made with EVS when one or more of the following circumstances were present: 1) locally invasive mass, 2) endometrial (mass) borders focally or diffusely ill-defined, 3) abnormal endometrial (mass) echotexture defined as increased echogenicity, decreased echogenicity and/or heterogeneity. The presence of an endometrial mass or endometrial thickening alone, without any of the additional criteria for malignancy was insufficient to make a diagnosis of endometrial carcinoma. Conversely, a patient with a normal endometrial thickness (see definition below) but morphological features suggestive of malignancy would be diagnosed as having endometrial carcinoma. A diagnosis of *benign endometrium* was made when none of the above criteria were present. In patients for whom the sonologist was unable to differentiate a benign from a malignant process, the endometrium was classified as *indeterminate*.

A diagnosis of *endometrial pathology* was made on EVS when 1) any of the previously described criteria for malignancy were present, 2) an endometrial mass was diagnosed, or 3) the endometrium measured > 5 mm in women not on hormone replacement therapy and > 7 mm in women on a hormonal regimen. In the absence of any of these criteria, the endometrium was classified as *nonpathological*.

The presence of any pathology of the cervix, myometrium or adnexa was also noted and documented.

### *3.3.3 Doppler examination*

Doppler indices of the left and right uterine artery, the endometrium or endometrial-based mass, as well as the myometrium were obtained. The myometrium was sampled to potentially serve as a correction factor for Doppler indices obtained from the endometrial complex, since a number of systemic factors may influence the observed Doppler values (58,97,98). Care was taken to only sample areas of the myometrium that appeared normal on EVS. Maximal

systolic (PSV) and end-diastolic (EDV) velocities were averaged over three separate cardiac cycles. For each waveform, the resistive index (RI = PSV - EDV/PSV) and the pulsatility index (PI = PSV -EDV/mean) were calculated using standard software available on our sonographic equipment. The PSV, PI and RI were calculated for all anatomic regions sampled, except for the myometrium where only the RI was obtained. In addition to arterial waveforms, the maximal venous flow velocity was obtained from the endometrial complex in all patients. In the absence of arterial or venous flow, a value of 0 was assigned to the PSV or venous velocity, respectively.

The degree of vascularity of the endometrial complex using colour Doppler was graded from 0 to 3 as follows: Grade 0: absence of colour flow; Grade 1: minimal vascularity defined as the presence of one or two small flecks of colour flow; Grade 2: moderate vascularity defined as the presence of several areas showing colour flow; Grade 3: marked vascularity defined as areas of colour flow present throughout the endometrial complex. In the setting of an endometrial mass, the presence of a single or dominant feeding vessel (stalk flow) was documented.

### 3.3.4 *Standard of reference*

For the differentiation of benign versus malignant endometria, the standard of reference used was endometrial histology (biopsy, D &C, or hysterectomy) or clinical follow-up. Only histological examinations adequate for diagnosing endometrial pathology were used as the standard of reference. In patients without adequate endometrial histology, or in patients who had not undergone endometrial sampling, a presumptive diagnosis of benign endometrium was made if the patients remained asymptomatic with cessation of vaginal bleeding for at least one year after the EVS. Although a number of patients with malignant endometria may have been misclassified as benign in the clinical follow-up group, the probability of that occurring would be on the order of 0.1%. This estimate is drawn from the observation that the incidence of endometrial carcinoma in asymptomatic women is approximately 1%, and that the likelihood of disease decreases to 0.1% after a negative sonographic examination (101).

For the differentiation of nonpathologic versus pathologic endometria, only the subgroup of patients with adequate endometrial histology was analyzed, since patients with benign endometrial pathology may become asymptomatic.

All data analysis was performed with the assumption that the chosen standard of reference was 100% accurate.

### 3.4 Statistical Analysis

*3.4.1 Accuracy of real-time EVS in diagnosing endometrial carcinoma*

Descriptive statistics are provided where deemed appropriate. The sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and accuracy of EVS (using real-time parameters of endometrial thickness and morphology to diagnose endometrial carcinoma) were calculated. Accuracy is defined as the number of true positive and true negative diagnoses over the total patient population studied. Sensitivity calculations were performed for the total patient population, and were then stratified according to hormone use. In addition, all sensitivity calculations were performed three-ways as follows: 1) indeterminate category (real-time EVS diagnosis) treated as malignant, 2) indeterminate category treated as benign, and 3) indeterminate category excluded. For each test characteristic, a 95% confidence interval (CI) around each point estimate of test performance was obtained by using the standard normal approximation of the binomial distribution.

*3.4.2 Logistic regression analysis*

To estimate the predictive value of various morphological features and Doppler ultrasound in diagnosing endometrial carcinoma, logistic regression analysis was performed.

To compare cases (malignant endometria) and controls (benign endometria) on all baseline variables, univariate analysis was used (SPSS 6.0 software). For continuous variables, results are presented as mean ± standard deviation (SD) and as median with interquartile range for variables with heavily skewed distributions. Dichotomous and categorical variables are presented as proportion (%) affected in each outcome group. Means were compared using

Student's t-test. Medians (where appropriate) were compared using the Mann-Whitney test, a nonparametric two sample median test. Proportions were compared using chi-square test, or Fisher exact test for cells with expected cell frequency less than 5. Differences in means and proportions and the associated 95% confidence intervals are presented.

To determine the presence of highly correlated variables that may be collinear or confounders in the association between a given variable and case status, Spearman's rank correlations were calculated. To investigate the role of covariates as potential confounders and/or effect modifiers based on substantive evidence and preliminary results, stratified analysis of the predictor / disease relationship controlling for the various covariates was performed. The odds ratios obtained from the stratified analysis were then compared to the crude odds ratio. When the strength of the association between a given predictor variable and disease status was different across strata, effect modification by the stratified variable on the predictor variable was further investigated in the multivariate analysis.

Logistic regression analysis was performed using the bic.logit procedure (S-plus software), which is based on a Bayesian method of model selection (108). Standard frequentist model selection methods typically rely on P-value based significance tests. A limitation of the frequentist approach is that P values for individual variables change, depending on the number of variables entered into a model, and the selection methods used. Therefore, using a P value cut-off as a basis for variable inclusion or exclusion, can be extremely misleading. A second limitation is that although several models may appear quite appropriate, each can lead to an entirely different conclusion about the question being studied. Therefore, selecting a single model in this context, ignores issues of model uncertainty and the resulting uncertainty of inferences.

The Bayesian approach to hypothesis testing and model selection overcomes these difficulties to a large extent. Uncertainty about the unknown parameters of the model is expressed in terms of probability of the parameter given the data, using Bayes' Theorem. The bic.logit procedure provides estimates of the beta coefficients for each predictor variable using the mean of individual model estimates, weighted by the posterior probabilities of each parameter. For

model selection, the Bayesian Information Criterion (BIC) or Schwartz Criterion (SC) provides an accurate approximation to Bayes' factor, which is the ratio of the integrated likelihoods of the models being compared. For example, if we compare two models, $M_2$ vs. $M_1$ and this ratio > 1, the data favour $M_2$ over $M_1$. The magnitude of the Bayes' factor in comparing the two models can be used to assess the strength of this evidence, where the BIC is a function of the likelihood ratio statistic or model deviance:

$$BIC_K = L_K^2 - df_K \log n$$

Where $L_K^2$ is the deviance for model $M_K$, n is the sample size, and $df_K$ is the corresponding number of degrees of freedom. Models can be compared by taking the difference of their BIC values. Models with smaller values for BIC (i.e. more negative), will have a better model fit. In addition, the magnitude of the difference in BICs can be used to assess the strength of one model against another. Finally, model uncertainty is expressed as the posterior probability that each model is true, given that one of the models must be true. This probability is derived from the BIC value (108). Use of these methods and their interpretation will be presented in Section 4.4.8 on Multivariate analysis.

Variables judged to be important predictors of malignant endometria on clinical grounds, univariate or bivariate analysis, and interaction terms that were judged potentially significant based on the results of the stratified analysis, were entered into the logistic regression program. Final model selection was based on the BIC value.

Since no model can be expected to precisely predict case status, some deviation from predicted outcome is to be expected. The width of the 95% CI surrounding the parameter was used as an estimate of the degree of accuracy with which the parameter was known.

*3.4.3 Receiver operating characteristics curves analysis*

Receiver operating characteristics (ROC) curves were generated to determine the optimal threshold value of endometrial thickness and the various

Doppler indices in distinguishing benign from malignant endometrial pathology. The LABROC1 program for continuous variables (IBM version 1.2, University of Chicago) was used for generating ROC curves. Only variables judged to be important predictors of endometrial carcinoma on clinical grounds, or by the univariate and bivariate analysis, were used in generating the ROC curves. The 95% CI for the difference in the areas ($A_Z$) under the ROC curve in patients with and without hormone use were calculated using the formula: $(A_{Z1} - A_{Z2}) \pm 1.96 \sqrt{(SE_1^2 + SE_2^2)}$. For comparing the areas under two ROC curves derived form the same sample of patients, the method described by Hanley and McNeil was used (109). In addition, a sensitivity analysis was performed for the different cut-off points for each variable.

## 4 - RESULTS

This chapter begins with a description of the study population, as well as the group of patients excluded from the study. Following this the accuracy of real-time EVS in differentiating benign from malignant endometria, and normal from pathological endometria are presented. Finally, details of the logistic regression analysis and ROC curve analysis are provided.

### 4.1 Description of study population

Five hundred and fifty-seven patients met our inclusion criteria and were entered into our study protocol. Of these, 136 patients were excluded for the following reasons: 1) no endometrial histology or clinical follow-up [n = 58], 2) no endometrial histology, and clinical follow-up performed less than 1 year after the EVS [n = 64], 3) no endometrial histology, and patients remained symptomatic at 1 year follow-up [n = 7], 4) uterine malignancy other than endometrial carcinoma [n = 3], 4) non-diagnostic EVS [n = 3], and 5) partial hysterectomy prior to the EVS [n = 1]. Therefore, 421 patients comprised our final study population. The mean age was 60 years (range 38 – 88 years). Of the 421 patients, 170 (40.4%) were not on a hormonal regimen, while 251 (59.6%) were on hormonal replacement therapy as follows: estrogen [n = 84], progesterone [n = 20], or a combination regimen [n = 147].

*Table II* compares age, hormonal status, mean endometrial thickness at EVS, and overall diagnosis at EVS (benign, malignant, indeterminate) in the study population and in the group of patients excluded from our study protocol.

***Table II. Demographic and Endovaginal Sonographic Diagnosis on Study Population and Patients Excluded from Study Protocol.***

| | Study population | Excluded patients |
|---|---|---|
| **Mean Age (± SD)** | 60.0 ± 9.2 years | 60.4 ± 9.2 years |
| **Hormones n (%)** | 251/421 (59.6%)<br>(95% CI: 55%-64%) | 72/136 (52.9%)<br>(95% CI: 45%-62%) |
| **Mean endo thick† (± SD)** | 6.83 (± 6.77)<br>(95% CI: 6.18-7.46) | 4.16 (± 5.25)<br>(95% CI: 3.26-5.06) |
| **EVS diagnosis** | | |
| **Benign** | 334 (79%)<br>(95% CI: 75%-83%) | 125 (92%)<br>(95% CI: 86%-96%) |
| **Malignant** | 43 (10%)<br>(95% CI: 8%-14%) | 0<br>(95% CI: 0%-2%) |
| **Indeterminate** | 44 (11%)<br>(95% CI: 8%-14%) | 11 (8%)<br>(95% CI: 4%-14%) |
| **TOTAL** | **421 (100%)** | **136 (100%)** |

† Mean endometrial thickness at EVS (mm)

The differences in the two populations as outlined in Table II, reflects the trend in current clinical practice, where patients with a thin endometrium and benign morphologic features on EVS, may not undergo further investigation. Although the proportion of patients on hormonal replacement therapy in the excluded group is slightly lower than in the study population, the 95% CIs overlap considerably. This trend is not unexpected given the more benign appearance of endometria on EVS in patients not on hormonal replacement. In addition, there may have been less concern on the part of the physician and patient regarding underlying endometrial pathology in patients not taking exogenous hormones.

Of the 421 patients in our final study group, 307 or 72.9% underwent endometrial sampling or surgery as follows: endometrial biopsy [n = 104, 33.9%], D & C [n = 178, 58.0%], or hysterectomy [n = 25, 8.1%]. Seven of 104

(6.7%) endometrial biopsies were interpreted as inadequate for diagnosis, while 14/178 (7.9%) D & Cs were judged inadequate. Therefore, endometrial histology was used as the standard of reference in 286 patients or 67.9%. The time delay between EVS and histology was 75.3 days (range 1 – 768 days). All endometrial carcinomas except two were diagnosed at histology within 127 days of the EVS. In two patients, the diagnosis of endometrial carcinoma was made after a time delay of 302 and 336 days, respectively. As both of these patients continued to be symptomatic, and the neoplasms were well to moderately differentiated, these tumours were likely present at the time of the EVS, and therefore were included for analysis.

In the remaining 135 patients or 32.1% without endometrial histology, a presumptive diagnosis of benign endometrium was made, since all patients remained asymptomatic with cessation of vaginal bleeding after a mean follow-up of 3 years (range 1 – 5 years).

### 4.2 Accuracy of real-time EVS in differentiating benign from malignant endometria

Of the 421 patients in our study population, 390 patients or 92.6% had benign endometria, while 31 or 7.4% had malignant endometria. Real-time EVS using a combination of biometric and morphologic criteria characterized the endometria as follows: benign [n = 333], malignant [n = 45], or indeterminate [n = 43].

Although EVS was considered diagnostic in all patients included for final analysis (see inclusion criteria detailed in section 3.1), in 20 patients or 4.8%, the visualization of the endometrium was considered suboptimal.

*Table III* lists the distribution of patients according to hormone use, diagnosis at real-time EVS, and presence of malignancy at final diagnosis using the previously defined standard of reference.

***Table III. Distribution of patients according to results of EVS versus final diagnosis in the differentiation of benign from malignant endometria.***

| *EVS diagnosis* † | *Hormone use* †† | *Final diagnosis* † | *Total (n = 421)* |
|---|---|---|---|
| – | – | – | n = 127 |
| – | + | – | n = 199 |
| – | – | + | n = 2 |
| – | + | + | n = 5 |
| + | – | + | n = 12 |
| + | + | + | n = 9 |
| + | – | – | n = 9 |
| + | + | – | n = 15 |
| ± | – | – | n = 18 |
| ± | + | – | n = 22 |
| ± | – | + | n = 2 |
| ± | + | + | n = 1 |

† + positive for malignancy, – negative for malignancy, ± diagnosis indeterminate

†† + patients receiving hormones , – patients *not* receiving hormones

*Table IV* lists the sensitivity, specificity, PPV, NPV and accuracy of real-time EVS using a combination of biometric and morphologic criteria to differentiate benign from malignant endometria. In addition to presenting the results for the total patient population, the results are stratified according to hormone use. The patients with an indeterminate diagnosis at real-time EVS were analyzed separately in *Tables IV-A,B,C* as follows: 1) indeterminate EVS diagnoses counted as malignant, 2) patients with indeterminate EVS diagnoses excluded, and 3) indeterminate EVS diagnoses counted as benign.

***Table IV-A. Accuracy of EVS in differentiating benign from malignant endometria (indeterminate EVS diagnoses counted as malignant)***

| | *All patients* | *Patients without hormones* | *Patients on hormones* |
|---|---|---|---|
| *TOTAL* | *421* | *170* | *251* |
| Sensitivity† (95%CI) | 77% (59% – 90%) | 88% (62% - 98%) | 67% (38% - 88%) |
| Specificity (95%CI) | 84% (80% - 87%) | 82% (75% - 88%) | 84% (79% - 89%) |
| PPV †† (95%CI) | 27% (18% - 38%) | 34% (20% - 51%) | 21% (11% - 36%) |
| NPV (95%CI) | 98% (96% - 99%) | 98% (95% - 100%) | 98% (94% - 99%) |
| Accuracy (95%CI) | 83% (79% - 87%) | 83% (76% - 88%) | 83% (78% - 88%) |

† Sensitivity: 95% CI for difference in patients with and without hormones = - 0.08, 0.50

†† PPV: 95% CI for difference in patients with and without hormones = - 0.06, 0.32

***Table IV-B. Accuracy of EVS in differentiating benign from malignant endometria (patients with indeterminate EVS diagnoses excluded)***

| | *All patients* | *Patients without hormones* | *Patients on hormones* |
|---|---|---|---|
| *TOTAL* | *378* | *150* | *228* |
| Sensitivity† (95%CI) | 75% (55% - 89%) | 86% (57% - 98%) | 64% (35% - 87%) |
| Specificity (95%CI) | 93% (90% - 95%) | 93% (88% - 97%) | 93% (89% - 96%) |
| PPV†† (95%CI) | 47% (32% - 62%) | 57% (34% - 78%) | 38% (19% - 59%) |
| NPV (95%CI) | 98% (89% - 94%) | 98% (95% - 100%) | 98% (94% - 99%) |
| Accuracy (95%CI) | 92% (89% - 94%) | 93% (87% - 96%) | 91% (87% - 95%) |

† Sensitivity: 95% CI for difference in patients with and without hormones = - 0.10, 0.52

†† PPV: 95% CI for difference in patients with and without hormones = - 0.09, 0.48

***Table IV-C. Accuracy of EVS in differentiating benign from malignant endometria (indeterminate EVS diagnoses counted as benign)***

| | *All patients* | *Patients without hormones* | *Patients on hormones* |
|---|---|---|---|
| *TOTAL* | *421* | *170* | *251* |
| Sensitivity† (95%CI) | 68% (49% - 83%) | 75% (48% - 93%) | 60% (32% - 84%) |
| Specificity (95%CI) | 94% (91% - 96%) | 94% (89% - 97%) | 94% (90% - 96%) |
| PPV†† (95%CI) | 47% (32% - 62%) | 57% (34% - 78%) | 38% (19% - 59%) |
| NPV (95%CI) | 97% (95% - 99%) | 97% (93% - 99%) | 97% (94% - 99%) |
| Accuracy (95%CI) | 92% (89% - 94%) | 92% (87% - 96%) | 92% (87% - 95%) |

† 95% CI for difference in sensitivity in patients with and without hormones : - 0.18, 0.48

†† 95% CI for difference in PPV in patients with and without hormones : - 0.09, 0.48

As illustrated in the preceding tables, our data do not present evidence for a difference in the sensitivity, specificity, PPV, NPV and accuracy of diagnosing endometrial carcinoma in patients with and without hormone use. The 95% confidence intervals for differences in proportions all include the null value (see tables footnotes). Nevertheless, there appears to be a trend towards a lower sensitivity and PPV in patients receiving hormonal replacement therapy that can be interpreted in one of two ways. First, the observed differences may simply represent random variation. Alternatively, given the wide 95% confidence intervals around the estimated parameters, the number of observations may have been too small to demonstrate a true difference. Exogenous hormones are known to result in morphological alterations of the endometrium on EVS that may decrease the accuracy in diagnosing endometrial carcinoma in this group of patients. However, the decreased accuracy in patients on hormones is usually attributable to a *lower specificity* when biometric criteria are used to differentiate malignant from benign endometria. The trend towards a *decrease in sensitivity* that we observed using real-time sonographic findings most probably reflects the sonologists application of more

stringent criteria, given the expected proliferative changes of the endometrium in patients receiving hormone replacement therapy.

The indeterminate category on EVS was treated in three different ways in the calculation of accuracy to account for both extremes in diagnostic probabilities. Nevertheless, this group of patients would be managed clinically as potentially malignant, and therefore require further investigation.

### 4.3 Accuracy of real-time EVS in differentiating normal from pathological endometria

In order to investigate the accuracy of EVS in differentiating normal from pathological endometria, a separate analysis of patients that had undergone histopathological correlation [n= 286] was undertaken. This group of patients was further subdivided into normal endometria or endometria demonstrating physiological change [n = 129, or 45%], and pathological endometria [n= 157, or 55%]. The final histological diagnoses of pathological endometria were as follows: simple hyperplasia [n = 28], complex hyperplasia [n = 7], proliferative and simple hyperplasia [n = 5], atypical hyperplasia [n = 12], polyps [n = 71], polyps and atypical hyperplasia [n = 3], and carcinoma [n = 31].

*Table V* lists the distribution of patients according to hormone use, diagnosis at real-time EVS, and presence or absence of endometrial pathology in the subgroup of patients for whom histopathological correlation was available.

***Table V. Distribution of patients according to results of EVS versus final diagnosis in the differentiation of normal from pathological endometria.***

| *EVS diagnosis* † | *Hormone use* †† | *Final diagnosis* † | *Total (n = 286)* |
|---|---|---|---|
| – | – | – | n = 24 |
| – | + | – | n = 54 |
| – | – | + | n = 9 |
| – | + | + | n = 8 |
| + | – | + | n = 61 |
| + | + | + | n = 79 |
| + | – | – | n = 18 |
| + | + | – | n = 33 |

† + positive for endometrial pathology, – negative for endometrial pathology

†† + patients receiving hormones , – patients *not* receiving hormones

*Table VI* lists the sensitivity, specificity, PPV, NPV and accuracy of real-time EVS using a combination of biometric and morphologic criteria to differentiate normal from pathologic endometria. In addition to presenting the results for all patients with histopathological correlation, the results are stratified according to hormone use.

***Table VI. Accuracy of EVS in differentiating pathologic endometria from normal endometria***

| | *All patients* | *Patients without hormones* | *Patients on hormones* |
|---|---|---|---|
| *TOTAL* | *286* | *112* | *174* |
| Sensitivity (95%CI) | 89% (83% - 94%) | 87% (77% - 94%) | 91% (83% - 96%) |
| Specificity (95%CI) | 60% (52% - 69%) | 57% (41% - 72%) | 62% (51% - 72%) |
| PPV (95%CI) | 73% (66% - 79%) | 77% (66% - 86%) | 71% (61% - 79%) |
| NPV (95%CI) | 82% (73% - 89%) | 73% (54% - 87%) | 87% (76% - 94%) |
| Accuracy (95%CI) | 76% (71% - 81%) | 76% (67% - 83%) | 76% (69% - 83%) |

Again, we did not find evidence of differences in the sensitivity, specificity, PPV, NPV and accuracy of diagnosing endometrial pathology in patients with and without hormone use, although some confidence intervals are wide. When comparing the results of real-time EVS in diagnosing pathological endometria (*Table VI*) versus endometrial malignancy (*Table IV-A*) we found a number of important differences. The accuracy for detecting all endometrial pathology was significantly lower than the accuracy for detecting malignancy alone (95% CI for the difference: 0.01, 0.13). This was primarily due to a decrease in the specificity (95% CI: 0.14, 0.32) and NPV (95% CI: 0.08, 0.24). Although the sensitivity improved from 77% to 89%, the confidence interval was wide and included 0 (95% CI: -0.27, 0.04). There was evidence of an increase in the PPV from 27% to 73% (95% CI: -0.57, -0.35). However, this increase can be explained,

in part, by the high prevalence of endometrial pathology relative to endometrial malignancy, 55% versus 7.4%, respectively.

## 4.4 Logistic regression analysis

We begin this section by defining all variables considered in the regression analysis. Following this, a descriptive analysis is presented for all continuous and dichotomous/categorical variables. Histograms for the continuous variables deemed to be of interest for further modeling are then provided. Linearity verification for all continuous variables is performed, and selected plots illustrated. The treatment of missing values is discussed. Finally, the results of the univariate, bivariate and multivariate analyses are presented.

### *4.4.1 Definition of Variables*

Dependent / outcome variable

**HISTDXLR**: Dichotomous variable.

The outcome variable is the presence (code = 1) or absence (code = 0) of endometrial carcinoma using our previously defined standard of reference.

Covariates

**AGE:** Continuous variable measured in years (see Section 4.4.4, Linearity Verification).

The incidence of endometrial carcinoma is known to have a bimodal shape with two peaks. However, the greatest increase in endometrial carcinoma occurs after menopause, and since only post-menopausal women were included in our study, the influence of age on outcome will be less marked.

**MEDICLR**: Dichotomous variable.

The presence (code = 1) or absence (code = 0) of hormonal replacement therapy. Since the ingestion of estrogen-like substances increases the thickness and vascularity of the endometrium, hormone use is a potential confounder and effect modifier.

**THICKLR1/THICKLR2:** Continuous variable.

Two subvectors (THICKLR1 /THICKLR2) were created for this continuous variable (ENDO_THICK) (see Section 4.4.4, Linearity Verification). The variable ENDO_THICK refers to endometrial thickness (measured in mm) at EVS. Extensive literature exists on the use of endometrial thickness as a predictor of endometrial carcinoma, with a thickness of ≤ 5mm indicating a low probability of disease (THICKLR2), and a thickness of > 5mm indicating a higher probability of disease (THICKLR1). In addition, irrespective of the association with endometrial carcinoma, a thicker endometrium, due to increased tissue volume, may be associated with positive Doppler findings. Therefore, this variable should be examined for effect modification and confounding.

**USM_NOMA**: Dichotomous variable.
Presence (code = 1) or absence (code = 0) of an endometrial mass at EVS. Endometrial carcinomas may present as an endometrial mass or as diffuse thickening of the endometrial complex. Conversely, an endometrial mass even in a postmenopausal woman frequently represents a benign endometrial polyp.

**US_MASS**: Continuous variable.
This variable refers to the mean size (measured in cm) of an endometrial mass when present. Since it only applies to patients with a measurable endometrial mass at EVS, it will only be used in a subset analysis of this group of patients.

**EVS_CYST:** Dichotomous variable.
Presence (code = 1) or absence (code = 0) of cysts within the endometrial complex or an endometrial-based mass. The presence of cysts tends to favour a benign process, although cysts may also be encountered in the setting of malignant endometria.

**EVS_CAL:** Dichotomous variable.
Presence (code = 1) or absence (code = 0) of calcification within the endometrial complex or an endometrial-based mass. Although calcification tends to favour a slow growing process, little is known regarding the predictive value of this variable.

**EVS_DEFLR**: Dichotomous variable.
This variable refers to the endometrial borders as being poorly defined (code = 1) or well defined (code = 0). Poorly defined endometrial borders may be seen in association with malignant endometria. Patients with poorly defined endometrial borders attributable to the presence of adenomyosis (a benign uterine condition) were coded as 0, since the small number of observations in this category precluded separate analysis, resulting in the generation of unstable parameter estimates.

**EVS_ECHO**: Categorical variable.
Defines the echogenicity of the endometrium as hyperechoic (code = 1), hypoechoic (code = 2), or heterogeneous (code = 3). Although the echotexture of endometrial carcinoma is variable, one would expect a higher proportion of malignancies in categories 2 and 3.

**VENOUS**: Continuous variable (see Section 4.4.4, *Linearity Verification).*
Measures the peak venous velocity (cm/sec) of the endometrial complex or an endometrial-based mass. Little is known regarding the predictive value of this variable in differentiating benign from malignant endometria. However, studies of other malignancies have demonstrated increased venous flow. This variable should be examined for confounding, since increased venous velocities may be associated with increased tissue volumes (thick endometria, large mass size) independent of any association with endometrial carcinoma.

**LUA_PSV / RUA_PSV** : Continuous variables.
Left and right uterine artery peak systolic velocity (PSV, cm/sec). Increased PSV of the uterine arteries has been associated with endometrial carcinoma. However, it is generally felt to be a less sensitive indicator than endometrial PSV.

**LUA_PI / RUA_PI**: Continuous variables.
Left and right uterine artery pulsatility index (PI). Decreased PI of the uterine arteries has been associated with endometrial carcinoma. However, it is generally felt to be a less sensitive indicator than endometrial PI.

**LUA_RI / RUA_RI**: Continuous variables.
Left and right uterine artery resistive index (RI). Decreased RI of the uterine arteries has been associated with endometrial carcinoma. However, it is generally felt to be a less sensitive indicator than endometrial RI.

**END _DOPP**: Dichotomous variable.
Presence (code = 1) or absence (code = 0) of pulsed Doppler arterial flow from the endometrial complex or an endometrial-based mass. The presence of endometrial flow is more likely to be associated with malignant endometria. This variable was created to facilitate modeling the variables RI and PI, since no meaningful values of PI and RI exist in the absence of arterial Doppler flow (PSV = 0). A correlation is expected between END_DOPP and the covariates VASCUL, PSV since both are measures of arterial flow. Therefore, particular attention should be paid to confounding by this variable.

**PSV**: Continuous variable (see Section *4.4.4, Linearity Verification*).
PSV (cm/sec) of arterial flow obtained from the endometrial complex or an endometrial-based mass. Malignant endometria demonstrate higher mean peak velocities relative to benign endometria.

**PI**: Continuous variable (see Section *4.4.4, Linearity Verification*).
PI of arterial flow obtained from the endometrial complex or an endometrial-based mass. Malignant endometria demonstrate lower mean values for PI relative to benign endometria.

**PISQ**: Continuous variable (see Section *4.4.4, Linearity Verification*).
The square term of the variable PI.

**RI**: Continuous variable (see Section *4.4.4, Linearity Verification*).
RI of arterial flow obtained from the endometrial complex or an endometrial-based mass. Malignant endometria demonstrate lower mean values for RI relative to benign endometria.

**MY_PSV**: Continuous variable.
PSV (cm/sec) of arterial flow obtained from areas of normal myometrium. The myometrium was sampled to potentially serve as a correction factor for values of PSV obtained from the endometrial complex, since a number of systemic factors may influence the observed Doppler values.

**MY_RI**: Continuous variable.
RI of arterial flow obtained from areas of normal myometrium. The myometrium was sampled to potentially serve as a correction factor for the RI obtained from the endometrial complex, since a number of systemic factors may influence the observed Doppler values.

**MY_EN_PS**: Continuous variable.
This variable was generated by forming a simple ratio of myometrial PSV to endometrial PSV. The decision to form a simple ratio as a potential correction factor is somewhat arbitrary, given the absence of available data on relative changes in endometrial and myometrial PSV under varied systemic conditions.

**MY_EN_RI**: Continuous variable.
This variable was generated by forming a simple ratio of myometrial RI to endometrial RI. The decision to form a simple ratio as a potential correction factor is somewhat arbitrary, given the absence of available data on relative changes in endometrial and myometrial RI under varied systemic conditions.

**VASCUL**: Ordinal variable (see Section *4.4.4, Linearity Verification*).
Refers to the vascularity of the endometrium using color Doppler and is graded from 0 to 3. A correlation is expected between VASCUL and the covariate END _DOPP since both measure blood flow albeit in different ways and with different

sensitivities. Therefore, particular attention should be paid to confounding by this variable.

**STALK**: Dichotomous variable.
Presence (code = 1) or absence (code = 0) of stalk flow due to a single dominant feeding vessel within an endometrial-based mass. Stalk flow is frequently associated with benign endometrial polyps.

**EVS_DX**: Categorical variable.
This refers to the diagnosis made with real-time EVS, using morphological features, but none of the Doppler indices. The categories used included benign endometrium (code = 1), malignant endometrium (code = 2), or indeterminate (code = 3). Although one may have treated this as an ordinal variable, we elected not to do so, since the category "indeterminate" has special implications clinically and cannot just be considered half-way between benign and malignant. The relationship between the covariates END_DOPP, VASCUL and our dependent variable may change depending on the particular category of EVS_DXLR, therefore this variable has potential for effect modification.

*4.4.2 Descriptive Analysis*

*Table VII-A* presents means and standard deviations (SD), as well as quartiles, for each continuous variable for the total patient population. *Table VII-B* presents frequencies (percentages) of events for dichotomous / categorical variables for the total patient population.

***Table VII-A. Continuous variables***

| *Variable* | *Mean (SD)* | *25 %* | *Median (Min - Max)* | *75%* |
|---|---|---|---|---|
| AGE | 60.0 (9.2) | 53 | 58 (38 - 88) | 66 |
| THICKLR1 | 5.1 (7.6) | 0 | 0.0 (0.0 - 44.0) | 8.1 |
| THICKLR2 | 1.7 (1.7) | 0 | 1 .6 (0 - 5.3) | 3.0 |
| US_MASS | 1.7 (1.1) | 0.9 | 1.5 (0.2 - 4.6) | 2.3 |
| VENOUS | 2.7 (4.1) | 0 | 2 (0 - 54) | 4 |
| LUA_PSV | 29 (19) | 17 | 24 (3 - 163) | 37 |
| LUA_PI | 2.48 (1.01) | 1.80 | 2.40 (0.50 – 8.87) | 3.04 |
| LUA _RI | 0.87 (0.11) | 0.81 | 0.89 (0.38 – 1.00) | 0.94 |
| RUA _PSV | 26 (19) | 17 | 26 (4 - 150) | 37 |
| RUA _PI | 2.50 (0.99) | 1.80 | 2.37 (0.58 – 7.87) | 3.01 |
| RUA _RI | 0.87 (0.10) | 0.82 | 0.89 (0.43 – 1.00) | 0.94 |
| PSV | 5 (8) | 0 | 2 (0 - 53) | 7 |
| PI | 1.06 (0.41) | 0.78 | 1.01 (0.37 - 3.62) | 1.25 |
| RI | 0.61 (0.13) | 0.52 | 0.61 (0.3 - 1.0) | 0.7 |
| MY_PSV | 10 (8) | 5 | 8 (0 - 65) | 13 |
| MY_RI | 0.77 (0.13) | 0.67 | 0.78 (0.26 – 1.00) | 0.86 |
| VASCUL | 0.9 (0.8) | 0 | 1 (0 – 3) | 1 |

***Table VII-B. Categorical variables (N = 421)***

| *Variable* | *Coding Scheme* | *Frequency (Percentage)* |
|---|---|---|
| HISTDXLR | 0 | 390 (92.6) |
| | 1 | 31 (7.4) |
| MEDICLR | 0 | 170 (40.4) |
| | 1 | 251 (59.6) |
| USM_NOMA | 0 | 308 (73.2) |
| | 1 | 113 (26.8) |
| EVS_CYST | 0 | 353 (83.8) |
| | 1 | 68 (16.2) |
| EVS _CAL | 0 | 405 (96.2) |
| | 1 | 16 (3.8) |
| EVS _DEFLR | 0 | 356 (84.6) |
| | 1 | 65 (15.4) |
| EVS _ECHO | 1 | 333 (79.1) |
| | 2 | 12 (2.8) |
| | 3 | 76 (18.1) |
| END _DOPP | 0 | 208 (49.4) |
| | 1 | 213 (50.6) |
| STALK | 0 | 347 (82.4) |
| | 1 | 74 (17.6) |
| EVS_DX | 1 | 333 (79.1) |
| | 2 | 45 (10.7) |
| | 3 | 43 (10.2) |

As can be seen from *Table VII-B*, the frequency of positive outcomes (HISTDXLR = 1) is 31 or 7.4% (95% CI: 5 - 10%). Although this result is in keeping with the range of 8 - 10% reported in the literature, the trend towards a slightly lower prevalence of disease in our patient population is likely due to the large number of women on hormonal replacement therapy. Of patients *not* taking exogenous hormones, 16/170 or 9.4% (95% CI: 5 - 15%) were diagnosed with endometrial carcinoma; while 15/251 or 6.0% (95% CI: 3 - 10%) carcinomas were encountered in patients on hormonal replacement therapy. Women receiving hormones present more frequently with abnormal vaginal bleeding, due to the occurrence of a variety of benign endometrial conditions associated with hormone use.

*4.4.3 Distribution of data and outliers*

The examination of boxplots and histograms revealed several univariate *outliers* for many of the continuous variables, both extreme (3 x IQR) and others (1.5 x IQR). Verification of the raw data revealed that these were not data entry errors, but represented true values that were biologically plausible. Therefore, the univariate analysis was run with and without the outliers. Although the ORs changed minimally for several of the variables, the overall inferences did not change. Therefore, these observations were included in all further analysis.

Histograms for the continuous variables deemed to be of interest for further modeling after the univariate analysis (see *Section 4.4.6, Univariate Analysis*), are included below.

***Figure 1. Histogram for variable AGE***

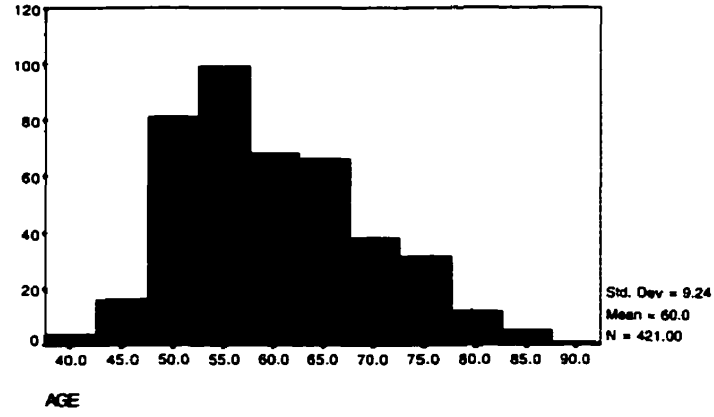



***Figure 2. Histogram for variable ENDO _THICK***

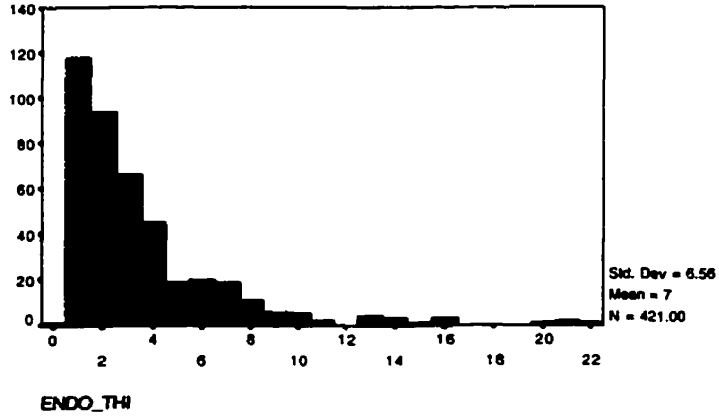


*Figure 3. Histogram for variable PSV*

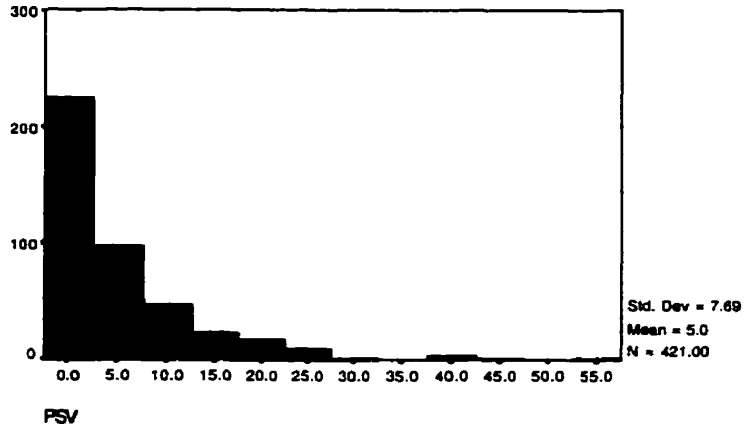



*Figure 4. Histogram for variable RI*

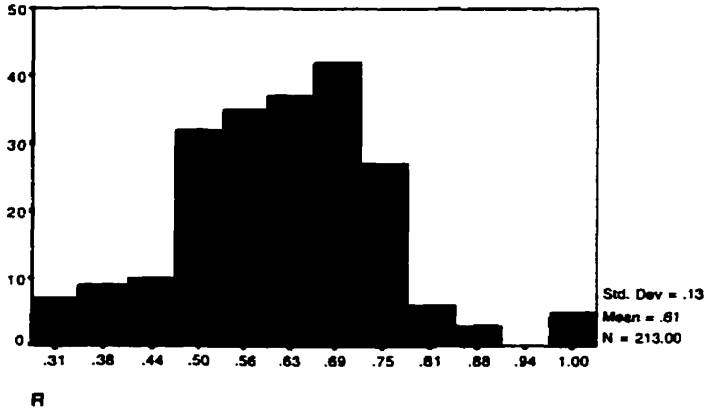



*Figure 5. Histogram for variable PI*

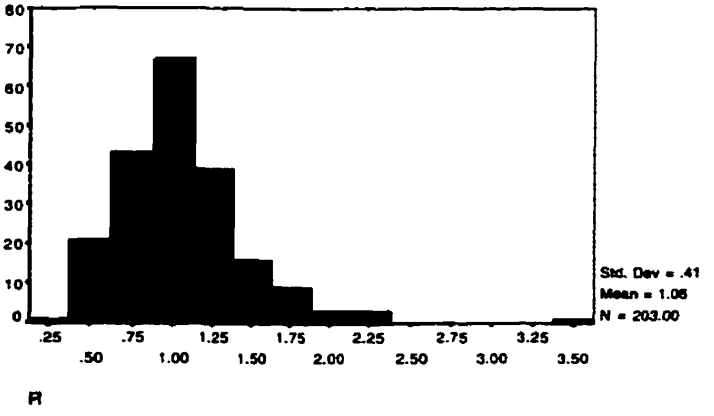


*Figure 6. Histogram for variable VENOUS*

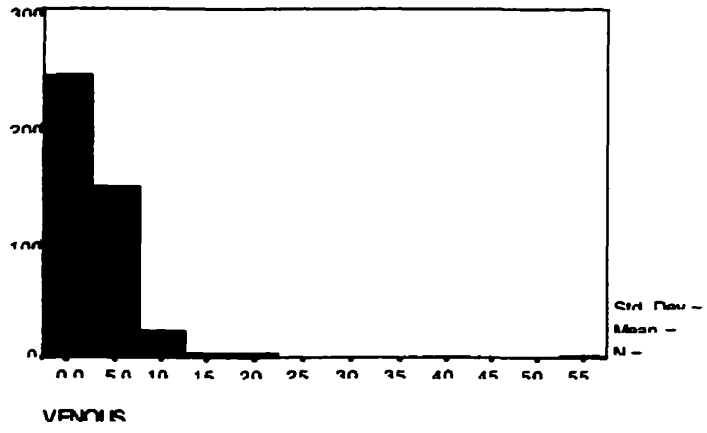
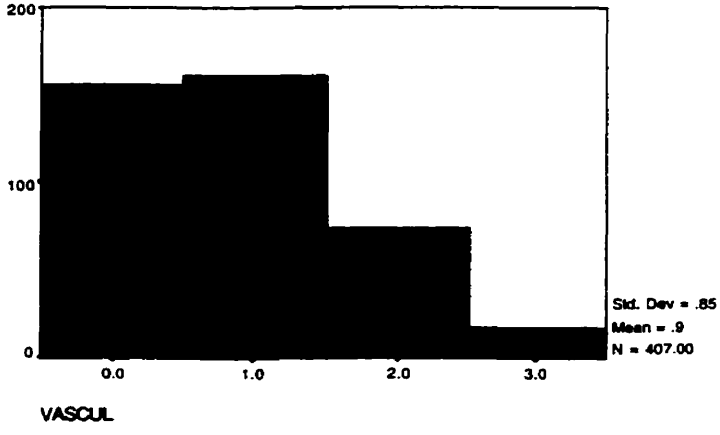



*Figure 7. Histogram for variable VASCUL*




### 4.4.4 *Linearity Verification*

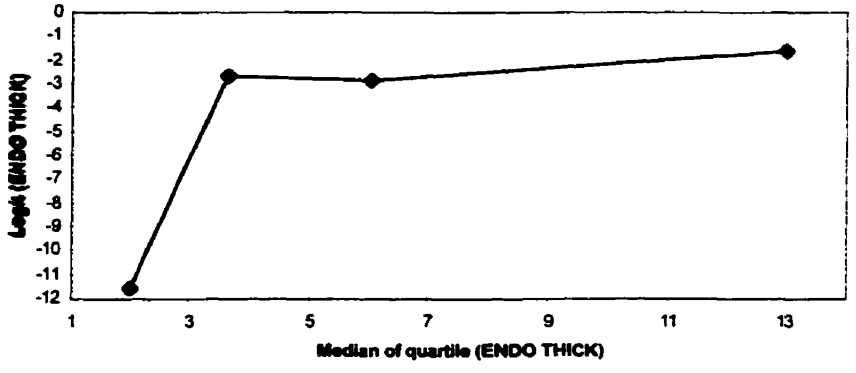
There are 15 continuous variables within this data set. To verify the assumption of linearity in the logit, the observations for each variable were separated into quartiles, and then the median of each quartile was plotted against the logit [ln (cases/controls)] for each quartile. Care was taken to use the same scale for the logit (y-axis) for all variables during the initial evaluation. Plots for the variables LUA_PSV, LUA_PI, LUA_RI, RUA_PSV, RUA_PI, RUA_RI, MY_PSV, MY_RI did not appear to violate the assumption of linearity. Since these

variables were removed from further modeling after the univariate analysis (see *Section 4.4.6, Univariate Analysis*), these plots are not provided here.

Plots of linearity for the remaining variables are illustrated in the ensuing pages. Except for the variables ENDO-THICK and PI, no assumptions of linearity are clearly violated. Note that the logit range for the variable ENDO_THICK is considerably wider than for the other variables, suggesting that ENDO_THICK is more predictive of disease outcome. For purposes of illustration, a narrow logit range was used for all variables except ENDO_THICK to avoid excessive compression in the y-axis.

***Figure 8. Linearity verification for ENDO_THICK***

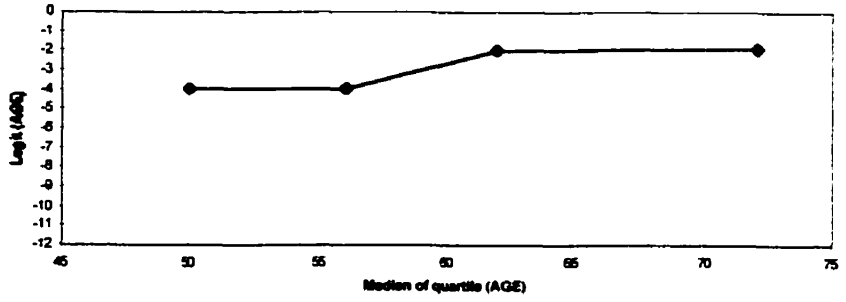
| Quartiles | 1st | 2nd | 3rd | 4th |
|---|---|---|---|---|
| **Median of quartiles** | 2 | 3.6 | 6 | 13 |
| **Logit** | -11.57 | -2.64 | -2.80 | -1.58 |




The plot for ENDO_THICK does not appear to be linear. Based on the above one can consider a number of options. Although our data suggests that ENDO_THICK may be dichotomized at 3mm, this goes against a large body of substantiative knowledge indicating that the risk of cancer begins to increase with ENDO_THICK > 5mm. Given the appearance of the plot, it appears most reasonable to model the 2 slopes by parameterizing each indicator. Therefore we generated two subvectors as follows: 1) THICKLR1 = ENDO_THICK, if ENDO_THICK > 5mm; and 0 otherwise 2) THICKLR2 = ENDO_THICK, if ENDO_THICK ≤ 5mm, and 0 otherwise.

*Figure 9. Linearity verification for AGE*

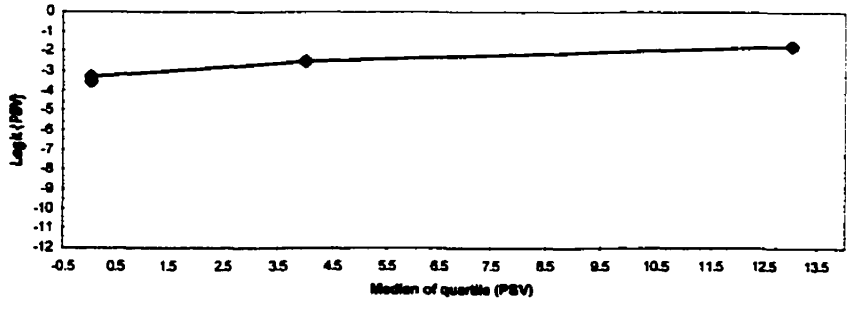
| Quartiles | 1st | 2nd | 3rd | 4th |
|---|---|---|---|---|
| **Median of quartiles** | 50 | 56 | 62 | 72 |
| **Logit** | -3.95 | -3.94 | -1.96 | -1.87 |




As illustrated, the plot for AGE appears to be essentially linear. Based on the above we will maintain AGE as a continuous variable in the regression analysis.

*Figure 10. Linearity verification for PSV*

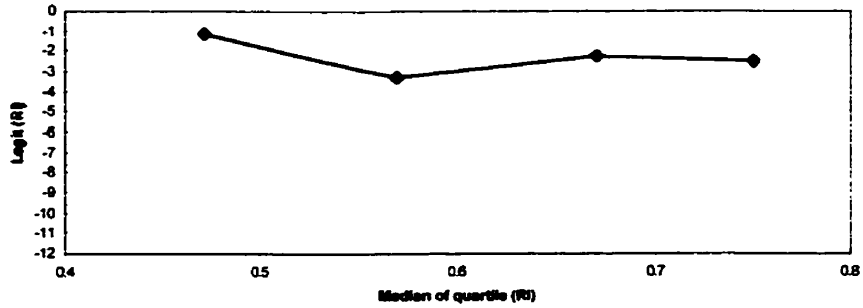
| Quartiles | 1st | 2nd | 3rd | 4th |
|---|---|---|---|---|
| **Median of quartiles** | 0 | 0 | 4 | 13 |
| **Logit** | -3.54 | -3.23 | -2.50 | -1.72 |




The plot for PSV does appear to be relatively linear. Based on the above we will continue to use PSV as a continuous variable.

*Figure 11. Linearity verification for RI*

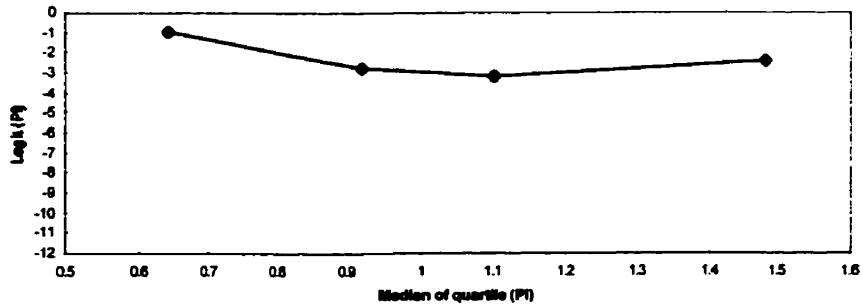
| Quartiles | 1st | 2nd | 3rd | 4th |
|---|---|---|---|---|
| **Median of quartiles** | 0.47 | 0.57 | 0.67 | 0.75 |
| **Logit** | -1.15 | -3.24 | -2.26 | -2.51 |




The plot for RI does appear to be relatively linear. Based on the above we will continue to use RI as a continuous variable.

*Figure 12. Linearity verification for PI*

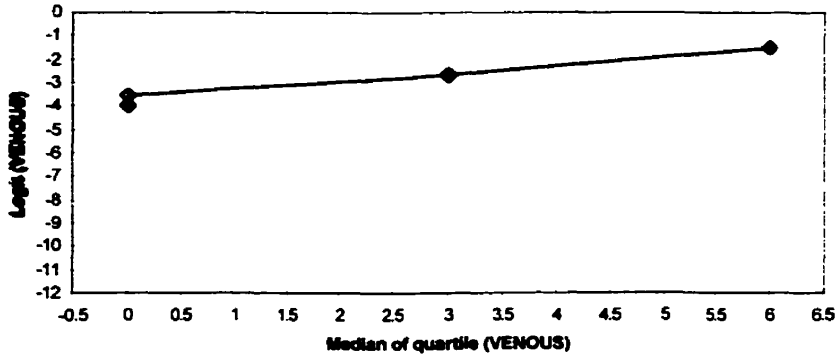
| Quartiles | 1st | 2nd | 3rd | 4th |
|---|---|---|---|---|
| **Median of quartiles** | 0.64 | 0.92 | 1.10 | 1.48 |
| **Logit** | -0.97 | -2.77 | -3.20 | -2.4 |




The plot for PI suggests a slight parabolic shape. Although disease status has been shown to correlate with low values of PI, a potential correlation at high levels of PI has some theoretical basis. Therefore, a square term of PI (PISQ) will be investigated in the univariate analysis.

*Figure 13. Linearity verification for VENOUS*

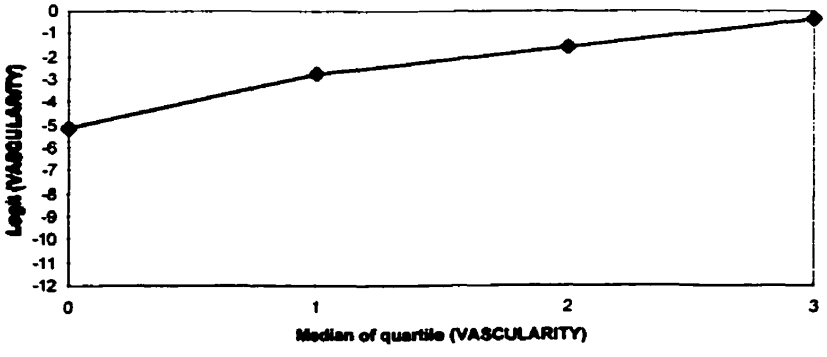
| Quartiles | 1st | 2nd | 3rd | 4th |
|---|---|---|---|---|
| **Median of quartiles** | 0 | 0 | 3 | 6 |
| **Logit** | -3.95 | -3.53 | -2.64 | -1.51 |




The plot for VENOUS does appear to be relatively linear. Based on the above we will continue to use VENOUS as a continuous variable.

*Figure 14. Linearity verification for VASCUL (Vascularity)*

| Quartiles | 1st | 2nd | 3rd | 4th |
|---|---|---|---|---|
| **Median of quartiles** | 0 | 1 | 2 | 3 |
| **Logit** | -5.12 | -2.71 | -1.55 | -0.36 |




The plot for VASCULAR does appear to be relatively linear. Based on the above we will continue to use VASCULAR as a continuous variable.

*4.4.5 Missing values*

No missing values were encountered for any of the dichotomous/ categorical variables. The maximum number of missing values for the continuous variables were as follows: VASCUL [n = 14], VENOUS [n = 2], RI [n = 208], PI [n = 218]. Since the variables RI and PI are highly correlated (r = .84), and therefore provide similar information, only RI was entered into the multivariate analysis (see *Section 4.4.7, Bivariate analysis*). Given the large number of missing values for the predictor RI, the multivariate analysis was run with and without this variable. Since the most predictive models returned by the bic.logit procedure were similar for both runs, and the best models excluded RI, the variable RI was omitted for the final analysis (see *Section 4.4.8, Multivariate analysis*). Therefore, we did not loose much precision in estimation due to missing data since overall less than 5% of cases had any missing values.

*4.4.6 Univariate analysis*

Means and standard deviations (SD), as well as quartiles, are presented for each continuous variable in *Table VIII-A* according to disease status. The first row of each variable represents patients with benign endometria (HISTDXLR = 0) while the second row represents patients with malignant endometria (HISTDXLR = 1). A 95% CI for the difference in the mean between the two groups for each variable is given.

As is presented in *Table VIII-A*, the difference in the means according to disease status for the variables AGE, endometrial thickness (THICKLR1/THICKLR2), and Doppler indices of the endometrium (VENOUS, PSV, RI, PI, VASCUL), results in 95% CIs that exclude the null value. Nevertheless, only the parameter estimate and corresponding 95% CI for the variables endometrial thickness and VENOUS, have any potential clinical significance. In addition, none of the Doppler indices of the uterine arteries or myometrium, demonstrate a clinically important difference between the two groups when considering the point estimates and 95% CI.

## *Table VIII-A. Continuous variables*

| *Variable* | *HISTDXLR* † | *Mean (SD)* | *25 %* | *Median (Range)* | *75%* | *95% CI for Δ in mean* |
|---|---|---|---|---|---|---|
| AGE | 0 | 59.6 (9.2) | 52 | 57 (38 - 85) | 66 | (-9.18, -2.45) †† |
| | 1 | 65.4 (8.3) | 60 | 65 (47 - 88) | 72 | |
| THICKLR1 | 0 | 4.6 (6.9) | 0 | 0.0 (0 - 42.0) | 7.8 | (-9.94, -4.56) †† |
| | 1 | 11.8 (11.5) | 0 | 9.0 (0 - 44.0) | 17.0 | |
| THICKLR2 | 0 | 1.8 (1.7) | 0 | 1.8 (0 - 5.3) | 3.0 | (0.20, 1.46) †† |
| | 1 | 0.9 (1.6) | 0 | 0.0 (0 - 5.3) | 3.0 | |
| US_MASS | 0 | 1.6 (1.1) | 0.9 | 1.2 (0.2 - 4.6) | 2.2 | (-0.94, 0.08) |
| | 1 | 2.1 (0.8) | 1.5 | 1.9 (0.9 - 3.8) | 2.6 | |
| VENOUS | 0 | 2.4 (2.9) | 0 | 1 (0 - 21) | 4 | (-6.62, -3.74) †† |
| | 1 | 7.5 (10.1) | 3 | 5 (0 - 54) | 7 | |
| LUA_PSV | 0 | 30 (19) | 17 | 25 (3 - 163) | 38 | (0.09, 13.89) †† |
| | 1 | 23 (14) | 13 | 21 (6 - 70) | 28 | |
| LUA_PI | 0 | 2.51 (1.02) | 1.80 | 2.40 (0.50 - 8.87) | 3.06 | (-0.35, 0.72) |
| | 1 | 2.17 (0.79) | 1.66 | 2.14 (0.51 - 3.60) | 2.77 | |
| LUA _RI | 0 | 0.87 (0.11) | 0.81 | 0.90 (0.40 - 1.00) | 0.94 | (-0.01, 0.07) |
| | 1 | 0.84 (0.14) | 0.79 | 0.88 (0.38 - 1.00) | 0.93 | |
| RUA _PSV | 0 | 30 (19) | 17 | 26 (4 - 150) | 38 | (-2.44, 11.69) |
| | 1 | 25 (11) | 18 | 23 (7 - 47) | 33 | |
| RUA _PI | 0 | 2.51 (1.00) | 1.80 | 2.37 (0.58 - 7.87) | 3.01 | (-0.25, 0.49) |
| | 1 | 2.39 (0.83) | 1.72 | 2.48 (0.68 - 4.00) | 2.93 | |
| RUA _RI | 0 | 0.87 (0.10) | 0.82 | 0.89 (0.43 - 1.00) | 0.94 | (-0.02, 0.06) |
| | 1 | 0.85 (0.12) | 0.80 | 0.87 (0.52 - 1.00) | 0.94 | |
| PSV | 0 | 4 (7) | 0 | 0 (0 - 46) | 7 | (-9.94, -4.47) †† |
| | 1 | 12 (12) | 3 | 8 (0 - 53) | 19 | |
| PI | 0 | 1.08 (0.41) | 0.83 | 1.04 (0.37 - 3.62) | 1.26 | (0.05, 0.41) †† |
| | 1 | 0.85 (0.34) | 0.63 | 0.69 (0.47 - 1.51) | 1.04 | |
| RI | 0 | 0.62 (0.13) | 0.54 | 0.61 (0.32 - 1.00) | 0.70 | (0.03, 0.14) †† |
| | 1 | 0.54 (0.15) | 0.42 | 0.50 (0.30 - 0.75) | 0.68 | |
| MY_PSV | 0 | 10 (8) | 5 | 8 (0 - 65) | 13 | (-1.48, 4.21) |
| | 1 | 9 (6) | 6 | 7 (0 - 25) | 14 | |
| MY_RI | 0 | 0.77 (0.13) | 0.68 | 0.78 (0.26 - 1.00) | 0.86 | (-0.04, 0.07) |
| | 1 | 0.75 (0.16) | 0.66 | 0.80 (0.35 - 1.00) | 0.86 | |
| VASCUL | 0 | 0.8 (0.8) | 0 | 1 (0 - 3) | 1 | (-1.33, -0.74) †† |
| | 1 | 1.8 (0.8) | 1 | 2 (0 - 3) | 2 | |

† HISTDXLR = 0 refers to benign endometria, HISTDXLR = 1 refers to malignant endometria

†† 95% CI does not include the null value

*Table VIII-B* presents frequencies (percentages) of events for dichotomous / categorical variables according to disease status. A 95% CI for the difference in the proportion between the two groups for each variable is given.

Note that for all variables *except* MEDICLR, EVS_CYST, EVS CAL, the 95% CI does not include the null value. In particular, the observed differences in proportions and corresponding 95% CI for the variables USM_NOMA, EVS_DEFLR, and EVS_DX demonstrate a clinically important difference between the two groups.

***Table VIII-B. Categorical variables***

| *Variable* | *Coding Scheme* | *HISTDXLR = 0* † *(n = 390)* | *HISTDXLR = 1* † *(n = 31)* | *95% CI for Δ in $p_0/p_1$* |
|---|---|---|---|---|
| MEDICLR | 0 | 154 (39.5) | 16 (51.6) | (-0.30, 0.06) |
| | 1 | 236 (60.5) | 15 (48.4) | |
| USM_NOMA | 0 | 295 (76.4) | 10 (32.3) | (0.26, 0.60) †† |
| | 1 | 92 (23.6) | 21 (67.7) | |
| EVS_CYST | 0 | 325 (83.3) | 28 (90.3) | (-0.18, 0.04) |
| | 1 | 65 (16.7) | 3 (9.7) | |
| EVS _CAL | 0 | 376 (96.4) | 29 (93.5) | (-0.06, 0.12) |
| | 1 | 14 (3.6) | 2 (6.5) | |
| EVS _DEFLR | 0 | 344 (88.2) | 12 (38.7) | (0.32, 0.67) †† |
| | 1 | 46 (11.8) | 19 (61.3) | |
| EVS _ECHO | 1 | 319 (81.8) | 14 (45.2) | (0.19, 0.55) †† |
| | 2 | 10 (2.6) | 2 (6.5) | |
| | 3 | 61 (15.6) | 15 (48.4) | |
| END _DOPP | 0 | 201 (51.5) | 7 (22.6) | (0.13, 0.44) †† |
| | 1 | 189 (48.5) | 24 (77.4) | |
| STALK | 0 | 327 (83.8) | 20 (64.5) | (0.02, 0.37) †† |
| | 1 | 63 (16.2) | 11 (35.3) | |
| EVS_DX | 1 | 326 (83.6) | 7 (22.6) | (0.46, 0.76) †† |
| | 2 | 24 (6.2) | 21 (67.7) | |
| | 3 | 40 (10.3) | 3 (9.7) | |

† HISTDXLR = 0 refers to benign endometria, HISTDXLR = 1 refers to malignant endometria

†† 95% CI does not include the null value

A univariate analysis was run for each predictor tabulated above with the dependent variable being HISTDXLR. For dichotomous predictors, dummy variables were created with the groups assigned code "0" representing the reference category to facilitate interpretation. For categorical predictors the indicator contrast was used and dummy variables created such that the first category represents the reference category.

***Table IX. Results: Univariate analysis***

| *Variable* | *β* | *S.E.* | *df* | $e^{\beta}$(OR) | *95% CI* ($e^{\beta}$) |
|---|---|---|---|---|---|
| **AGE** | .0634 | .0193 | 1 | 1.0654 | (1.03, 1.11) |
| Constant | -6.4842 | 1.2633 | 1 | | |
| **MEDICLR** | -.4909 | .3740 | 1 | .6121 | (0.29, 1.27) |
| Constant | -2.2643 | .2627 | 1 | | |
| **THICKLR1** | .0813 | .0231 | 1 | 1.0847 | (1.04. 1.13) |
| **THICKLR2** | -.0138 | .1545 | 1 | .9863 | (0.73, 1.34) |
| Constant | -3.1249 | .3987 | 1 | | |
| **USM_NOMA** | 1.9172 | .4023 | 1 | 6.8018 | (3.09, 14.96) |
| Constant | -3.3945 | .3215 | 1 | | |
| **US_MASS** | .7005 | .1392 | 1 | 2.0147 | (1.53, 2.65) |
| Constant | -3.0902 | .2524 | 1 | | |
| **EVS_CYST** | -.5878 | .6236 | 1 | .5556 | (0.16, 1.89) |
| Constant | -2.4880 | .2003 | 1 | | |
| **EVS_CAL** | .6162 | .7801 | 1 | 1.8519 | (0.40, 8.54) |
| Constant | -2.5621 | .1927 | 1 | | |
| **EVS_DEFLR** | 2.4715 | .4008 | 1 | 11.8401 | (5.40, 25.97) |
| Constant | -3.3557 | .2937 | 1 | | |
| **EVS_ECHO** | | | | | |
| EVS_ECHO(2) | 1.5167 | .8213 | 1 | 4.5571 | (0.91, 22.79) |
| EVS_ECHO(3) | 1.7233 | .3970 | 1 | 5.6030 | (2.57, 12.20) |
| Constant | -3.1261 | .2731 | 1 | | |
| **VENOUS** | .2221 | .0471 | 1 | 1.2487 | (1.14, 1.37) |
| Constant | -3.4263 | .3000 | 1 | .0000 | |
| **LUA_PSV** | -.0288 | .0143 | 1 | .9717 | (0.94, 1.0) |
| Constant | -1.8127 | .3793 | 1 | | |
| **LUA_PI** | -.4133 | .2281 | 1 | .6615 | (0.42, 1.03) |
| Constant | -1.5780 | .5340 | 1 | | |
| **LUA_RI** | -2.0624 | 1.5154 | 1 | .1271 | (0.01, 2.48) |
| Constant | -.7835 | 1.2950 | 1 | .5452 | |

***Table IX. Results: Univariate analysis (continued)***

| *Variable* | *β* | *S.E.* | *df* | $e^{\beta}$(OR) | *95% CI* ($e^{\beta}$) |
|---|---|---|---|---|---|
| **RUA_PSV** | -.0164 | .0127 | 1 | .9837 | (0.96, 1.01) |
| Constant | -2.1017 | .3761 | 1 | | |
| **RUA_PI** | -.1308 | .2060 | 1 | .8774 | (0.59, 1.31) |
| Constant | -2.2133 | .5293 | 1 | | |
| **RUA_RI** | -1.5401 | 1.7198 | 1 | .2144 | (0.01, 6.23) |
| Constant | -1.2197 | 1.4841 | 1 | | |
| **END_DOPP** | 1.2937 | .4413 | 1 | 3.6461 | (1.54, 8.66) |
| Constant | -3.3574 | .3845 | 1 | | |
| **PSV** | .0759 | .0176 | 1 | 1.0788 | (1.04, 1.12) |
| Constant | -3.0813 | .2544 | 1 | | |
| **PSV** | .0723 | .0189 | 1 | 1.0750 | (1.04, 1.12) |
| **MY_PSV** | -.0353 | .0309 | 1 | .9653 | (0.91, 1.03) |
| Constant | -2.7003 | .3664 | 1 | | |
| **PSV** | .0767 | .0178 | 1 | 1.0798 | (1.04, 1.12) |
| **MY_EN_PS** | -.0036 | .0134 | 1 | .9964 | (0.97, 1.02) |
| Constant | -3.0773 | .2546 | 1 | | |
| **PI** | -1.9758 | .7619 | 1 | .1386 | (0.03, 0.62) |
| Constant | -.1719 | .7034 | 1 | | |
| **PISQ** | -.0027 | .0115 | 1 | .9973 | (0.98, 1.02) |
| Constant | -2.0489 | .2243 | 1 | | |
| **PI** | -3.3410 | 1.7413 | 1 | .0354 | (0.00, 1.07) |
| **PISQ** | .6584 | .6768 | 1 | 1.9317 | (0.51, 7.28) |
| Constant | .4553 | 1.0289 | 1 | | |
| **RI** | -5.3231 | 1.7911 | 1 | .0049 | (0.00, 0.16) |
| Constant | 1.0121 | 1.0041 | 1 | | |
| **RI** | -4.8465 | 1.9122 | 1 | .0079 | (0.00, 0.33) |
| **MY_RI** | .0638 | 1.8005 | 1 | 1.0659 | (0.03, 36.33) |
| Constant | .6794 | 1.4453 | 1 | | |
| **RI** | -5.0160 | 1.8273 | 1 | .0066 | (0.00, 0.24) |
| **MY_EN_RI** | .0081 | .0121 | 1 | 1.0081 | (0.98, 1.03) |
| Constant | .8084 | 1.0350 | 1 | | |
| **VASCUL** | 1.3523 | .2353 | 1 | 3.8664 | (2.44, 6.13) |
| Constant | -4.2613 | .4433 | 1 | | |
| **STALK** | 1.0490 | .3998 | 1 | 2.8548 | (1.30, 6.25) |
| Constant | -2.7942 | .2303 | 1 | | |
| **EVS_DX** | | | | | |
| EVS_DX(2) | 3.7065 | .4848 | 1 | 40.7105 | (15.74, 105.29) |
| EVS_DX(3) | 1.2497 | .7100 | 1 | 3.4895 | (4.26, 14.31) |
| Constant | -3.8400 | .3818 | 1 | | |

The decision as to whether to consider a given covariate for multivariate analysis was based on a combination of apriori knowledge on the importance of the predictor, and the strength of the association as indicated by the crude odds ratio (OR, expressed as $e^{\beta}$ in Table IX) and associated 95% CI.

Based on this premise, the covariates LUA_PSV, LUA_RI, LUA_PI, RUA_PSV, RUA_RI, RUA_PI, were excluded from further analysis. For all of these predictors the 95% CI around the odds ratio includes the null value. In addition, it is generally believed that Doppler indices of the endometrial complex are better predictors of disease status than Doppler indices of the uterine arteries. The covariates EVS_CAL and EVS_CYST, were also excluded from further analysis due to lack of evidence of any important association with disease status. The covariates MY_PSV, MY_EN_PS, MY_RI, and MY_EN_RI, did not add to the predictive power of PSV or RI, respectively, and therefore were removed from further modeling. Similarly, the strength of association of PI did not improve with the addition of the square term, PISQ, indicating that the linearity assumption is not clearly violated.

The most important risk factors judging from the crude odds ratio appear to be the covariates USM_NOMA, EVS_DEFLR, VASCUL, and EVS_DX where the odds of disease in patients with real-time sonographic findings suggestive of malignancy relative to those with a benign endometrium is substantially increased. Other covariates that appear to be risk factors for the outcome include AGE, THICKLR1/THICKLR2, US_MASS, EVS_ECHO, VENOUS, END_DOPP, PSV, STALK, RI and PI. Note that high values of RI and PI appear to have a protective effect, in keeping with what is known about these Doppler indices. Although the covariate MEDICLR does not appear to be an important predictor of outcome as indicated by the crude odds ratio, due to its potential role as a confounder and effect-modifier, it will be entered into the multivariate analysis.

*4.4.7 Bivariate analysis*

To determine the presence of highly correlated variables, which may be collinear, or confounders in the association between a given variable and case

status, Spearman's rank correlations were run. *Table X* details the covariates with high correlation coefficients $|r| \geq .4$.

***Table X. Spearman's rank correlation with variables with*** $|r| \geq .4$

| | VASCUL | END_DOPP | PSV | RI | EVS_DX | US_MASS | USM_NOMA |
|---|---|---|---|---|---|---|---|
| VASCUL | — | .65 | .73 | — | — | .45 | .42 |
| PSV | — | — | — | — | — | .45 | .43 |
| END_DOPP | — | — | .92 | — | — | — | — |
| PI | — | — | — | .84 | — | — | — |
| EVS_DEFLR | — | — | — | — | .55 | — | — |
| EVS_ECHO | — | — | — | — | .42 | — | — |
| STALK | .47 | — | .51 | — | — | .49 | .49 |
| VENOUS | .61 | — | — | — | — | — | — |

As is illustrated in *Table X*, the covariates RI and PI are highly correlated and therefore, only one of these predictors will be run in the multivariate analysis. Since both RI and PI are measures of impedance to flow, and there is no strong evidence as to which variable is a more sensitive indicator, RI will be maintained in the regression analysis since it has less missing data.

The covariates END_DOPP, PSV and VASCUL are also highly correlated indicating that all three variables provide similar information. This will make it extremely difficult to separate out what effects are attributable to one or more of these variables. Furthermore, any effects due to one variable may be split among the others, making it appear that none of these variables are important predictors. Since END_DOPP was created largely to facilitate modeling the variables RI and PI (no meaningful values of PI and RI exist when PSV = 0), it would appear reasonable to omit END_DOPP from the regression analysis. Instead, the complete model can be run first with RI included, and then rerun with RI omitted. The covariates PSV and VASCUL will be investigated separately by adding them into the most predictive models returned by the Bayesian Information Criterion (BIC), and determining whether this improves the fit of the model.

The other variables demonstrating high correlation coefficients will be further investigated in the stratified analysis discussed below to evaluate for any important confounding effects.

To investigate the role of covariates as potential *confounders*, stratified analysis of the predictor / disease relationship controlling for the various covariates was performed, and these odds ratios were compared to the crude odds ratios presented in *Table IX*. The decision to investigate a pair of covariates using stratified analysis was both hypothesis-driven, based on previously known confounding effects between variables, and data-driven. Important confounding was felt to be supported by the data when the parameter estimate and associated 95% CI for the total population fell outside of the range of parameter estimates presented by the different strata.

The following pairs of variables were examined for confounding based on the high correlation coefficients presented in *Table X*: EVS_DX and EVS_DEFLR, EVS_DX and EVS_ECHO, VASCUL and US_MASS, VASCUL and USM_NOMA, PSV and US_MASS, PSV and USM_NOMA, STALK and US_MASS, STALK and USM_NOMA, STALK and PSV, VENOUS and VASCUL. There was no evidence for confounding effects among any of these variable pairs (stratified analysis not shown).

Stratified analysis was also used to explore the role of covariates as potential *effect-modifiers*. Effect modification was suspected if there was a difference in the strength of association between disease status and a predictor variable for the different levels of the stratification variable. For effect modification, only a priori, substantive effects were considered. Thus, only variables with evidence of an association with disease status in one level of the stratum and a non-significant association in the other level of the stratum (as determined by the 95% CI around the parameter estimate) were considered for entry into the multivariate model.

The following variables were investigated for confounding and effect-modification based on apriori theory: EVS_DX and VASCUL, EVS_DX and PSV, THICKLR1/LR2 and VASCUL, THICKLR1/LR2 and PSV. VENOUS and US_MASS. In addition the following variables were stratified according to hormone use

(MEDICLR): EVS_DX, THICKLR1/LR2, VASCUL, VENOUS. Only selected variables demonstrating positive effects are presented in the following tables.

***Table XI. Stratification of EVS_DX according to hormone use (MEDICLR)***

| | MEDICLR = 0 | MEDICLR = 1 | MEDICLR = ALL |
|---|---|---|---|
| | $e^{\beta}$ (95% CI) | $e^{\beta}$ (95% CI) | $e^{\beta}$ (95% CI |
| EVS_DX(2) † | 84.6659 (16.38 - 434.55) | 23.8705 (7.10-80.24) | 40.7105 (15.74-105.29) |
| EVS_DX(3) †† | 7.0555 (0.94 - 53.24) | 1.8084 (0.20-16.18) | 3.4895 (0.87-14.03) |

† Malignant, †† Indeterminate

The odds ratios ($e^{\beta}$) of EVS_DX(2) / EVS_DX(3) for the total patient population (MEDICLR = ALL) are contained within the range obtained across the different strata, indicating no strong evidence for confounding effects, even though the small sample size for EVS_DX(2) leads to large CIs. In addition, since the association with disease status does not change across the different strata, there is unlikely to be substantial effect modification. It is of interest to note that most of the effect of EVS_DX(3) appears to be in the MEDICLR = 0 group. To our knowledge there is no strong apriori theory for this observation.

***Table XII. Stratification of VASCUL according EVS_DX***

| | EVS_DX = 1† | EVS_DX = 2†† | EVS_DX = 3††† | EVS_DX = ALL |
|---|---|---|---|---|
| | $e^{\beta}$ (95% CI) | $e^{\beta}$ (95% CI) | $e^{\beta}$ (95% CI) | $e^{\beta}$ (95% CI) |
| VASCUL | 12.2755 (3.30-45.73) | 1.2075 (0.62-2.34) | 1.0683 (0.28-4.07) | 3.8664 (2.44-6.13) |

† Benign, †† Malignant, ††† Indeterminate

Stratifying the variable VASCUL (endometrial vascularity) according to EVS_DX results in substantial differences in the association with disease status across the strata. An interaction term will be added to the model.

*4.4.8 Multivariate analysis*

A matrix of thirteen predictor variables was entered into the bic.logit program of S-plus. As discussed in Section *3.4 Statistical Analysis*, this logistic

regression program uses the Bayesian Information Criterion (BIC) to select the set of best models, from among the set of all possible models (for our purposes, $2^{13}$ possible combinations). Variables entered into the regression analysis were those associated with case status in the univariate analysis, and those associated with potential confounding or interaction effects in the bivariate analysis. The dependant variable was case status (HISTDXLR), where cases had malignant endometria and controls had benign endometria. For model selection we excluded models that were 20 or more times *less* likely than the best model to predict case status, according to the BIC.

*Table XIII* displays the characteristics of the four most likely models *including* the variable RI, while *Table XIV* displays the characteristics of the four most likely models *excluding* the variable RI. Although RI was not in any of the top models in either case, results are different because missing RI data caused deletion of some cases.

***Table XIII. Logistic regression analysis, variable RI included***

| *Model order* | *Variables in Model* | *P(Model \| data)* | *BIC* † | *Δ BIC* |
|---|---|---|---|---|
| 1st | AGE, EVS_DX, VENOUS | .25 | -1009.674 | – |
| 2nd | AGE, EVS_DX | .23 | -1009.486 | 0.2 |
| 3rd | EVS_DX, VENOUS | .12 | -1008.141 | 1.5 |
| 4th | AGE, USM_NOMA, EVS_DX | .06 | -1006.907 | 2.8 |

† Bayesian Information Criterion

***Table XIV. Logistic regression analysis, variable RI excluded***

| *Model order* | *Variables in Model* | *P(Model \| data)* | *BIC* † | *Δ BIC* |
|---|---|---|---|---|
| 1st | EVS_DX, VENOUS | .59 | -2375.539 | – |
| 2nd | AGE, EVS_DX, VENOUS | .14 | -2372.754 | 2.8 |
| 3rd | USM_NOMA, EVS_DX, VENOUS | .12 | -2372.531 | 3.0 |
| 4th | EVS_DX, VENOUS, STALK | .05 | -2370.660 | 4.9 |

† Bayesian Information Criterion

The most probable model generated by the bic.logit program (excluding the variable RI, *Table IV*), contained the variables EVS_DX (overall diagnosis with

EVS: 1 = benign, 2 = malignant, 3 = indeterminate) and VENOUS (peak venous velocity of endometrial complex. The posterior probabilities, or p (model | data), are interpreted as follows: considering all possible models, these are the relative probabilities of each model assuming that one of them must be correct. Therefore the posterior probability that the 1st model in *Table XIV* is the correct model is .59. For the 2nd and 3rd most likely models in *Table XIV*, the posterior probabilities are .14 and .12 respectively. The 4th model has a probability of .05, and therefore is approximately 12 times less likely to be the true model than the 1st model. The Δ BIC between the 1st and the 2nd or 3rd model indicates positive evidence that the 1st model is more likely, given the data. The 2nd and 3rd models are probably equally likely. *Table XV* adapted from Raftery (108) offers a number of guidelines for model selection.

***Table XV. Guidelines for model selection***

| *Δ BIC* | *P(Model \| data)* | *Evidence* |
|---|---|---|
| 0 - 2 | 50 – 75 | weak |
| 2 – 6 | 75 – 95 | positive |
| 6 – 10 | 95 – 99 | strong |
| > 10 | > 99 | very strong |

When evaluating the four most likely models generated by the bic.logit program with the variable RI included (*Table XIII*), a number of observations can be drawn. First, none of the four most likely models actually contain the variable RI, indicating that this variable is not a strong predictor of case status. Second, the 1st model generated by the data showed only weak-to-positive evidence of being more likely than the 4th model. Therefore, the first three models are approximately equally likely, and were only slightly more predictive than the 4th model. Note that the combination of variables EVS_DX, VENOUS appears as the 1st most likely model with the variable RI excluded (*Table XIV*), and the 3rd most likely model with RI included (*Table XIII*). Given the increased precision obtained by running the logistic regression without the variable RI, and the concordance of most likely models between the two runs, we have selected the 1st model from the run excluding the variable RI for further consideration. This model will be referred to as Model A in the ensuing discussion.

The selected model (Model A) was logit(p) = -4.7262 + 3.6621 EVS_DX(2) + 1.3323 EVS_DX(3) + 0.2021 VENOUS. The characteristics of Model A are detailed in *Table XVI.*

***Table XVI. Model A, logistic regression analysis***

| *Variable* | *Parameter estimate* | *S.E.* † | *OR* †† | *95%CI for OR* |
|---|---|---|---|---|
| EVS_DX(2) | 3.6621 | 0.5346 | 38.94 | (13.66, 111.04) |
| EVS_DX(3) | 1.3323 | 0.7408 | 3.79 | (0.89, 16.19) |
| VENOUS | 0.2021 | 0.0569 | 1.22 | (1.09, 1.37) |
| Intercept | -4.7262 | 0.5204 | — | — |

† Standard error, †† Odds ratio

Therefore in Model A, the adjusted odds ratio for EVS_DX(2) (malignant diagnosis with EVS) as a predictor for an actual malignant outcome was 38.94, for EVS_DX(3) (indeterminate diagnosis with EVS) the adjusted odds ratio was 3.79, and for VENOUS it was 1.22.

Since VASCUL was not included in the model, and the bivariate analysis suggested an interaction effect between EVS_DX and VASCUL, Model A was rerun with the addition of the variable VASCUL (Model B). Model B was logit(p) = -4.8852 + 3.3083 EVS_DX(2) + 1.1579 EVS_DX(3) + 0.1808 VENOUS + 0.3352 VASCUL. The characteristics of Model B are detailed in *Table XVII.* Note that the 95% CI for the odds ratio of VASCUL includes the null value.

***Table XVII. Model B, logistic regression analysis***

| *Variable* | *Parameter estimate* | *S.E.* † | *OR* †† | *95%CI for OR* |
|---|---|---|---|---|
| EVS_DX(2) | 3.3083 | 0.5834 | 27.34 | (8.71, 85.78) |
| EVS_DX(3) | 1.1579 | 0.7509 | 3.18 | (0.73, 13.87) |
| VENOUS | 0.1808 | 0.0626 | 1.20 | (1.06, 1.35) |
| VASCUL | 0.3352 | 0.3040 | 1.40 | (0.77, 2.54) |
| Intercept | -4.7262 | 0.5204 | — | — |

† Standard error, †† Odds ratio

Model C includes the interaction term and was logit(p) = -8.1702 + 7.4572 EVS_DX(2) + 5.1236 EVS_DX(3) + 0.2039 VENOUS + 2.3393 VASCUL – 2.5434

EVS_DX(2) * VASCUL – 2.6624 EVS_DX(3) * VASCUL. The characteristics of Model C are detailed in *Table XVIII.*

***Table XVIII. Model C, logistic regression analysis***

| *Variable* | *Parameter estimate* | *S.E.* † | *OR* †† | *95%CI for OR* |
|---|---|---|---|---|
| EVS_DX(2) | 7.4572 | 1.7468 | 1732.22 | (56, 53,152) |
| EVS_DX(3) | 5.1236 | 1.8159 | 167.93 | (4.79, 5900) |
| VENOUS | 0.2039 | 0.0661 | 1.22 | (1.08, 1.40) |
| VASCUL | 2.3393 | 0.7588 | | |
| EVS_DX(2)*VASCUL | -2.5434 | 0.8603 | | |
| EVS_DX(3)*VASCUL | -2.6624 | 1.0378 | | |
| Intercept | -8.1702 | 1.6257 | — | — |

† Standard error, †† Odds ratio

Since this was a prospective cohort study, we can compute the probability of a malignant outcome in an individual with different underlying risk factors. We can thereby investigate the interaction effect by entering the following covariate patterns, namely a low and high value for VASCUL for the benign and then the malignant EVS category respectively. A value of "2" will be entered for the variable VENOUS since this represents the median.

- EVS category benign, with VASCUL 0, VENOUS 2: 0,0,2,0,0,0
  Logit (p) = -7.7624, p(malignant) = 0.0004
- EVS category benign, with VASCUL 2, VENOUS 2: 0,0,2,2,0,0
  Logit (p) = - 3.0838, p(malignant) = 0.04
- EVS category malignant, with VASCUL 0, VENOUS 2: 1,0,2,0,0,0
  Logit (p) = -0.3052, p(malignant) = 0.42
- EVS category malignant, with VASCUL 2, VENOUS 2: 1,0,2,2,2,0
  Logit (p) = -0.7134, p(malignant) = 0.33

One can see from the above probabilities for disease outcome that the interaction effect is most marked for the benign category of EVS diagnosis

(EVS_DX). Therefore, the interaction term will be maintained in our final model (Model C).

### 4.4.9 *Goodness of fit*

To assess the internal validity of the models, the probability of endometrial carcinoma predicted by Models A and C for each covariate pattern, compared to that observed in the data was evaluated (see *Table XIX*). In this way the fit of the two models could be directly compared.

For covariate patterns with small or zero cell size, the fit cannot be detemined because the probability of observed malignancies becomes very unreliable. In general, Model C was slightly more accurate in predicting case status than Model A. Overall considering Model C, the observed and predicted probabilities were within 0.08 of each other, and often much closer, indicating adequate fit between the model and data.

***Table XIX-A. Comparison of observed vs. predicted p(malignant) – Model A***

| *Covariate Pattern* | *No. of patients with covariate pattern* | *p(malignant) Observed* | *p(malignant) Predicted* |
|---|---|---|---|
| 0,0,0 | 140 | 0.007 | 0.009 |
| 0,0,2, | 35 | 0 | 0.01 |
| 0,0,5 | 16 | 0 | 0.02 |
| 1,0,0 | 9 | 0.33 | 0.26 |
| 1,0,2 | 5 | 0.40 | 0.34 |

***Table XIX-B. Comparison of observed vs. predicted p(malignant) – Model C***

| *Covariate Pattern* | *No. of patients with covariate pattern* | *p(malignant) Observed* | *p(malignant) Predicted* |
|---|---|---|---|
| 0,0,0,0,0,0 | 111 | 0 | 0.0003 |
| 0,0,2,0,0,0 | 35 | 0 | 0.0004 |
| 0,0,5,0,0,0 | 16 | 0 | 0.0008 |
| 0,0,5,2,0,0 | 9 | 0 | 0.08 |
| 1,0,0,0,0,0 | 9 | 0.33 | 0.33 |
| 1,0,2,0,0,0 | 5 | 0.40 | 0.42 |
| 1,0,5,2,2,0 | 2 | 0.50 | 0.47 |

## 4.5 Receiver operating characteristic (ROC) curve analysis

Several of the more plausible continuous variables were further examined by receiver operating characteristic (ROC) curve analysis. The decision to investigate these variables was based on the results of the multivariate analysis, as well as substantive theory from the literature. For selected pairs of variables the areas under the ROC curves were compared. In addition, a sensitivity analysis was performed in an attempt to define an optimal threshold value for differentiating malignant from benign endometria.

### *4.5.1.Areas under the ROC curve for different variables*

For all variables, ROC curve analysis was performed for the total patient population and then stratified according to hormone use. *Table XX* details the areas under the ROC curve ($A_Z$) for the different variables investigated.

***Table XX. Areas under ROC curve ($A_Z$) for different variables***

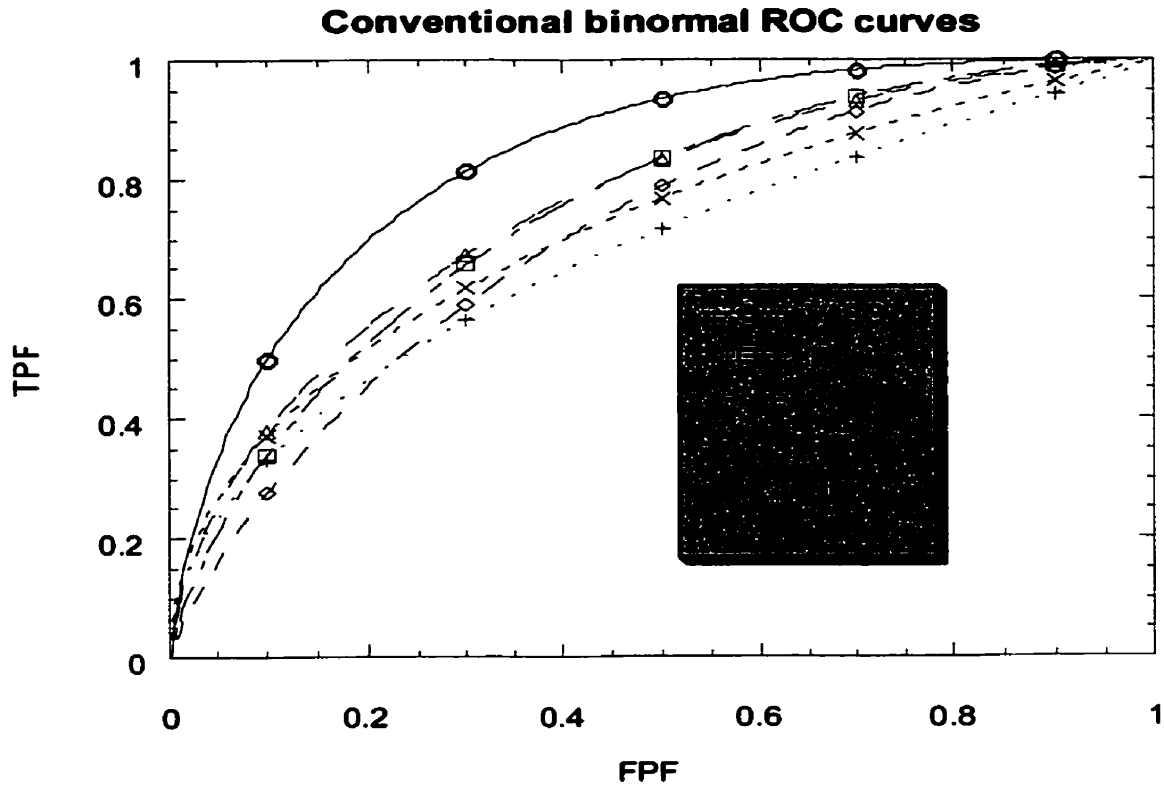
| | $A_Z \pm SE$ | | | |
|---|---|---|---|---|
| *Variable* | *All patients* | *Without hormones* | *With hormones* | *95% CI for $\Delta$ in $A_Z$*[††] |
| ENDO_THICK | 0.75 ± 0.04 | 0.78 ± 0.06 | 0.72 ± 0.07 | (-0.12, 0.24) |
| PSV | 0.71 ± 0.06 | 0.73 ± 0.08 | 0.71 ± 0.07 | (-0.19, 0.23) |
| PI | 0.71 ± 0.06 | 0.67 ± 0.09 | 0.69 ± 0.08 | (-0.25, 0.22) |
| RI | 0.67 ± 0.06 | 0.68 ± 0.09 | 0.70 ± 0.09 | (-0.27, 0.23) |
| VASCUL | 0.84 ± 0.03 | 0.82 ± 0.06 | 0.87 ± 0.04 | (-0.19, 0.09) |
| VENOUS | 0.75 ± 0.05 | 0.75 ± 0.07 | 0.75 ± 0.07 | (-0.19, 0.19) |

[††] In patients with and without hormones

As illustrated in the preceding table, our data do not present evidence for a difference in the areas under the ROC curve for patients with and without exogenous hormone use, as indicated by the 95% CI interval for the difference in $A_Z$ between the two patient populations.

For comparing areas under the ROC curve for selected pairs of variables in the total patient population (see *Figure 15*), we used the method by Hanley and McNeil (109) for correlated areas. We limited our comparisons to variable pairs with a potentially significant clinical impact. We found that the following pairs of variables were equally accurate in assigning case status : VASCUL/VENOUS (95% CI: -0.01, 0.19), VASCUL/ENDO_THICK (95% CI: 0.00, 0.18), PI/RI (95% CI: -0.04, 0.12), and ENDO_THICK/PSV (95% CI: -0.09, 0.17). Of the commonly used Doppler indices, endometrial vascularity was slightly more accurate in assigning case status than PSV [VASCUL/PSV (95% CI: 0.03, 0.23)] or RI [ VASCUL/RI (95% CI: 0.06, 0.28)].

*Figure 15. ROC curve for selected variables in the total patient population*




*4.5.2 Sensitivity analysis for selected variables*

In order to determine whether an optimal cut-off level exists to differentiate malignant from benign endometria for one or more of the above variables, a sensitivity analysis was performed. See *Tables XXI – XXIII* for sensitivity analysis of selected variables. Sensitivity analysis of the variables PSV, RI, and PI (not shown), revealed no clinically useful cut-off values for predicting malignancy.

***Table XXI. Sensitivity analysis for endometrial thickness (ENDO_THICK) Total number of patients = 421***

| *THICK* † | *TP* | *FP* | *FN* | *TN* | *SENS* | *SPEC* | *PPV* | *NPV* | *ACC* |
|---|---|---|---|---|---|---|---|---|---|
| 1 | 31 | 368 | 0 | 22 | 100% | 6% | 8% | 100% | 13% |
| (95%CI) | | | | | (91%-100%) | (4%-8%) | (5%-11%) | (87%-100%) | (10%-16%) |
| 2 | 31 | 296 | 0 | 94 | 100% | 24% | 9% | 100% | 29% |
| (95%CI) | | | | | (91%-100%) | (20%-29%) | (7%-13%) | (97%-100%) | (25%-34%) |
| 3 | 26 | 239 | 5 | 151 | 84% | 39% | 10% | 97% | 42% |
| (95%CI) | | | | | (66%-95%) | (34%-44%) | (6%-14%) | (93%-99%) | (37%-47%) |
| 4 | 24 | 197 | 7 | 193 | 77% | 49% | 11% | 97% | 52% |
| (95%CI) | | | | | (59%-90%) | (44%-55%) | (7%-16%) | (93%-99%) | (47%-56%) |
| 5 | 23 | 160 | 8 | 230 | 74% | 59% | 13% | 97% | 60% |
| (95%CI) | | | | | (55%-88%) | (54%-64%) | (8%-18%) | (93%-99%) | (56%-66%) |
| 6 | 23 | 127 | 8 | 263 | 74% | 67% | 15% | 97% | 67% |
| (95%CI) | | | | | (55%-88%) | (63%-72%) | (10%-22%) | (94%-98%) | (63%-72%) |
| 7 | 20 | 103 | 11 | 289 | 65% | 74% | 16% | 96% | 73% |
| (95%CI) | | | | | (45%-81%) | (69%-78%) | (10%-24%) | (93%-98%) | (69%-78%) |
| 8 | 18 | 82 | 13 | 308 | 58% | 79% | 18% | 96% | 76% |
| (95%CI) | | | | | (38%-75%) | (75%-83%) | (11%-27%) | (92%-97%) | (73%-81%) |
| 9 | 14 | 77 | 17 | 313 | 45% | 80% | 15% | 95% | 78% |
| (95%CI) | | | | | (27%-64%) | (76%-84%) | (9%-24%) | (93%-98%) | (73%-82%) |
| 10 | 13 | 65 | 18 | 325 | 42% | 83% | 17% | 95% | 80% |
| (95%CI) | | | | | (25%-61%) | (79%-87%) | (9%-27%) | (92%-97%) | (76%-84%) |

† A cut-off value of 2 indicates: patients with thickness > 2 mm = malignant, all else benign; a cut-off value of 3 indicates: patients with thickness > 3 mm = malignant, all else benign etc.

As is demonstrated in *Table XXI*, changing the threshold of endometrial thickness used to define endometrial malignancy resulted in the expected trade-off between sensitivity and specificity. Applying a cut-off value of 2 mm for endometrial thickness, resulted in a 100% sensitivity but only a 24% (95% CI: 20% - 29%) specificity for detecting endometrial carcinoma in our patient population. Using the more widely accepted cut-off value of 5 mm (83), increased the specificity to 59% (54% - 64%), however at the cost of a decrease in sensitivity from 100% to 74% (55% - 88%). Using a combination of biometric and morphologic criteria, EVS diagnosed malignancy with a sensitivity of 77% (59% - 90%) and a specificity of 84% (80% - 87%). Therefore the specificity of diagnosing

endometrial carcinoma was substantially higher when a combination of biometric and morphologic criteria were used, without a concomitant decrease in sensitivity.

Of the 31 patients with proven endometrial carcinoma, eight cases or 26% had an endometrial thickness of ≤ 5 mm. In five of these cases, the endometrial thickness measured only 3 mm. In seven of the eight false negative cases, a mass distending the endometrial cavity was present. As detailed in Section 3.3.2, the endometrial thickness in our study did not include measurements of an endometrial mass, in patients where the endometrium could be identified clearly as a separate structure. Had measurements of endometrial thickness included the mass, all eight cases would have been correctly classified as malignant using a threshold value of 5 mm. Real-time EVS diagnosed the eight cases as follows: malignant [n = 4], benign [n = 3], and indeterminate [n = 1].

***Table XXII. Sensitivity analysis for endometrial vascularity (VASCUL) Total number of patients = 407***

| *VASCUL* † | *TP* | *FP* | *FN* | *TN* | *SEN* | *SPEC* | *PPV* | *NPV* | *ACC* |
|---|---|---|---|---|---|---|---|---|---|
| 0 (95%CI) | 30 | 222 | 1 | 154 | 97% (83%-100%) | 41% (36%-46%) | 12% (8%-17%) | 99% (96%-100%) | 45% (40%-50%) |
| 1 (95%CI) | 20 | 71 | 11 | 305 | 65% (45%-81%) | 81% (77%-85%) | 22% (14%-32%) | 96% (94%-98%) | 80% (76%-84%) |
| 2 (95%CI) | 7 | 10 | 24 | 36 | 23% (10%-41%) | 97% (95%-99%) | 41% (18%-67%) | 94% (91%-96%) | 92% (89%-94%) |
| 3 (95%CI) | 0 | 0 | 31 | 376 | 0% (0%-10%) | 100% (98%-%100) | 0% (0%-1%) | 92% (89%-95%) | 92% (89%-95%) |

† A cut-off value of 0 mm indicates: patients with colour vascularity coded > 0 = malignant, all else benign, etc.

***Table XXIII. Sensitivity analysis for endometrial venous velocity (VENOUS) Total number of patients = 419***

| *VENOUS* † | *TP* | *FP* | *FN* | *TN* | *SENS* | *SPEC* | *PPV* | *NPV* | *ACC* |
|---|---|---|---|---|---|---|---|---|---|
| 0 | 26 | 228 | 4 | 161 | 87% | 41% | 10% | 98% | 45% |
| (95%CI) | | | | | (69%-96%) | (37%-46%) | (7%-15%) | (94%-99%) | (40%- 50%) |
| 1 | 26 | 193 | 4 | 196 | 87% | 50% | 12% | 98% | 53% |
| (95%CI) | | | | | (69%-96%) | (45%-56%) | (8%-17%) | (95%-99%) | (49%-58%) |
| 2 | 24 | 152 | 6 | 237 | 80% | 61% | 14% | 98% | 62% |
| (95%CI) | | | | | (61%-92%) | (56%-66%) | (9%-20%) | (95%-99%) | (57%-67%) |
| 3 | 21 | 110 | 9 | 279 | 70% | 72% | 16% | 97% | 72% |
| (95%CI) | | | | | (51%-85%) | (67%-76%) | (10%-23%) | (94%-99%) | (67%-76%) |
| 4 | 18 | 79 | 12 | 310 | 60% | 80% | 18% | 96% | 78% |
| (95%CI) | | | | | (41%-77%) | (75%-84%) | (11%-28%) | (94%-98%) | (74%-82%) |
| 5 | 12 | 54 | 18 | 335 | 40% | 86% | 18% | 95% | 83% |
| (95%CI) | | | | | (23%-59%) | (82%-89%) | (10%-30%) | (92%-97%) | (79%-86%) |
| 6 | 9 | 34 | 21 | 355 | 30% | 92% | 20% | 94% | 93% |
| (95%CI) | | | | | (15%-49%) | (88%-94%) | (10%-36%) | (92%-97%) | (83%-90%) |
| 7 | 7 | 22 | 23 | 367 | 23% | 94% | 24% | 94% | 89% |
| (95%CI) | | | | | (10%-42%) | (92%-96%) | (10%-44%) | (91%-96%) | (86%-92%) |
| 8 | 6 | 13 | 24 | 376 | 20% | 97% | 32% | 94% | 91% |
| (95%CI) | | | | | (8%-39%) | (94%-98%) | (13%-57%) | (91%-96%) | (88%-94%) |
| 9 | 6 | 8 | 24 | 381 | 20% | 98% | 43% | 94% | 92% |
| (95%CI) | | | | | (8%-39%) | (96%-99%) | (18%-71%) | (91%-96%) | (89%-95%) |

† A cut-off value of 1 indicates: patients with venous velocity > 1 cm/sec = malignant, all else benign, etc.

As is demonstrated in *Tables XXII - XXIII*, changing the threshold of endometrial vascularity (VASCUL) or venous flow (VENOUS) used to define endometrial malignancy resulted in the expected trade-off between sensitivity and specificity. Nevertheless, when comparing these results with those of endometrial thickness, one notes a substantially higher sensitivity for a given value of specificity and vice versa.

## 5 - DISCUSSION

We have prospectively studied a large consecutive population of postmenopausal women presenting with abnormal vaginal bleeding using EVS and Doppler ultrasound. The available body of literature suggests that EVS, using measurements of endometrial thickness, has a high sensitivity for detecting endometrial carcinoma. Most authors recommend using a low cut-off value such as 4 or 5 mm, which maintains a high sensitivity but sacrifices specificity. A meta-analysis of articles published between 1966 and 1996 (83) found that 96% (95% CI: 94%-98%) of women with cancer had an abnormal EVS, when a threshold value of > 5 mm was used to define abnormal endometrial thickening. The corresponding specificity for diagnosing endometrial carcinoma was 61% (95% CI: 59%-63%). The false-negative rate of 4% for EVS compares favourably with those achieved using office-based endometrial biopsy devices. These authors concluded that an endometrial thickness of ≤ 5 mm on EVS, excludes endometrial disease in the majority of postmenopausal women with vaginal bleeding.

In the above meta-analysis (83), endometrial thickness was defined as the width of the combined thickness of the anterior and posterior sides of the endometrium. The authors of this article, however, did not address whether polypoid masses distending the endometrial cavity were included in measurements of endometrial thickness. Measurements of endometrial thickness in our study population *excluded* the contents of a distended uterine cavity. We felt that including fluid or polypoid masses that distend the uterine cavity overestimates the thickness of the endometrium. Furthermore, no distinction is possible between conditions that result in localized or diffuse endometrial thickening, such as endometrial hyperplasia and carcinoma, versus endometrial polyps. In our patient population, using a threshold value of > 5 mm to define abnormal endometrial thickening, endometrial carcinoma was diagnosed with a sensitivity of 74% (95% CI: 55% - 88%) and a specificity of 59% (95% CI: 54% - 64%). If we include polypoid masses in measurements of endometrial thickness, endometrial carcinoma is diagnosed with a sensitivity of 97% (95% CI: 83% - 100%) and a specificity of 47% (95% CI: 42% – 52%). Of the 31 patients with proven endometrial carcinoma in our study population, eight cases or 26% had an endometrial thickness of ≤ 5 mm. In seven of the eight false negative cases, a mass distending the endometrial cavity was present at EVS.

At histopathology three of the seven masses were benign endometrial polyps. These findings underline the limitation of EVS in differentiating benign from malignant endometrial pathology. In our study, EVS differentiated pathologic from normal endometria with a sensitivity of 89% (95% CI: 83% - 94%) and a specificity of 60% (95% CI: 52% - 69%).

As is evident from our results, including polypoid masses in measurements of endometrial thickness substantially increases the sensitivity of EVS for detecting endometrial carcinoma from 74% (95% CI: 55% - 88%) to 97% (95% CI: 83% - 100%). These results are not unexpected, given the known association between benign polyps and endometrial carcinoma. In addition, although endometrial carcinoma typically presents as an infiltrative mass replacing the endometrium, endometrial carcinoma may present as a polypoid mass on EVS. When polypoid masses are included in measurements of endometrial thickness, the specificity decreases from 59% (95% CI: 54% - 64%) to 47% (95% CI: 42% – 52%) at the 5 mm cut-off level. However, the highest specificity is achieved with EVS when a combination of biometric and morphologic criteria are used to diagnose malignancy: sensitivity 77% (95% CI: 59% - 90%) and specificity 84% (95% CI: 80% - 87%). The decision to emphasize sensitivity versus specificity when evaluating the test performance of EVS, will depend largely on the clinical indication for performing the test. Since the role of EVS in evaluating patients with postmenopausal bleeding is primarily to identify patients who require further evaluation, an abnormal test result must have a high sensitivity for diagnosing endometrial carcinoma. Therefore, patients with findings on EVS indicating a low probability of endometrial disease will not be required to undergo routine endometrial sampling.

In our study population, EVS achieved only a modest sensitivity of 74% when an endometrial thickness of > 5 mm was used to indicate a positive test result. Similarly, using a combination of biometric and morphological sonographic criteria did not improve the sensitivity of diagnosing endometrial carcinoma, although the overall accuracy was substantially increased. These results are in keeping with the sensitivity of 85% and specificity of 78% reported by Hanggi et al. (23), when the following criteria for malignancy were applied: an endometrial thickness of greater than 5 mm, areas of decreased echogenicity or heterogeneity, and poor definition of the endo-myometrial junction. These authors concluded that dilatation and

curettage is necessary in the evaluation of women presenting with postmenopausal bleeding. Although a number of studies have published modest results in diagnosing endometrial carcinoma with EVS (57,83), most have found EVS to be highly sensitive (17,19,22,40,80,81,83,86,87). The reported sensitivity and specificity in these studies range from 94% - 100% and 24% - 68%, respectively. Although most authors agree on the role of EVS in the evaluation of patients with postmenopausal bleeding, they differ in their recommendations regarding optimal cut-off values for endometrial thickness. Nevertheless, there is a clear trend towards recommending cut-off values that yield a high sensitivity at the cost of specificity. Applying a threshold value of > 2 mm to define malignancy in our study population, resulted in a 100% sensitivity and a 24% (95% CI: 20% - 29%) specificity for detecting endometrial carcinoma. A threshold value of > 5 mm resulted in a sensitivity of 74% (95% CI: 55% - 88%) and a specificity of 59% (95% CI: 54% - 64%). The discrepancy between our results and those published in the literature at the 5 mm threshold level, may be explained, in part, by the different criteria used for measuring endometrial thickness. Although some authors clearly state that the contents of a distended uterine cavity were included in measurements of endometrial thickness, others are less precise regarding the method used to obtain endometrial thickness (5,17,19,80,83).

Our data do not present evidence for a difference in the area under the ROC curve for patients on hormonal replacement therapy. In addition, the 95% confidence intervals surrounding the sensitivity, specificity, PPV, NPV and accuracy of diagnosing endometrial carcinoma in patients with and without hormone use, demonstrate considerable overlap. Nevertheless, there appears to be a trend towards a lower sensitivity and PPV in patients receiving hormonal replacement therapy. Although the observed differences may simply represent random variation, the number of observations may have been too small to demonstrate a true difference. Endometrial thickness is known to increase after the initiation of hormone replacement therapy. The degree of increase in endometrial thickness, however, will vary depending on the type of hormonal regimen used (72). Using biometric criteria alone, EVS is unable to discriminate between proliferative endometrium, benign endometrial disease and carcinoma. Because hormone replacement therapy in postmenopausal women may result in proliferative changes of the endometrium,

EVS typically is less specific in this subgroup of patients. In particular, when endometrial thickening is the only criteria used to diagnose malignancy. In the meta-analysis of articles published between 1966 and 1996 (83), EVS was equally accurate at identifying women with endometrial disease, regardless of whether or not they were receiving hormonal replacement therapy. However, women on hormonal replacement therapy had a substantially higher false positive rate (specificity 77%, 95% CI: 75%-79%), compared to patients not taking hormones (specificity 92%, 95% CI: 90%-94%). For this reason, some authors have advocated a higher threshold value for endometrial thickness in postmenopausal women on hormonal replacement therapy compared to controls (8 mm versus 5 mm) (79, 84). The trend towards a decrease in sensitivity that we observed using real-time sonographic findings most probably reflects the sonologists application of more stringent criteria, given the expected proliferative changes of the endometrium in patients receiving hormone replacement therapy.

Using logistic regression analysis we found that none of the individual morphological parameters such as endometrial thickness, echogenicity, heterogeneity, presence of a mass, or definition of borders was a better predictor of endometrial carcinoma than the subjective impression of malignancy using a combination of sonographic criteria. Of the Doppler parameters examined, maximal venous velocity proved to be the most important predictor of case status.

Our results are in agreement with previous studies that reported Doppler indices of endometrial flow to be better predictors of malignant endometria than Doppler indices of uterine arterial flow (101,102). Although earlier studies reported high accuracy rates using RI or PI to differentiate malignant from benign endometria (56,57,101), these results have not been corroborated by our findings or those of other investigators (60,61,103,104). Of the endometrial Doppler indices investigated, the presence of vascularity using colour Doppler and the venous velocity had the highest accuracy for predicting case status. These findings are in keeping with the results of Sladkevicius et al. (60), who found that the best Doppler variable for differentiating between benign and malignant endometria was the presence of colour flow within the endometrium (sensitivity 88% [95% CI: 66-97%], specificity 81% [95% CI: 75-89%]).

Several limitations were encountered in our study. First, since only patients presenting with postmenopausal bleeding were included in our protocol, the results of this study may not be applicable to an asymptomatic population with a lower prevalence of disease. However, since up to 90% of patients with endometrial carcinoma present with abnormal vaginal bleeding, clinical efforts at detecting endometrial carcinoma are largely directed at symptomatic patients. Second, all EVS were performed by experienced radiologists specialized in this technique. The accuracy of EVS is highly operator-dependent, and the results of this study may not be reproducible in general clinical practice. A third limitation is that we used an imperfect standard of reference to assign case status. Although methods have been devised to take into account an imperfect standard of reference (110,111), we performed all data analysis with the assumption that the chosen standard of reference was 100% accurate. Of the 421 patients in our final study group, 261 or 62% were classified on the basis of endometrial sampling: D & C [n = 164, 39%], and endometrial biopsy [n = 97, 23%]. As discussed in Section 2.1, office-based endometrial biopsy devices are reported to have a false-negative rate ranging from 5% - 15%, while D & C has a false-negative rate ranging from 2% - 6%. Indeed, two patients with a final histological diagnosis of endometrial carcinoma had initial endometrial biopsies that were negative or inconclusive for malignancy. Although a negative endometrial biopsy was considered a study endpoint, several mechanisms were in place to minimize the possibility of false negative diagnoses. First, most patients with an underlying endometrial carcinoma continue to be symptomatic, and therefore undergo repeat EVS and/or more invasive sampling procedures. Second, the referring gynecologists were aware of the study protocol and would routinely contacts us about discrepancies between imaging findings and the clinical evaluation. Finally, although for study purposes the sonologists were asked to classify all endometria as benign or malignant, practically, all patients with endometrial thickening and/or an endometrial based mass were referred for histological sampling which resulted in a low false negative rate for EVS. Similar arguments can be made for the 135 patients (without endometrial histology) in whom a presumptive diagnosis of benign endometrium was made, since all patients remained asymptomatic with cessation of vaginal bleeding after a minimum follow-up of one year (mean 3 years).

In summary, this study supports the growing trend in practice to use EVS as an alternative to endometrial biopsy in evaluating patients with postmenopausal vaginal bleeding. We found that a thin measurement of endometrial thickness on EVS can exclude endometrial carcinoma in the majority of postmenopausal women with vaginal bleeding, regardless of hormonal replacement therapy. The recommended threshold value for endometrial thickness in our study is dependent on the method with which the endometrial stripe is measured. Using a combination of biometric and morphologic sonographic criteria achieved the best accuracy in diagnosing patients with endometrial carcinoma, however, at the cost of sensitivity. The presence of arterial or venous flow using Doppler can be used as an ancillary finding for predicting case status.

## 6 - REFERENCES


1. Nesse RE. Abnormal vaginal bleeding in perimenopausal women. Am Fam Physician 1989; 40:185-192.
2. Karlsson B, Granberg S, Hellberg P, Wikland M. Comparative study of transvaginal sonography and hysteroscopy for the detection of pathologic endometrial lesions in women with postmenopausal bleeding. J Ultrasound Med 1994; 13:757-762.
3. Requard CK, Wicks JD, Mettler FA. Ultrasonography in the staging of endometrial adenocarcinoma. Radiology 1981; 140:781-785.
4. Choyke PL, Thickman D, Kressel HY, Lynch JH, Jaffe MH, Clark LR, Zeman RK. Controversies in the radiologic diagnosis of pelvic malignancies. Radiol Clin North Am 1985; 23:531-549.
5. Granberg S, Wikland M, Karlsson B, Norstrom A, Friberg L. Endometrial thickness as measured by endovaginal ultrasonography for identifying endometrial abnormality. Am J Obstet Gynecol 1991; 164:47-52.
6. Nolan JF, Huen A: Prognosis in endometrial cancer. Gynecol Oncol 1976; 4:384-390.
7. Chen SS, Lee L: Retroperitoneal lymph node metastases in Stage I carcinoma of the endometrium: Correlation with risk factors. Gynecol Oncol 1983; 16:319-325.
8. Figge DC, Otto PM, Tamini HK, Greer BE: Treatment variables in the management of endometrial cancer. AM J Obstet Gynecol 1983; 146:495-500.
9. DiSaia PJ, Creasman WT, Boronow RC, Blessing JA: Risk factors and recurrent patterns in Stage I endometrial cancer. AM J Obstet Gynecol 1985; 151:1009-1015.
10. Word B, Gravlee LC, Wideman GL. The fallacy of simple uterine curettage. Obstet Gynecol 1925; 9:78
11. Stock RJ, Kanbour A. Prehysterectomy curettage. Obstet Gynecol 1975; 45:537-541.
12. Chambers JT, Chambers SK. Endometrial sampling: When? Where? Why? With what? Clinical Obstetrics and Gynecology 1992; 35:28-39.
13. Frishman G, Jacobs S. A randomized clinical trial comparing Pipelle and Tis-u-trap for endometrial biopsy (comments). Obstet Gynecol 1990; 76:315-316.
14. Van Den Bosch T, Vandendael A, Van Schoubroeck D, Wranz PAB, Lombard CJ. Combining vaginal ultrasonography and office endometrial smapling in the

diagnosis of endometrial disease in postmenopausal women. Obstet Gynecol 1995; 85(3):349-352.

15. Giusa-Chiferi MG, Goncalves WJ, Baracat EC. Transvaginal ultrasound, uterine biopsy and hysteroscopy for postmenopausal bleeding. Int J Gynecol Obsete 1996; 55(1):39-44.
16. Byrne AJ. Endocyte endometrial smears in the cytodiagnosis of endometrial carcinoma. Acta Cytologica 1990; 34(3):373-381.
17. Karlsson B, Granberg S, Wikland M, Ryd W, Norstrom A. Endovaginal scanning of the endometrium compared to cytology and histology in women with postmenopausal bleeding. Gynecologic Oncology 1993; 50:173-178.
18. Dijkhuizen PHLJ, Brolmann HAM, Potters AE, Bongers MY, Heintz APM. The accuracy of transvaginal ultrasonography in the diagnosis of endometrial abnormalities. Obstet Gynecol 1996; 87: 345-349.
19. Guner H, Tiras MB, Karabacak O, Sarikaya H, Erdem M, Yildirim M. Endometrial assessment by vaginal ultrasonography might reduce endometrial sampling in patients with postmenopausal bleeding: A prospective study. Aus NZ J Obstet Gynecol 1996; 36:175-178.
20. Kavak Z, Ceyhan N, Pekin S. Combination of vaginal ultrasonography and pipelle sampling in the diagnosis of endometrial disease. Aust NZ J Obstet Gynaecol 1996; 36:63-66.
21. Goldchmit R, Katz Z, Blickstein I, Caspi B, Dgani R. The accuracy of endometrial pipelle sampling with and without sonographic measurement of endometrial thickness. Obstet Gynecol 1993; 82:727-730.
22. Pertl B, Lahousen M, Pieber D, Heydarfadi HJ, Giuliani A. Stellenwert der Sonograhie bei der Früherkennung des Endometriumkarzinoms. Gynäkologisch-geburtshilfliche Rundschau 1996; 36:14-20.
23. Hanggi W, Brandenberger AW, Ammann M, Laely A, Dietz TU, Hermann U. Diagnosis of malignant tumours of the uterus by transvaginal sonography. Ultraschall in Med 1995; 16:2-7.
24. Fleischer AC, Kalemeris GC, Machin JE, Entman SS, James AE. Sonographic depiction of normal and abnormal endometrium with histopathologic correlation. J Ultrasound Med 1986; 5:445-452.

25. Johnson MA, Graham MF, Cooperberg PL. Abnormal endometrial echoes: sonographic spectrum of endometrial pathology. J Ultrasound Med 1982; 1:161-166.
26. Chambers CB, Unis JS. Ultrasonographic evidence of uterine malignancy in the post menopausal uterus. Am J Obstet Gynecol 1986; 154:1194-1199.
27. Nasri MN, Coast GJ. Correlation of ultrasound findings and endometrial histopathology in postmenopausal women. Br J Obstet Gynaecol 1989; 96:1333-1338.
28. Callen PW, DeMartini WJ, Filly RA. The central uterine cavity echo: a useful anatomic sign in the ultrasonographic evaluation of the female pelvis. Radiology 1979; 131:187-190.
29. Fleischer AC, Kalemeris GC, Entman SS. Sonographic depiction of the endometrium during normal cycles. Ultrasound in Med. & Biol. 1986; 12:271-277.
30. Osmers R, Volksen M, Schauer A. Vaginosonography for early detection of endometrial carcinoma? Lancet 1990; 335(8705):1569-1571.
31. Mendelson EB, Bohm-Velez M, Joseph N, Neiman HL. Endometrial abnormalities: evaluation with transvaginal sonography. AJR 1988; 150:139-142.
32. Rullo S, Piccioni MG, Framarino Dei Malatesta ML, Silvestrini J, Boni T, Marzetti L. Sonographic, hysteroscopic, histological correlation inthe early diagnosis of endometrial carcinoma. Eur J Gynaec Oncol 1991; 12:463-469.
33. Coleman BG, Arger PH, Grumbach K, Menard MK, Mintz MC, Allen KS, Arenson RL, Lamon KA. Transvaginal and transabdominal sonography: prospective comparison. Radiology 1988; 168:639-643.
34. Bourne T, Habmberger L, Hahlin M, Granberg S. Ultrasound in gynecology: endometrium (Review). Int J Gynaecol Obstet 1997; 56(2):115-127.
35. Ghirardini G, Montanari M, Gualerzic. Carcinoma and endometrial thickness (letter). Lancet 1990; 336(8728):1447.
36. Rudelstorfer R, Nanz S, Bernaschek G. Vaginosonography and its diagnostic value in patients with postmenopausal bleeding. Arch Gynecol Obstet 1990; 248:37-44.
37. Botsis D, Kassanos D, Pyrgiotis E, Zourlas PA. Vaginal sonography of the endometrium in postmenopausal women. Clin Exp Obst Gyn 1992; 19:189-192.

38. Smith P, Bakos O, Heimer G, Ulmstein U. Transvaginal ultrasound for identifying endometrial abnormality. Acta Obstet Gynecol Scand 1991; 70:591-594
39. Lerner JP, Timor-Tritsch IE, Monteagudo A. Use of transvaginal sonography in the evaluation of endometrial hyperplasia and carcinoma. Obstet Gynecol Survey 1996; 51:718-725.
40. Karlsson B, Granberg S, Wikland M, Ylostalo P, Torvid K, Marsal K, Valentin L. Transvaginal ultrasonography of the endometrium in women with postmenopausal bleeding - A nordic multicenter study. Am J Obstet Gynecol 1995; 172:1488-1494.
41. Weigel M, Friese K, Strittmatter HJ et al. Measuring the thickness — is that all we have to do for sonographic assessment of endometrium in postmenopausal women? Ultrasound Obstet Gynecol 1995; 6:97-102.
42. Hulka CA, Hall DA, McCarthy K, Simeone JF. Endometrial polyps,hyperplasia, and carcinoma in postmenopausal women: Differentiation with endovaginal sonography. Radiology 1994; 191:755-758.
43. Atri M, Nazarnia S, Aldis AE, Reinhold C, Bret PM, Kintzen G. Transvaginal US appearance of endometrial abnormalities. Radiographics 1994; 14:483-492.
44. Stovall TG, Solomon SK, Ling FW. Endometrial sampling prior to hysterectomy. Obstet Gynecol 1989; 73:405-409.
45. Taylor KJW, Ramos I, Carter D, Morse SS, Snower D, Fortune K. Correlation of Doppler US tumor signals with neovascular morphologic features. Radiology 1988; 166:57-62.
46. Goswamy RK, Steptoe PC. Doppler ultrasound studies of the uterine artery in spontaneous ovarian cycles. Human Reprod 1988; 3:721-726.
47. Scholtes MCW, Wladimiroff JW, van Rijen HJM, Hop WCJ. Uterine and ovarian flow velocity waveforms in the normal menstrual cycle: a transvaginal Doppler study. Fertil Steril 1989; 52:981-985.
48. Taylor KJW, Burns PN, Wells PNT, Conway DI, Hull MGR. Ultrasound Doppler flow studies of the ovarian and uterine arteries. Br J Obstet Gynaecol 1985; 92:240-246.
49. Salvatore CA. Arteriole alteration in normal and hyperplastic endometrium. Am J Obstet Gynecol 1968; 102:493-500.

50. Thompson RS, Trudinger BJ, Cook CM. Doppler ultrasound waveform indices: A/B ratio, pulsatility index and Pourcelot ratio. Br J Obstet Gynaecol 1988; 95:581-588.
51. Long MG, Boultbee JE, Hanson ME, Begent RHJ. Doppler time velocity waveform studies of the uterine artery and uterus. Br J Obstet Gynaecol 1989; 96:588-593.
52. Cosgrove DO, Bamber JC, Davey JB, McKinna JA, Sinnett HD. Color Doppler signals from breast tumors - work in progress. Radiology 1990; 176:175-180.
53. Schoenberger SG, Sutherland CM, Robinson AE. Breast neoplasms: Duplex sonographic imaging as an adjunct in diagnosis. Radiology 1988; 168:665-668.
54. Shimamoto K, Sakuma S, Ishigaki T, Makino N. Intratumoral blood flow: evaluation with colour Doppler echography. Radiology 1987; 165:683-685.
55. Long MG, Boultbee JE, Begent RHJ, Bagshawe KD. Doppler ultrasound of the uterine artery and uterus in invasive mole and choriocarcinoma. Br J Obstet Gynaecol 1987 p. 621.
56. Bourne TH, Campbell S, Whitehead MI, Royston P, Steer CV, Collins WP. Detection of endometrial cancer in postmenopausal women by transvaginal ultrasonography and colour flow imaging. Br Med J 1990; 301:369.
57. Bourne TH, Campbell S, Steer CV, Royston P, Whitehead MI, Collins WP. Detection of endometrial cancer by transvaginal ultrasonography with color flow imaging and blood flow analysis: a preliminary report. Gynecologic Oncology 1991; 40:253-259.
58. Hata T, Hata K, Senoh D, Makihara K, Aoki S, Takemiya O, Kitao M. Doppler ultrasound assessment of tumor vascularity in gynecologic disorders. J Ultrasound Med 1989; 8:309-314.
59. Kurjak A, Shalan H, Sosic A, Benic S, Zudenigo D, Kupesic S, Predanic M. Endometrial cancinoma in postmenopausal women: evaluation by transvaginal color doppler ultraonography. AMJ Obstet Gymecol 1993; 169:1597-1603.
60. Sladkevicius P, Valentin L, Marsal K. Endometrial thickness and doppler velocimetry of the uterine arteries as discriminators of endometrial status in women with postmenopausal bleeding: a comparative study. Am J. Obstet Gynecol 1994; 171:722-728.

61. Conoscenti G, Meir YJ, Fischer-Tamaro L, Maieron A, Natale R, D'ottavio G, Rustico M, Mandruzzato G. Endometrial assessment by transvaginal sonography and histologicla findings after D&C in women with postmenopausal bleeding. Ultrasound Obstet Gynecol 1995; 6:108-115.
62. Grimes DA. Diagnostic dilatation and curettage: a reappraisal. Am J Obstet Gynecol 1982; 142:1-6.
63. Koonings PP, Moyer DL, Grimes DA. A randomized clinical trial comparing Pipelle and Tis-U-Trap for endometrial biopsy. Obstet Gynecol 1990; 75:293-295.
64. Vuopala S. Diagnostic accuracy and clinical applicability of cytological and histological methods for investigating endometrial carcinoma. Acta Obstet Gynecol Scand Suppl 1977; 70:1-72.
65. Ferenczy A, Shore M, Guralnick M, Gelfand MM. The Kevorkian curette. An appraisal of its effectiveness in endometrial evaluation. Obstet Gynecol 1979; 54:262-267.
66. Lutz MH, Underwood PB Jr, Kreutner A, Mitchell KS. Vacuum aspiration: an efficient outpatient screening technic for endometrial disease. South Med J 1977; 70:393-395.
67. Reid PC, Brown VA, Fothergill DJ. Outpatient investigation of postmenopausal bleeding. Br J Obstet Gynaecol 1993; 100:498
68. Townsend DE, Fields G, McCausland A, et al. Diagnostic and operative hysteroscopy in the management of persistent postmenopausal bleeding. Obstet Gynecol 1993; 82:419-421.
69. Goldrath MH, Sherman AI. Office hysteroscopy and suction curettage: can we eliminate the hospital diagnostic dilatation and curettage? Am J Obstet Gynecol 1985; 152:220-229.
70. Brooks PG, Serden SP. Hysteroscopic findings after unsuccessful dilatation and curettage for abnormal uterine bleeding. Am J Obstet Gynecol 1988; 158:1354-1357.
71. Goldstein SR, Nachtigall M, Snyder JR, Nachitall L. Endometrial assessment by baginal ultrasonography before endometrial sampling in patients with post menopausal bleeding. Am J Obstet Gynecol 1990; 163(1)part1, 119-123.
72. Bonilla-Musoles F, Ballester MJ, Marti MC, Raga F, Osbome NG. Transvaginal color doppler assessment of endometrial status in normal postmenopausal

women: the effect of hormone replacement therapy. J Ultrasound Med 1995; 14:503-507.

73. Varner RE, Sparks JM, Cameron CD, Roberts LL, Soong SJ. Transvaginal sonography of endometrium in postmenopausal women. Obstet & Gynecol 1991; 78(2):195-199.
74. Pepper JM, Oyesanya OA, Dewart PH, Howell A, Seif AW. Indices of differential endometrial: myometrial growth may be used to improve the reliability of detecting endometrial neoplasia in women on tamoxifen. Ultrasound in Obstretrics and Gynecology 1996; 408-411.
75. Osmers R, Volksen M, Rath W, Kuhn W. Vaginosonographic detection of endometrial cancer in postmenopausal women. Int J Gynecol Obstet 1990; 32:35-37.
76. Nasri MN, Shepherd JH, Setchell ME, Lowe DG, Chard T. The role of vaginal scan in measurement of endometrial thickness in postmenopausal women. Br J Obstet Gynecol 1991; 98:470-475.
77. Nasri NM, Coast GJ. Correlation of ultrasound findings and endometrial histopathology in postmenopausal women. Br J Obstet Gynecol 1989; 96: 1333-1338.
78. Mortakis AE, Mavrelos K. Transvaginal ultrasonography and hysteroscopy in the diagnosis of endometrial abnormalities. J Am Assoc Gynecol Laparosc 1997; 4:449-452.
79. Lin MC, Gosink BB, Wolf SI, Feldesman MR, Stuenkel CA, Braly PS, Pretorius DH. Endometrial thickness after menopause: Effect of hormone replacement. Radiology 1991; 180:427-432.
80. Haller H, Matejcic N, Rukavina B, Krasevic M, Rupcic S, Mozetic D. Transvaginal sonography and hysteroscopy in women with postmenopausal bleeding. Int J Gynecol Obstet 1996; 54:155-159.
81. Malinova M, Pehlivanov B. Transvaginal sonography and progesterone challenge for identifying pathology in postmenopausal women. Int J Gynecol Obstet 1996; 52:49-53.
82. Kurjak A, Shalan H, Kupesic S, Kosuta D, Sosic A, Benic S, Ilijas M, Jukic S, Predanic M. An attempt to screen asymptomatic women for ovarian and

endometrial cancer with transvaginal color and pulsed doppler sonography. J Ultrasound Med 1994; 13:295-301.

83. Smith-Bindman R, Kerlikowske K, Feldstein VA, Subak L, Scheidler J, Segal M, Brand R, Grady D. Endovaginal ultrasound to exclude endometrial cancer and other endometrial abnormalities. JAMA 1998; 280 (17):1510-1517.
84. Doren M, Suselbeck B, Schneider HPG, Holzgreve W. Uterine perfusion and endometrial thickness in postmenopausal women on long-term continuous combined estrogen and progestogen replacement. Ultrasond Obstet. Gynecol 1997; 9:113-119.
85. Duda VF, Juhnke I, Kaevel K, Rode G. Inter- und intrasbservergenauigkeit der vaginosonographischen endometriumbiometrie. Ultraschall Klin Prax 1992; 7:104-106.
86. Brandner P, Gnirs J, Neis KJ, Hetterbach A, Schmidt W. Vaginosonography in the noninvasive evaluation of the endometrium of the postmenopausal uterus. Geburtsh u Frauenheik 1991; 51:734-740.
87. Emanuel MH, Verdel MJ, Wamsteker K, Lammes FB. A prospective comparison of transvaginal ultrasonography and diagnostic hysteroscopy in the evaluation of patients with abnormal uterine bleeding: Clinical implications. Am J Obstet Gynecol 1995; 172:547-552.
88. Schiller VL, Grant EG. Doppler ultrasonography of the pelvis. Radiologic Clinics of North America 1992; 30(4):735-742.
89. In: Echocardiography, 5th ed., editor H. Feigenbaum. Lea & Febiger, Malvern, Philadelphia, 1994; P.26.
90. Burns PN, Wells PNT. A Doppler Glossary, In: Clinical application of Doppler ultrasound, editors K.J.W. Taylor, P.N. Burms, P.N.T. Wells. Raven Press, New York, New York, 1988; p.355.
91. Gosling RG, King DH. Ultrasound angiology. In : Marcus AW, Adanson L, eds. Arteries and veins. Edinburgh: Churchill livingstone; 1975:61-98.
92. Pourcelot L. Applications cliniques de l'examen Doppler transcutane. In: Peronneau P, ed. Velocimetre Ultrasonore Doppler, 34. Paris: Seminaire I.N.S.E.R.M. 1974:213-240.
93. Stuart B, Drumm J, Fitzgerald DE, Duignan NM. Fetal blood flow waveforms in normal pregnancy. Br J Obstet Gynaecol 1980; 87:780-785.

94. Achiron R, Lipitz S, Frenkel Y Mashiach S. Endometrial blood flow response to estrogen replacement therapy and tamoxifen in asymptomatic, postmenopausal women: a transvaginal Doppler study. Ultrasond Obstet Gynecol 1995; 5:411-414.
95. Achiron R, Lipitz S, Sivan E, Goldenberg M, Horovitz A, Frenkel Y, Mashiach S. Changes mimicking endometrial neoplasia in postmenopausa, tamoxifen-treated women with breast cancer: a transvaginal doppler study. Ultrasound Obstet Gynecol 1995; 6:116-120.
96. Bonila-Musoles F, Marti MC, Ballester MJ, Raga F, Osborne NG. Normal uterine arterial blood flow in postmenopausal women assessed by transvaginal color doppler sonography. J Ultrasound Med 1995; 14:497-501.
97. Bonila-Musoles F, Marti MC, Ballester MJ, , Raga F, Osborne NG. Normal uterine arterial blood flow in postmenopausal women assessed by transvaginal color doppler ultrasonography. J Ultrasound Med 1995; 14:491-494.
98. Mostbeck GH, Goessinger HD, Mallek R, Siostrzonek P, Schneider B, Tscholakoff D. Effect of heart rate on Doppler measurements of resistive index in renal arteries. Radiology 1990; 175:511-513.
99. Cacciatore B, Lehtovirta P, Wahlstroem T, Yloestalo P. Preoperative sonographic evaluation of endometrial cancer. Am J Obstet Gynecol 1989; 160:133-137.
100. Kurjak A, Zalud I, Jurkovic D, Alfirovic Z, Miljan M. Transvaginal color Doppler for the assessment of pelvic circulation. Acta Obstet Gynecol Scand 1989; 68:131-135.
101. Merce LT, Garcia GL, De la Fuente F. Doppler ultrasound assessement of endometrial pathology. Acta Ostet Gynecol Scand 1991; 70:525-530.
102. Aleem F, Predanic M, Calame R, Noukhtar M, Pennisi J. Transvaginal color and pulsed doppler sonography of the endometrium: a possible role in reducing the number of dilatation and curettage procedures. J Ultrasound Med 1995; 14:139-145.
103. Flam F, Almstrom H, Hellstrom AC, Moberger B. Value of uterine artery doppler in endometrial cancer. Acta Oncologica 1995; 34(6):779-782.

104. Chan FY, Chau MT, Pun TC, Lam C, Ngan HYS, Leong L, Wong RLC. Limitation of transvaginal sonography and color doppler imaging in the differentiation of endometrial carcinoma from benign lesions. J Ultrasound Med 1994; 13:623-628.
105. White CD. Postmenopausal bleeding as a risk factor for endometrial carcinoma. W V Med J 1991; 87(1):15-17.
106. Timor-Tritsch IE, Bar-Yam Y, Elgali S, Rottem S. The technique of transvaginal sonography with the use of a 6.5 MHz probe. Am J Obstet Gynecol 1988; 158:1019-1024.
107. Kleinbaum DG, Kupper LL, Muller KE. Applied regression analsysis and other multivariate methods. Belmont, California: Duxbury Press, 1988.
108. Kass RE, Raftery AE. Bayes Factors. J Am Stat Assoc 1995; 90:773-795.
109. Hanley JA, McNeil BJ. A method of comparing the areas under receiver operating characteristic curves derived from the same cases. Radiology 1983; 148:839-843.
110. Walter S and Irwig L. Estimation of test error rates, disease prevalence and relative risk from misclassified data: A review. J Clin Epi 1988; 41:923-937.
111. Joseph L, Gyorlcos T, Coupul L. Bayesian estimation of disease prevalence and the parameters of diagnostic tests in the absence of a Gold Standard. A J Epi 1995; 141:263-272.