Assessment of tubal patency through sono-hysterosalpingography using B-mode and colour Doppler: a comparative study

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ABSTRACT

Background: Tubal factors constitute a major proportion of infertility causes, and include disorders affecting one or more components of the fallopian tube. Demonstration of tubal patency using imaging modalities can help lower the rate of invasive diagnosis and management. Hysterosalpingography provides optimal delineation of fallopian tube anatomy, allowing detection of tubal patency, tubal occlusion, and peri-tubal disease. Sonosalpingography with saline infusion was started as an alternative because of its added benefits of structural and morphological abnormality detection.

Methods: Patients referred for primary and secondary infertility were assessed on B-mode ultrasound trans-abdominally, followed by cervical catheterization and trans-vaginal ultrasound. Sterile saline fluid was pushed through the cervix, to use as a contrast medium, and its movement was assessed on B-mode and as a colour Doppler artefact.

Results: B-mode, colour Doppler and presence of free-fluid in pouch of Douglas showed presence of tubal patency in 59.6%, 82.7% and 78.8% cases respectively and presence of bilateral and unilateral tubal block in 60.4%, 17.3% and 21.2% respectively. Diagnostic accuracy of B-mode and colour Doppler is 84.6% and 95.2% respectively.

Conclusions: Sonosalpingography can be used to pinpoint the exact location of the tubal block, due to its real-time imaging benefits. Normal saline infusion can be used effectively. Colour Doppler provides higher sensitivity for evaluation of tubal patency and has higher specificity for tubal block. Used in combination, they improve diagnostic accuracy and efficacy of sonosalpingography.

Keywords: B-Mode, Colour Doppler, Diagnostic accuracy, Sonosalpingography, Tubal patency

INTRODUCTION

Infertility is defined as one year of unprotected intercourse without conception. It is a major gynecological problem with complex etiology and about 10% cases remain unexplained after investigation.1 Testing for infertility is usually divided into three groups, depending upon the correlation with pregnancy rates, by the European Society of Human Reproduction and Embryology 2000 (ESHRE):2 1) With established association with pregnancy. 2) Not consistently associated with pregnancy. 3) No association with pregnancy.

Infertility can have several causes, however, tubal infertility accounts for nearly 30% of it.3 Functional competence of the fallopian tubes implies both tubal patency as well as mucosal integrity of the endosalpinx. Tubal factor infertility (TFI) includes an array of disorders affecting one or more components of the fallopian tubes. Demonstration of tubal patency using imaging modalities can help to lower the rate of invasive management. Laparoscopic assessment of tubal patency is the gold standard for the diagnosis of tubal patency.

Hysterosalpingography (HSG) is a widely used method for assessment of tubal patency. This method is fairly...
accurate in detecting proximal tubal disease, is safe, not much expensive and may potentially be associated with increased pregnancy rates. HSG provides optimal delineation of the fallopian tubes, allowing detection of tubal patency, tubal occlusion, tubal irregularity and peritubal disease. This method involves exposing the patient to unnecessary radiation. There is an added risk of iodine intolerance and hypersensitivity reactions associated with the procedure.

Sonosalpingography (SSG) using contrast media like Echovist, provides good results. However, the contrast dye is expensive and not available in all the countries. Many patients cannot afford it. SSG using saline infusion was started as an alternative to using ultrasound contrast media. Here, saline and air are used as contrast for ultrasound.

Normally, there is no air-fluid interface when the tubes are normal. This makes their visualization difficult. The fallopian tubes can only be visualized if there is hydrosalpinx, or fluid in the pelvic cavity, or if fluid is introduced into the tubes, externally. Initial studies used about 200 ml of saline, introduced trans-vaginally, through the cervix into the uterus. Fluid would fill up the uterine cavity and pass through the tubes into the pelvic cavity. Retro-uterine fluid seen on abdominal ultrasound was accepted as a criterion for patency.

The basic principle of SSG is to distend the uterine cavity with isotonic saline, which helps to delineate the contour, identify any intrauterine pathology, thickness of the endometrium, and also measure the amount of free-fluid in the pouch of Douglas (PoD).

Use of colour Doppler to further the assessment of tubal status has not been used popularly. While, B-mode alone may not be able to pin point the exact location of the tubal obstruction, facilitating the procedure with colour Doppler, can increase the sensitivity of the procedure. Movement of fluid through the fallopian tubes, can be visualized simultaneously using colour Doppler to find the exact location of obstruction. It gives better information regarding the assessment of each individual tube, rather than assessing only the spill of fluid.

There are studies that have compared colour Doppler to B-mode ultrasound for the assessment of tubal patency. Still others, have compared the efficacy of each in respect to laparoscopy. No other study has combined the use of colour Doppler and B-mode to collectively assess the tubal patency and assess the sensitivity of the procedure for the same.

**Methods**

It was a prospective study conducted at Dr. D.Y. Patil Hospital, Nerul, Navi Mumbai from December 2018 to October 2020.

**Inclusion criteria**

All females in the reproductive age group who presented with primary or secondary infertility. (Infertility was defined as the inability to conceive despite one year of unprotected intercourse). The procedure was done between 9th-11th day of the menstrual cycle to standardize the findings.

**Exclusion criteria**

Females presenting with lower abdominal pain or symptoms and clinical features suggestive of genitourinary tract infection and pelvic inflammatory disease.

**Procedure**

The procedures were done in the mid-proliferative phase of the menstrual cycle to reduce the risk of disturbing an early pregnancy, in females presenting with primary or secondary infertility. A detailed trans-vaginal ultrasound scan was done to assess the position of pelvic organs and to rule out any pathologies, which would come in the way of the procedure. Moreover, any free fluid in pelvis was also checked for, before beginning.

Strict asepsis was maintained. A 7F disposable balloon catheter specially devised for HSG was introduced through cervix into the uterus. The balloon was distended with 1-2 ml of distilled water or normal saline and was placed just beyond the internal os. Alternatives to this catheter are paediatric feeding tubes or small gauge Foley’s catheter. Once the catheter is fixed, trans-vaginal probe is introduced into the vagina for further assessment.

Saline was injected through the catheter slowly, and scanning was done to assess the uterine cavity that was subsequently distended by saline and also passage of saline (fluid) seen through the tubes. Once the uterine cavity filled with saline, endometrial pathologies were evaluated, if any. Spill of saline from fimbrial end is seen as fluid flow surrounding the ovary and its collection in pelvis on B-mode scanning. Absence of spill may indicate blockage. Absence of spill in the presence of pain on uterine distention is taken as a sign of tubal occlusion, whereas, retro-uterine spill is taken as a sign of tubal patency. Movement of fluid in each Fallopian tube with subsequent collection of fluid, or increase in the quantity of free-fluid, in the pouch of Douglas was taken as a sign of tubal patency.

On ultrasound Doppler, the colour box was placed on the transverse section of the uterus. Colour signals following-up the uterine cavity confirm the passage of fluid in the uterus as saline was injected through the cannula in the uterus in short bouts. The field of vision was immediately changed over to ovary and adnexa, by spanning the probe from transverse section of uterus, laterally.
injecting saline, colour box was placed to visualize the adnexa and ovary. Filling up of the box with colour signals indicates patency of the tube and absence of such signals indicate block.\textsuperscript{10} The same procedure was repeated on the other side.

The following parameters were studied for every patient: morphology of uterus, fallopian tubes and ovaries; movement of normal saline through the cervix into the uterus and fallopian tubes on transabdominal and transvaginal ultrasound; movement of normal saline through each fallopian tube individually on B-mode; fluid movement through each of the fallopian tubes on color Doppler; presence of obstruction/occlusion at the level of the fallopian tubes.

Written informed consent was obtained from each patient after explaining the radiological procedure.

**Ethical approval**

Ethical approval was obtained from the institution, Dr. D. Y. Patil University School of Medicine before carrying out the study.

**Statistical analysis**

We examined 52 patients and 104 tubes individually (52 right and 52 left fallopian tubes). R analysis was done for calculating results.

**RESULTS**

31 (59.6\%) of the participants had tubal block (B-mode USG): absent. 4 (7.7\%) of the participants had tubal block (B-mode USG): right. 14 (26.9\%) of the participants had tubal block (B-mode USG): left. 3 (5.8\%) of the participants had tubal block (B-mode USG): bilateral.

43 (82.7\%) of the participants had tubal block (colour Doppler): Absent. 2 (3.8\%) of the participants had tubal block (colour Doppler): right. 7 (13.5\%) of the participants had tubal block (colour Doppler): left.

41 (78.8\%) of the participants had tubal block (PoD fluid): absent. 3 (5.8\%) of the participants had tubal block (PoD fluid): right. 7 (13.5\%) of the participants had tubal block (PoD fluid): left. 1 (1.9\%) of the participants had tubal block (PoD fluid): bilateral.

**Table 1**: Distribution of imaging findings.

| Findings                          | Mean±SD; median (IQR); min-max; frequency (%) |
|----------------------------------|-----------------------------------------------|
| Movement of fluid (yes)          | 80 (76.9\%)                                    |
| Colour Doppler artifact (yes)    | 95 (91.3\%)                                    |
| Tubal block (PoD fluid) (present)| 12 (11.5\%)                                    |

**Table 2**: Association between tubal block (PoD fluid) and movement of fluid (n=104).

| Movement of fluid | Tubal block (PoD fluid) | Fisher’s exact test |
|-------------------|-------------------------|---------------------|
|                   | Present | Absent | Total | \( \chi^2 \) | P value |
| Yes               | 2 (2.5\%) | 78 (97.5\%) | 80 (100.0\%) | 27.746 | <0.001 |
| No                | 10 (41.7\%) | 14 (58.3\%) | 24 (100.0\%) |             |        |
| Total             | 12 (11.5\%) | 92 (88.5\%) | 104 (100.0\%) |             |        |

80 (76.9\%) of the participants had movement of fluid: yes. 24 (23.1\%) of the participants had movement of fluid: no.

95 (91.3\%) of the participants had colour Doppler artifact: yes. 9 (8.7\%) of the participants had colour Doppler artifact: no.

12 (11.5\%) of the participants had tubal block (PoD fluid): present. 92 (88.5\%) of the participants had tubal block (PoD Fluid): absent.

Fisher’s exact test was used to explore the association between ‘Tubal Block (PoD Fluid)’ and ‘Movement of Fluid’ as more than 20\% of the total number of cells had an expected count of less than 5.
There was a significant difference between the various groups in terms of distribution of tubal block (PoD fluid) ($\chi^2=27.746$, $p<0.001$).

2.5% of the participants in the group (movement of fluid: yes) had [tubal block (PoD fluid): present], 41.7% of the participants in the group (movement of fluid: no) had [tubal block (PoD fluid): present]. 97.5% of the participants in the group (movement of fluid: yes) had [tubal block (PoD fluid): absent], 58.3% of the participants in the group (movement of fluid: no) had [tubal block (PoD fluid): absent].

Participants in the group movement of fluid: no had the larger largest proportion of tubal block (PoD fluid): present. Participants in the group movement of fluid: yes, had the larger largest proportion of tubal block (PoD fluid): absent.

Fisher’s exact test was used to explore the association between ‘Tubal Block (PoD Fluid)’ and ‘Colour Doppler Artifact’ as more than 20% of the total number of cells had an expected count of less than 5.

There was a significant difference between the various groups in terms of distribution of tubal block (PoD fluid) ($\chi^2=57.753$, $p<0.001$).

4.2% of the participants in the group (colour Doppler artifact: yes) had [tubal block (PoD fluid): present], 88.9% of the participants in the group (colour Doppler artifact: no) had [tubal block (PoD fluid): present]. 95.8% of the participants in the group (colour Doppler artifact: yes) had [tubal block (PoD fluid): absent]. 11.1% of the participants in the group (colour Doppler artifact: no) had [tubal block (PoD fluid): absent].

Participants in the group colour Doppler artifact: no had the larger largest proportion of tubal block (PoD fluid): present. Participants in the group colour Doppler artifact: yes had the larger largest proportion of tubal block (PoD fluid): absent.

### Table 3: Association between tubal block (PoD fluid) and colour Doppler artifact (n=104).

| Color Doppler artifact | Tubal block (PoD fluid) | Fisher's exact test |
|-----------------------|------------------------|-------------------|
|                       | Present | Absent | Total | $\chi^2$ | P value |
| Yes                   | 4 (4.2%) | 91 (95.8%) | 95 (100.0%) | 57.753 | <0.001 |
| No                    | 8 (88.9%) | 1 (11.1%) | 9 (100.0%)  |          |        |
| Total                 | 12 (11.5%) | 92 (88.5%) | 104 (100.0%) |  |  |

Table 4: Comparison of tubal block (B-mode USG) with tubal block (PoD fluid) (n=52).

| Tubal block (B-mode USG) | Tubal block (PoD fluid) | Cohen’s Kappa |
|--------------------------|-------------------------|---------------|
|                         | Absent | Right | Left | Bilateral | Total | k   | P value |
| Absent                  | 30 (57.7%) | 1 (1.9%) | 0 (0.0%) | 0 (0.0%) | 31 (59.6%) | 0.409 | <0.001 |
| Right                   | 1 (1.9%) | 2 (3.8%) | 0 (0.0%) | 1 (1.9%) | 4 (7.7%)  |      |        |
| Left                    | 9 (17.3%) | 0 (0.0%) | 5 (9.6%) | 0 (0.0%) | 14 (26.9%) |      |        |
| Bilateral               | 1 (1.9%) | 0 (0.0%) | 2 (3.8%) | 0 (0.0%) | 3 (5.8%)  |      |        |
| Total                   | 41 (78.8%) | 3 (5.8%) | 7 (13.5%) | 1 (1.9%) | 52 (100.0%) |      |        |

Table 5: Comparison of tubal block (colour Doppler) with tubal block (PoD fluid) (n=52).

| Tubal block (colour Doppler) | Tubal block (PoD fluid) | Cohen's Kappa |
|-----------------------------|-------------------------|---------------|
|                            | Absent | Right | Left | Bilateral | Total | k   | P value |
| Absent                      | 40 (76.9%) | 2 (3.8%) | 0 (0.0%) | 1 (1.9%) | 43 (82.7%) | 0.765 | <0.001 |
| Right                       | 1 (1.9%) | 1 (1.9%) | 0 (0.0%) | 0 (0.0%) | 2 (3.8%)  |      |        |
| Left                        | 0 (0.0%) | 0 (0.0%) | 7 (13.5%) | 0 (0.0%) | 7 (13.5%)  |      |        |
| Bilateral                   | 0 (0.0%) | 0 (0.0%) | 0 (0.0%) | 0 (0.0%) | 0 (0.0%)  |      |        |
| Total                       | 41 (78.8%) | 3 (5.8%) | 7 (13.5%) | 1 (1.9%) | 52 (100.0%) |      |        |

The two methods agreed in 71.2% of the cases and disagreed in 28.8% of the cases.

There was moderate agreement between the two methods, and this agreement was statistically significant (Cohen’s Kappa =0.409, p<0.001).
The disagreements observed between the two methods were as follows:

1 (1.9%) cases classified as absent by tubal block (PoD fluid) were classified as right by tubal block (B-mode USG). 9 (17.3%) cases classified as absent by tubal block (PoD fluid) were classified as left by tubal block (B-mode USG). 1 (1.9%) cases classified as absent by tubal block (PoD fluid) were classified as bilateral by tubal block (B-mode USG). 1 (1.9%) cases classified as right by tubal block (PoD fluid) were classified as right by tubal block (PoD fluid) were classified as right by tubal block (PoD fluid) were classified as left by tubal block (PoD fluid) were classified as left by tubal block (PoD fluid). 2 (3.8%) cases classified as absent by tubal block (PoD fluid) were classified as bilateral by tubal block (PoD fluid). 1 (1.9%) cases classified as bilateral by tubal block (PoD fluid) were classified as right by tubal block (PoD fluid).

The two methods agreed in 92.3% of the cases and disagreed in 7.7% of the cases.

There was Substantial agreement between the two methods, and this agreement was statistically significant (Cohen’s Kappa =0.765, p<0.001).

### Table 6: Performance of study parameters for predicting tubal block (PoD fluid): present versus absent.

| Description of variables | Category(s) | Category(s) | Total positives | True positives | True negatives | False positives | False negatives |
|--------------------------|-------------|-------------|-----------------|---------------|---------------|----------------|----------------|
| Tubal block (PoD fluid)  | Present     | Absent      | 12 (11.5%)      | -             | -             | -              | -              |
| Movement of fluid        | No          | Yes         | 24 (23.1%)      | 10 (10%)      | 78 (75%)      | 14 (13%)       | 2 (2%)         |
| Color Doppler artifact   | No          | Yes         | 9 (8.7%)        | 8 (8%)        | 91 (88%)      | 1 (1%)         | 4 (4%)         |

### Primary diagnostic parameters

| Variable                     | Sensitivity | Specificity | PPV    | NPV    | Diagnostic accuracy |
|------------------------------|-------------|-------------|--------|--------|---------------------|
| Movement of fluid            | 83.3% (52-98) | 84.8% (76-91) | 41.7% (22-63) | 97.5% (91-100) | 84.6% (76-91) |
| Color Doppler artifact       | 66.7% (35-90) | 98.9% (94-100) | 88.9% (52-100) | 95.8% (90-99) | 95.2% (89-98) |

### Other diagnostic parameters

| Variable                     | LR+       | LR-       | Yuden index | Odds ratio | Kappa | P value |
|------------------------------|----------|----------|-------------|------------|-------|---------|
| Movement of fluid            | 5.48     | 0.20     | 68.1        | 27.86 (5.51-140.94) | 0.47  | <0.001  |
| Color Doppler artifact       | 61.33    | 0.34     | 65.6        | 182.00 (18.11-1828.83) | 0.74  | <0.001  |

The disagreements observed between the two methods were as follows:

1 (1.9%) cases classified as absent by tubal block (PoD fluid) were classified as right by tubal block (color Doppler). 2 (3.8%) cases classified as right by tubal block (PoD fluid) were classified as absent by tubal block (color Doppler) were classified as absent by tubal block (color Doppler).

**DISCUSSION**

Tubal evaluation is critical in the evaluation and work-up of sub fertile and infertile patients. For a long time, HSG was used for the assessment of tubal patency. Since the introduction of sonosalpingography and its technical modifications, it has emerged as a fairly reliable technique for the evaluation of tubal status. Introduction of contrast media, HyCoSy and HyFoSy have bettered its efficacy in diagnosing tubal patency and true tubal occlusion, respectively.

All studies point towards the importance of using sonosalpingography as a preliminary screening test, as it allows evaluation of the pelvic structures and ovaries in the same sitting, helping in the diagnosis of other potential causes of infertility. SSG has also been suggested as a basic test to guide laparoscopic evaluation for tubal patency. Among the 52 patients we examined, nearly 10 patients had concomitant presence of other pathologies as described below:

**Uterine pathologies**

Adenomyosis, intramural fibroids and submucosal fibroids. The most common being intramural fibroids.
(40%). One patient had bulky uterus, likely secondary to the presence of fibroids.

**Ovarian pathologies**

Ovarian cyst, and polycystic ovarian morphology. One patient presented with bulky unilateral ovary.

**Peritoneal pathologies**

Presence of free-fluid in the anterior pouch of Douglas with adhesions within the fluid.

Some studies say that SSG cannot pinpoint the location of the exact block and presence of free-fluid in the pouch of Douglas is suggestive of patency of at least one tube, without providing information as to the patency of bilateral tubes. While, other studies believe that diagnostic laparoscopy is required for the final and complete evaluation of infertility. But studies also show near identical efficacy and sensitivity of sonosalpingography, in comparison with diagnostic laparoscopy and HSG.

According to the study conducted by Malhotra et al, colour Doppler was not considered an essential part of the “Sion procedure” for assessing the flow of echo enhancing agent in the tube and spillage into the peritoneal cavity. The same study also suggests that SSG is unsuccessful in pin-pointing the location of the tubal block, for which HSG is better.

With similar results, the study conducted by Scanlan et al, showed, that while SSG can assess the patency of at least one tube, it cannot evaluate the block on either side or confirm bilateral tubal patency. However, according to Sheth et al, SSG meets all the requirements of a screening test for tubal patency, with an extremely low false positive rate.

In our study, real-time imaging, followed by careful tracing of each individual fallopian tube shows that SSG can be used to pin-point the exact location of the tubal block. Nearly 76.9% fallopian tubes showed movement of fluid through them, and 91.3% showed presence of colour aliasing artefact. Around 88.5% tubal patency was observed based on the presence of free-fluid pockets in the peritoneum, adjacent to either ovary.

The study using a lasting contrast agent like SonoVue, either through 2D or 3D reconstruction imaging, showed that each fallopian tube can be assessed in detail, from the uterine horn to their fimbrial ends. The contrast agent produces harmonic signals based only on the contrast enhancing microbubble spheres, filtering out signals from the surrounding tissue. Another similar study used gaseous spring water as contrast agent, without further ancillary investigations for confirmation, showing that HyCoSy is sufficient for evaluation of tubal patency.

When comparing this to our study, we found that using flow assessment through B-mode and aliasing through colour Doppler, has a high sensitivity (83.3% and 66.7% respectively) and specificity (84.8% and 98.9%) while assessing tubal patency. The diagnostic accuracy is 84.6% and 95.2% respectively, showing that further evaluation may not be required.

Adding colour Doppler may increase the efficiency and accuracy of SIS for assessment of tubal patency. In a study by Peters et al of 129 infertile patients, Doppler SIS showed complete agreement with HSG in 81% cases. When compared with the gold standard test of tubal assessment, chromopertubation, Doppler SIS showed agreement in 86% of cases, while HSG agreed with chromopertubation only in 75% cases. Correlation of colour Doppler sonosalpingography and HSG with chromopertubation was 81% versus 60% respectively in one study.

In our study by using B-mode only, we got a false positive ratio (study showed tubal block, when there was none) of 58.3%, whereas it was only 11.1% when using colour Doppler. The true positive ratio (study shows tubal block and it was present) was also higher using colour Doppler (88.9%), as compared to using B-mode (41.7%). The diagnostic accuracy with colour Doppler (95.2%) is higher than accuracy of using B-mode (84.6%). There was only 71.2% agreement with the presence of free-fluid in pouch of Douglas and hence tubal patency, while using B-mode, while the agreement was as high as 92.3% with colour Doppler. However, there was substantial agreement between the two methods, and the agreements were statistically significant (Cohen’s Kappa =0.765, p<0.001).

With HyCoSy, it is easy to diagnose tubal patency by evaluating the free flow of contrast through the fallopian tubes, but it difficult to diagnose true occlusion from false occlusion.

While tracing the flow of contrast through the entire tubal length increases confidence in the diagnosis of proximal and distal patency, this is more technically demanding. Sometimes only proximal patency can be demonstrated by visualizing paracornual flow of contrast.

Even in our study, tubal block was evaluated at the cornua of the uterus. In addition, slight overestimation of tubal block was noted using B-mode, likely due to the tubal spasm, also known as false occlusion.

However, there was slight underestimation of tubal occlusion using colour Doppler (lower sensitivity), possibly due to the presence of surrounding vessels, which may contribute to the contamination of the colour aliasing artifact, due to fluid movement.

Procedure failure due to technical difficulties of either visualization or canalization are higher with SSG and lie
between 2-8%.14 Within our study, among 55 women examined, 3 procedures had to be aborted due to technical difficulties viz. leaking of fluid from the catheter, improper placement of the inflatable catheter within the cervix, severe pain to the patient, and vasovagal symptoms.

Limitations of the study is that while imaging collection of fluid, in bilateral pockets of pouch of Douglas, in real-time is highly sensitive, due to high attrition rate, the study findings could not be compared to other standard and gold-standard methods of evaluating tubal patency.

CONCLUSION

Tubal evaluation forms a crucial element of infertility investigation among the female factors. Among the various methods, HyCoSy is a fairly reliable diagnostic tool for the evaluation of tubal patency. Technical modifications have further enhanced the utility of this technique, which is nearly as accurate and sensitive as HSG, without exposing the patient to radiation. B-mode is used to see the flow of saline fluid (or contrast media) from the endometrial cavity to the uterine cornua, and through the fallopian tubes till the fimbrial ends. Colour Doppler is used to see the colour aliasing artifact noted within the fallopian tube due to movement of fluid through it.

Use of colour Doppler, increases the sensitivity for evaluation of tubal patency, with slightly underestimation of tubal block (possibly due to contamination by surrounding vessels). As compared to B-mode, it has significantly higher sensitivity and specificity for the diagnosis of tubal patency and higher specificity for diagnosis of tubal block. Overall, it has a better diagnostic accuracy. However, B-mode evaluation has better sensitivity and agreement in the diagnosis of tubal block, with slight overestimation of tubal block (likely due to diagnosis of false occlusion). Using B-mode and colour Doppler in combination and combining results before reporting, significantly improve the diagnostic accuracy and efficacy of sonosalpingography, alleviating the need for further diagnostic interventions that are painful, and associated with higher risks and procedural complications.

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