Three-dimensional scan of the uterine cavity of infertile women before assisted reproductive technology use

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Abstract

The primary objective was to assess the utility of routine 3-dimensional (3D) ultrasound in the evaluation of infertile women and to estimate the prevalence of uterine anomalies before the use of assisted reproductive technology (ART), using the European Society of Human Reproduction and Embryology and the European Society for Gynaecological Endoscopy classification system. A second objective was to assess the effect of uterine anomalies on the pregnancy rate in patients who underwent assisted reproductive techniques.

We retrospectively studied 668 patients treated in the Department Obstetrics Gynecology and Neonatology “St Ioan” Clinical Emergency Hospital and in the Department of Medical and Surgical Specialties, Faculty of Medicine “Transilvania” University of Brasov between July 2016 and February 2017 for subfertility. Patients were examined using 2-dimensional (2D) and 3-dimensional (3D) transvaginal ultrasound. Müllerian duct anomalies were present in 6.13% of patients, with the most common anomaly being a dysmorphic uterus (class U1c in 42.68% of patients), 17 patients (20.73%) with incompletely septeate uterus (class U2a), 12 patients (14.63%) with a completely septeate uterus (class U2b), 8 patients (9.75%) with a party bicornpealre uterus (class U3a), and 6 patients (7.31%) with a completely bicornpealre uterus (class U3b). Only 1 (1.21%) patient had an aplastic uterus without a rudimentary cavity (class U5b). The pregnancy rate in the presence of uterine anomalies was 55% and the pregnancy rate in control group patients was 39.8%. The incidence of pregnancy in the group with uterine anomalies was statistically similar with the control group of normal uterus (P<0.1). For ongoing pregnancy rate and live birth rate, our data indicated a slightly elevated rate for both of those indexes in the anomalies group. The incidence of miscarriage in the presence of uterine anomalies was 24% and 6.7% in the control group, which is statistically significant (P=0.05).

3D ultrasound evaluation of the uterus should be considered before ART in order to make an accurate diagnosis of the uterine congenital anomaly and improve ART results.

Abbreviations: 2D = 2-dimensional, 3D-US = 3-dimensional ultrasound, AFS = American Fertility Society, ART = assisted reproductive technologies, ESGE = European Society of Gynaecological Endoscopy, ESHRE = European Society of Human Reproduction and Embryology, hCG = human chorionic gonadotropin, IUD = intrauterine device, MRI = magnetic resonance imaging, r-FSH = recombinant follicle stimulating hormone, ROI = region of interest.

Keywords: 3D-ultrasound, ART, infertility, transvaginal ultrasound, uterine malformations

1. Introduction

The impact of uterine malformations on reproductive outcomes is controversial and depends on the type of the malformation and the nature of the study that quantifies them.[1] Subfertility appears to be more frequent than absolute infertility in which conception is impossible. The Müllerian ducts are the embryological structures that eventually develop into the fallopian tubes, uterus, uterine cervix, and superior part of the vagina. A carefully regulated cascade of events regulates their development and several processes can interfere during intrauterine life and result in congenital uterine malformations. Previous studies have documented a 4% to 7% incidence of uterine malformations, which is often associated with urinary tract anomalies such as renal agenesis, cross fused renal ectopia, and duplex kidney.[1,2]

Two main classifications of Mullerian anomalies are used: the American Fertility Society (AFS), which was published in 1988, and the more recent the European Society of Human Reproduction and Embryology and the European Society of Gynaecological Endoscopy (ESHRE/ESGE) classification system, which is simplified and more logically based on a patient’s anatomy (Table 1). In the later system, Müllerian anomalies are classified into 6 main classes: U0 normal uterus; U1 dysmorphic uterus; U2 septate
Before using assisted reproductive technologies (ARTs), it is essential to correctly evaluate the uterine cavity in order to avoid poor pregnancy outcomes of pregnancies or ART failure.

In the last decade, several methods have been used to evaluate uterine anomalies, including 2-dimensional (2D) transvaginal ultrasound. Meanwhile, hysteroscopy and laparoscopy as well as magnetic resonance imaging (MRI) have been considered the gold standards in the diagnosis of uterine anomalies. Ultimately, 3-dimensional ultrasound (3D-US) and MRI are the most reliable for examining uterine morphology, as determined by the ESHRE/ESGE classification system on reproductive function. Before using assisted reproductive technologies (ARTs), it is essential to correctly evaluate the uterine cavity in order to avoid poor pregnancy outcomes of pregnancies or ART failure.

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Recent studies have compared the sensitivity of MRI and 3D-US and found a sensitivity of 100% in establishing a diagnosis of uterine anomalies. MRI and ultrasound are the most reliable for diagnosing uterine anomalies. In this way, achieving a differential diagnosis between a septate and bicornuate uterus is facilitated by the MRI scan is too expensive. In comparison with laparoscopy and vaginal anomalies being classified separately into subclasses. Previous studies have been based on the AFS classification system; however to the best of our knowledge, no studies have reported the impact of uterine morphology, as determined by the ESHRE/ESGE classification system on reproductive function.

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## Table 1

| Uterine anomaly class | Subclass | Description |
|-----------------------|----------|-------------|
| D0                    | (a) T-shaped | Dysmorphic uterus |
|                       | (b) Infants | Septate uterus |
|                       | (c) Others | Internal indentation >50% of the uterine wall thickness and external contour straight or with indentation <50% |
| U2                    | (a) Partial | Bicornual uterus |
|                       | (b) Complete | External indentation >50% of the uterine wall thickness, |
| U3                    | (a) Partial | Congenital uterus with cavity |
|                       | (b) Complete | Congenital uterus without cavity |
| U4                    | (a) With rudimentary cavity | Unilaterally formed uterus/hemi uterus |
|                       | (b) without rudimentary cavity | Rudimentary horn with cavity communicating or not |
| U5                    | (a) With rudimentary cavity | Rudimentary horn without cavity/aplasia (no horn) |
|                       | (b) without rudimentary cavity | Aplastic uterus |
| U6                    |          | Unclassified cases |

ESHRE = European Society of Human Reproduction and Embryology, ESHRE = European Society of Gynecological Endoscopy.

## 2. Methods

### 2.1. Study design

We conducted a retrospective study from July 2016 to January 2017 of all patients treated in 2 tertiary units, in the Department Obstetrics Gynecology “SfIoan” Clinical Emergency Hospital and in the Department of Medical and Surgical Specialties, Faculty of Medicine “Transilvania” University of Brasov, for infertility or subfertility complaints. The primary objective of our study was to assess the utility of 3D as routine evaluation for infertile women before ART in order to exclude uterine anomalies and to avoid unnecessary intervention such as MRI, laparoscopy, or hysteroscopy. In total, there were 668 patients with primary or secondary infertility. In the evaluation protocol, we included routine 3D transvaginal ultrasound in addition to conventional examination technique by 2D ultrasound. We additionally excluded patients with associated uterine pathology associated, one or more polyps, synechiae, or submucosa myoma. Also, we excluded patients in whom the ultrasound image was not sufficient for a definitive diagnosis. As the conventional 2D ultrasound cannot identify the uterine anomaly and the MRI scan is too expensive (5–7 times more than 3D ultrasound), our study had no comparative arm with one of those methods. Before the examination, we obtained written informed consent for transvaginal ultrasound examination. As the study was retrospective, ethical approval was not necessary.

### 2.2. Patient data collection

A Voluson E6 Ultrasound (General Electric Medical System Zipf, Austria) machine, equipped with a RIC5-9H 5 to 9 MHz 4D endocavitary probe, was used to acquire 2D and 3D images during the early follicular phase of menstrual cycle. Scanning was performed by 4 certified obstetrics and gynecology specialists in accordance with the following image acquisition protocol:

1. Examinations were performed during the early follicular phase, in order to facilitate the assessment of the uterine cavity, endometrial thickness, and cervical appearance, as well as in the early luteal phase for any signs of ovulation (i.e., presence of a corpus luteum, fluid in the Douglas pouch).
2. Patients were placed in the lithotomy position, after bladder evacuation.
3. A condom-covered probe was used after filling the tip of the condom with ultrasound gel.
4. A 3-step examination protocol was followed:
   a. First, 2D examination of the pelvis was performed to exclude the possibility of gross pathology such as uterine myoma or ovarian masses. Then, a mid-longitudinal (sagittal) section of the uterus in 2D was obtained. The region of interest (ROI) was adjusted in order to obtain the optimal 3D volume. The angle sweep was 90°. One to three static uterine volumes were obtained.
   b. Then, to display the 3 orthogonal planes, the longitudinal, transverse, and coronal planes were assigned to Boxes A, B, and C, respectively. The rendered image was obtained by adjusting the rendering box containing the ROI in Windows A to include the uterine fundus. The automated mode was used for acquisition. The multplanar display examined must have the whole uterus captured. The 3D dataset was
initially analyzed in the multiplanar view with longitudinal, transverse, and coronal sections. The endometrium must be evident in all 3 planes and also the upper endometrial cavity and the isthmic portions of each tube.

(c) Finally, the contrast and gain were adjusted and the uterine morphology was analyzed in different modes (rendered, TUI-tomographic ultrasound image, OmniView). OmniView (GE Medical Systems, Zipf, Austria) is a new display technology for 3D ultrasound that allows interrogation of volumes datasets.

The most important anatomical features to visualize for the characterization of the uterine anatomy include the uterine fundus, cavity borders, fallopian interstitial portions, and uterine wall thickness. These features are required for use of the ESHRE classification system. All images were recorded and analyzed both in real time and after examination. The average time for 3D image acquisition and analysis ranges between 2 and 4 minutes.

We also examined the association between uterine anomalies and pregnancy. We used the control group consisting of the patients without uterine anomalies. The ART procedure used was with different strategies, because it does not take place in the same clinic, using either short protocol or long protocol with recombinant follicle-stimulating hormone (r-FSH, Gonal-F; Merck Serono Biotechnology Pharmaceuticals, Darmstadt, Germany) or purified human menopausal gonadotropin (Menopur; Ferring Pharmaceuticals, Saint Prex, Switzerland). The starting dose was 150UI on day 3 of the follicular phase of the menstrual cycle. The ovarian response was monitored using ultrasound on alternate days. Serum estradiol measurements were done on alternative days starting with day 3. Human chorionic gonadotropin (Ovitrelle, Merck Serono Biotechnology Pharmaceuticals, Darmstadt, Germany; 0.5 mL per 6500 UI) was administered for inducing ovulation, when there were 3 or more follicles measuring 18 mm or more in diameter. Fertilization was achieved using standard in vitro fertilization technique, with transvaginal oocyte retrieval performed 36 to 48 hours after Ovitrelle injection. Sixteen days after transferring the embryo, o beta hCG levels were determined, and if the level was 50 UI/L, an ultrasound was done 2 weeks later to confirm the pregnancy. The patients were followed on the first trimester up to 12 weeks gestation.

2.3. Statistical analysis

Data analyzed are reported as frequencies as well as percentages after been analyzed using the IBM SPSS Statistics for Windows Version 23.0 (IBM Corp, Armonk, NY) and EpiInfo 3.5.4 (Statistics Program for Public Health Professionals for use on Windows 2000, CDC; Atlanta, Georgia). Fisher exact test or the Chi-squared test was used to compare pregnancy rates and miscarriage rates in women with uterine anomalies and compared those rates in woman with a normal uterus.

3. Results

Of the 668 patients, 82 (12.27%) were diagnosed with Müllerian duct anomalies. Subjects were between 23 and 44 years of age (with a median age 34.5 years) and only 1 had a history that
suggested Mullerian duct anomalies. A normal uterus was identified in 586 patients (Fig. 1). From these subjects, we selected a control group of 148 patients who underwent ART, with normal uterus and the same characteristics as the group with uterus anomalies, which have ART. This was the control group.

The pregnancy rate in the presence of uterine anomalies was 55% and the pregnancy rate in control group patients was 39.8%, so they were statistically similar ($P = .09$). The incidence of pregnancy in the group with uterine anomalies was statistically similar to that of the control group of normal uteri ($P < .11$). The incidence of miscarriage in the presence of uterine anomalies was 24% and in the control group was 6.7%, which is statistically significant ($P = .04$).

Surprisingly, 4 women with irregular menstrual bleeding and a history of infertility were pregnant at the time of examination. Among these women, 2 were classified with an U2a uterus (partial septate) and 2 were classified with an U2b (septate) (Fig. 2A, B). In this latter group, one patient’s pregnancy was at 32 weeks; however, she had been experiencing metrorrhagia throughout the pregnancy. This complication was particularly notable during the first trimester.

Some patients did not require or undergo ART in our unit and could not undergo follow-up properly. Therefore, we included only those patients who were treated in our unit. In the group of patients that exhibited pregnancy, data concerning the ART protocols or embryo quality were not available for all the patients and we could not use those variables to determine the relationship between fertilization success and therapy. Those patients who presented with an ovular sac in the uterine cavity during the first trimester of pregnancy were considered as having succeeded at achieving conception, while those who were pregnant but were unable to carry the embryo anytime during the first 12 weeks were considered to have miscarried.

Following ultrasonography evaluation, we diagnosed 2 (2.43%) patients with a dysmorphic uterus (class U1a), 1 patient (1.21%) with a hypoplastic uterus (class U1b), 35 patients (42.68%) with a generally dysmorphic uterus (class U1c), 17 patients (20.73%) with an incompletely septate uterus (class U2a), 12 patients (14.63%) with a completely septate uterus (class U2b), 8 patients (9.75%) with a partly bicornoreal uterus (class U3a), 6 patients (7.31%) with a completely bicornoreal uterus (class U3b), and 1 patient (1.21%) with an aplastic uterus without a rudimentary cavity (class U5b) (Table 2). Representative images of U1, U2, U3, and U4 classes are shown in Figs. 3–7, respectively.

The pregnancy rates were significantly higher for women with U1c class uterus (77.7%). For the U2 group, the rates were comparable (52% for U2a and 40% for U2b). We were unable to account for the influence of other factors that could affect fertility in the control group (e.g., follicular antral count, anti-Mullerian hormone levels, and endometriosis).

Miscarriage rates were significantly lower in the control group (6.7%) than those women included in this study. In particular, the highest miscarriage rate was for women with a class U2b uterus (30%), while women with classes U2a and U1c uteruses exhibited similar rates (22.2% and 21.4%, respectively) (Table 2). The overall miscarriage rate for woman with

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**Table 2**

| Uterine type | No. of patients with infertility (%) | ART submitted (No./%) | Conceiving after ART (No./%) | Miscarriage (No./%) |
|-------------|--------------------------------------|-----------------------|-----------------------------|-------------------|
| Normal uterus U0 | 586 | 148 (25.25%) | 59 (39.8%) | 4 (6.77%) |
| U1a | 2 (2.4%) | 1 (1.9%) | 0 | 0 |
| U1b | 1 (1.2%) | 0 | 0 | 0 |
| U1c | 35 (42.6%) | 18 (34.6%) | 14 (77.7%) | 3 (21.4%) |
| U2a | 17 (20.7%) | 17 (32.6%) | 9 (52%) | 2 (22.2%) |
| U2b | 12 (14.6%) | 10 (19.2%) | 4 (40%) | 2 (50%) |
| U3a | 8 (9.7%) | After surgery | 2 (33%) | 0 |
| U3b | 6 (7.3%) | 0 | 0 | 0 |
| U5b | 1 (1.2%) | 0 | 0 | 0 |
| Anomalies | 82 (13.9%) | 52 (63%) | 29 (55%) | 7 (24%) |

**ART** = assisted reproductive technologies.
uterine anomalies who underwent ART was 24%, which is considerably higher than the rate in the control group, which was 6.7%. Our data indicated that the ongoing pregnancy rate and live birth rate were slightly elevated rate for both of those indexes in the anomalies group without any identifiable explanation after analyzing other confounding variables implication (i.e., maternal age, body mass index, or smoking) as is illustrated in Table 3.

4. Discussion

Our findings suggest that uterine anomalies per se can result in infertility, which is supported by previous studies that have found that uterine congenital anomalies can affect fertility. This incidence appears to be higher than that reported in other studies; however, our findings exhibit a strong correlation with the rate of infertility reported elsewhere. This study demonstrates that...
results in a more accurate determination of the shape, dimensions, morphology, and wall thickness of the uterine cavity, which is comparable to the data obtained by MRI.\textsuperscript{13} Meanwhile, surgically invasive methods such as hysteroscopy have been shown to achieve a diagnostic sensitivity of 99.1%.\textsuperscript{14} Furthermore, 3D-US is efficient for diagnosing the possible abnormal placement of an IUD (intrauterine device) in a malformed uterus.\textsuperscript{16}

The most common anomaly that was reported in our data was the type U1c (former arcuate uterus), which occurred in 42.6% of patients. This is a minor anomaly that can be often improperly missed during hysteroscopy and laparoscopy. Meanwhile, the existence of uterine anomalies was observed more frequently than in other studies\textsuperscript{12} as we found that such a cluster of malformations occurred in nearly 20% of all uterine anomalies.

In comparing the rates of ART conception, we found that the overall rate of conception was higher (53%) in the treated group than in the normal uterine control group (39.8%), thereby indicating that an individual’s ability to achieve conception is affected by the uterine congenital anomalies.

Assessing the uterine morphology is important for reducing the miscarriage rate. Indeed, several patients could benefit from surgery before pregnancy whether it results in spontaneous or ART-mediated conception. Women with class U2 uterine malformations have a reduced level of fertility and higher rates of miscarriage and preterm delivery.\textsuperscript{13} This was confirmed by our study in which the miscarriage rate in women with class U2b uteri was 50%. Moreover, the overall miscarriage rate after ART was higher in women with anomalies group (24%) than in women with healthy uteri (6.7%), although the sample size in study was reduced. There was no significant difference in the rate of miscarriage between women with minor uterine anomalies such as classes U1c and U2a. However, assessing the impact of uterine anomalies on pregnancy outcomes would be more informative if performed using data from the second half of pregnancy.

Our study is based on the more recently developed classification system for uterine anomalies, which differs from previous studies\textsuperscript{12,14,17} that are based on the former AFS classification system. We adopted the ESHRE classification system for Müllerian duct congenital anomalies because it is based on the anatomy of the uterus. In the AFS classification system, some classes were difficult to assign (i.e., “didelphys uterus” or “bicornuate uterus”) or were simply confusing (i.e., “arcuate uterus”) given the difficulty in separating the uterus with partial septa.\textsuperscript{17,18} In addition, the ESHRE class U1c provides a classification for minor or subtle deformities of the uterine cavity that does not result in confusion with a clearly septate uterus.

Interestingly, patients with classes U3 or U4 uterine defects do not exhibit a reduced rate of fertility, but instead present with miscarriage or preterm delivery.\textsuperscript{13} The patients included in class U1a often present second-trimester miscarriage.\textsuperscript{13} Some of the

| Table 3 | Ongoing pregnancy and live births. | Births and number of cycles of treatment |
|---------|-----------------------------------|-----------------------------------------|
| Categories | Cycles and (P) | Pregnancies | Rate (%) |
| Control group (normal uterus) | Ongoing pregnancies | 312 (P=.376) | 59 | 18.91% |
| | Live births | 312 (P=.246) | 55 | 17.62% |
| Study group (uterine anomalies) | Ongoing pregnancies | 114 (P=.049) | 29 | 25.43% |
| | Live births | 114 (P=.067) | 22 | 19.29% |

Data are rate (%). No. are numbers of cycles, or pregnancies.
* Fischer exact test.
anomalies can be surgically resolved and improved pregnancy outcomes. The ESHRE-ESGE consensus generates some objective parameters that allow a precise categorization of uterine anomalies into 1 of 6 classes (U0-U5) and also a seventh class (U6) for unclassified cases. Several studies have reported improved reproductive outcomes following surgical interventions, but there are insufficient data regarding the safety and efficacy of such procedures when performed for minor anomalies. The ability to differentiate between anomalies is crucial for planning surgery and counseling. Furthermore, hysteroscopic septa resection can be useful for subfertile patients before ART, but further randomized controlled trials are needed to offer evidence-based management options.

Our study is limited by the lack of data concerning those patients with uterine anomalies and who did not undergo ART as well as by the reduced number of patients with ART. The short time of pregnancy follow-up was also a limit in evaluating the impact of uterine anomalies on pregnancy outcomes. Comparing to MRI or laparoscopy and hysteroscopy, 3D ultrasound is safer, cheaper, and more acceptable but needs a trained operator. The cost of 3D scan ranged between 40 and 50 USD compared with MRI scan of the pelvis range between 250 and 300 USD. Another advantage of the method is the possibility to be performed at the first ultrasound examination for infertility evaluation instead or in addition with the 2D conventional scan that cannot identify uterine congenital anomalies. A strength of this study is its inclusion of a large number of examined patients who were not pre-selected, which allowed us to confirm that there may be higher rates of uterine anomalies than previously reported in our population. Such patients could benefit from diagnosis before spontaneous or assisted conception.

5. Conclusion

Three-dimensional ultrasound evaluation of the uterus is useful before ART given the association between uterine congenital anomalies and subfertility. In order to ensure an accurate anatomical diagnosis, the ESHRE/ESGE classification system should be adopted. The ability of an individual to conceive is not prevented merely by the presence of a uterine anomaly, so uterine congenital anomalies do not affect the ART conception rate but are instead involved in first trimester pregnancy loss. For this reason, the accurate differentiation between uterine anomaly types is crucial in planning surgery and improving ART results.

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