





Original Research

Fallopian tube patency diagnosed by laparoscopic chromopertubation in women with polycystic ovarian syndrome and non-subfertile controls: a retrospective case-control study

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Abstract

Background: Hyperandrogenism has been associated with tubal dysfunction. Previous studies have not used non-subfertile controls when assessing hyperandrogenic women through laparoscopic chromopertubation (LC), the gold standard for tubal patency testing. **Methods:** The objective of this retrospective study was to compare the results of LC between women with medication-resistant anovulatory polycystic ovary syndrome (PCOS, $n = 202$) and non-subfertile women with ovarian cysts (controls, $n = 48$). **Results:** There were no statistically significant differences between PCOS women and controls for the prevalence of bilateral tubal occlusion (2.5% versus 4.2%, respectively; $p = 1.000$) or overall rates for occlusion (7.4% of tubes assessed versus 6.3%, respectively; $p = 0.828$). More PCOS patients needed high pressure chromopertubation to demonstrate bilateral tubal patency than controls (12.4% versus 2.1%; $p = 0.036$). In the PCOS group, patients needing higher pressure to achieve patency demonstrated higher testosterone levels than women requiring typical pressure (0.66 ± 0.17 versus 0.47 ± 0.24 ng/mL; $p = 0.001$). **Conclusions:** For women with ovarian cysts and without subfertility, six percent of all Fallopian tubes are occluded. PCOS patients have similar rates of tubal occlusion, making these women a reasonable but more available control group for LC studies. Higher total testosterone levels were associated with higher LC pressures to demonstrate bilateral patency.

Keywords: Fallopian tube; Polycystic ovary syndrome; Chromopertubation; Androgens; Fertility

1. Introduction

It is estimated that 25 to 35 percent of female infertility is associated with tubal abnormalities through functional or anatomic causes [1]. Extensive emphasis is given to mechanical injury, such as through peritubal adhesions, fimbrial agglutination, and intraluminal obstruction and ciliary damage. Though there has been meaningful investigation of how peritonitis influences patency, understanding of how hormonal shifts affect tubal patency is limited.

Distinct from mechanical obstruction, it is established that androgens can affect tubal function [2,3]. Ciliary flagellation decreases with testosterone exposure [2]. Accumulation of intraluminal secretions and cellular debris increases for testosterone-pretreated transmen with supra-physiologic hyperandrogenism [3]. In a retrospective case series of our study group, eight percent of women with PCOS (showing signs of elevated testosterone levels) were found to have occluded fallopian tubes during laparoscopy [4]. Though hormonal effects may hinder tubal patency, they have not been proven to cause complete or lasting obstruction.

With several studies suggesting that hyperandrogenism is associated with multiple risks for tubal dysfunction [2,3], it is natural to question whether hormonal shifts increase occlusion in general when compared with otherwise healthy women. Further, the literature also lacks data regarding the incidence of tubal blockage in a general population of women without subfertility. Since laparoscopic chromopertubation is rarely performed in women without infertility, it would be of interest to study the tubal patency of a healthy population and to compare it to women with hyperandrogenism. To achieve this end, we explored the role of hyperandrogenism on tubal patency by comparing infertile, anovulatory women undergoing laparoscopy with chromopertubation for PCOS and non-subfertile women undergoing the procedure for ovarian cyst indications, excluding participants with concurrent endometriosis or pelvic inflammatory disease (PID).



Table 1. Inclusion and exclusion criteria for both study groups.

	PCOS group	Control group
Inclusion criteria	Combined hysteroscopy/laparoscopy for ovarian drilling PCOS according to the revised Rotterdam criteria [8] Clomiphene or letrozole resistance [9]	Combined hysteroscopy/laparoscopy for removal of an ovarian cyst Non-infertile women
Exclusion criteria	- Previous removal of one or both Fallopian tubes, hysteroscopically proven endometrial polyps, uterine myomas, endometriosis, adenomyosis, hydrosalpinx, intraabdominal adhesions	Polycystic ovarian morphology

2. Methods

2.1 Patient population

In a retrospective study, all women were included who between January, 2008 to December, 2020 underwent combined hysteroscopy/laparoscopy for laparoscopic ovarian drilling with chromopertubation at the Clinical Division of Gynaecologic Endocrinology and Reproductive Medicine of the Medical University of Vienna, Austria. Details relating to the majority of these patients have already been published [4–7]. PCOS had been diagnosed according to the revised Rotterdam criteria [8]. All PCOS patients underwent ovarian drilling for clomiphene or letrozole resistance [9]. As a control group, we included all non-infertile women ages 18–44 years who had not actively planned to become pregnant so far but underwent hysteroscopy/laparoscopy with chromopertubation for removal of a non-endometrioma ovarian cyst over this same time period. Hysteroscopy and chromopertubation were performed in order to provide additional information to these patients. Women were excluded from both groups in case of previous removal of one or both Fallopian tubes, hysteroscopically proven endometrial polyps (since the removal of these polyps by curettage or operative hysteroscopy might have influenced Fallopian tube function), uterine myomas (since they might obstruct tubes or alter the pressure needed for chromopertubation), endometriosis, adenomyosis, hydrosalpinx, and intraabdominal adhesions. All women in the control group had regular cycles and did not have polycystic ovarian morphology on ultrasound. Accordingly, there were no PCOS women in the control group. Details about in- and exclusion criteria can be found in Table 1 (Ref. [8,9]).

This resulted in a final patient population of 250 women (PCOS group, $n = 202$, control group, $n = 48$). The study was approved by the Institutional Review Board of the Medical University of Vienna (IRB number 2371/2020). Data in this retrospective study was anonymized; thus, the requirement for informed consent was waived.

2.2 Surgical technique

All surgical procedures were conducted under general anaesthesia and either directly performed or supervised by

experts in infertility surgery. For women with laparoscopic ovarian drilling, a hysteroscopy was performed first followed by laparoscopy. The technique has been published previously [7,10]. For diagnostic hysteroscopy, a forward-oblique 30° hysteroscope (Karl Storz GmbH & Co. KG, Tuttlingen, Germany; 5 mm outer sheath diameter) was used.

Before laparoscopy, a *Spackmann* uterine manipulator with clamp fixation and a rubber cone with a diameter of 18 mm (Reference number 1264; WISAP® Medical Technology GmbH Brunnthal/Hofolding, Brunnthal, Germany) was placed through the cervix and advanced to one centimeter from the uterine fundus for each patient before laparoscopy with chromopertubation. During laparoscopy, a thorough inspection of the pelvis, internal genitalia, and liver region was conducted. Chromopertubation was performed using a 50 mL syringe with a dilute solution of indigo carmine blue dye (Amino AG, Gebenstorf, Switzerland) through the uterine manipulator. Parameters recorded include patency of the Fallopian tubes, the volume of dye utilized with chromopertubation, and an assessment of the subjectively required pressure to achieve tubal patency as reported previously [6,7].

2.3 Parameters analysed

The major outcome parameter was tubal patency as assessed by laparoscopic chromopertubation, with documentation for each tube. This also included information about the subjectively required pressure to achieve tubal patency (normal, i.e., low pressure, and high pressure, which required deliberate exertion). All operations were performed or supervised by reproductive surgeons experienced in the assessment of Fallopian tube patency and integrity. The pressure needed to achieve tubal patency was defined as low, when the tubes were patent using only minimal pressure and only a small amount of blue dye was required. If meaningful pressure was required or indigo carmine was egressing more readily through the cervix than tubes despite appropriate uterine manipulator placement, high pressure was diagnosed. Demographic parameters include: patients' age and body mass index (BMI); the surgical indication; and additional findings/surgical procedures in the

course of hysteroscopy/laparoscopy, including intracavitary abnormalities, and the presence of endometriosis, hydrosalpinx or hydrosalpinges, and peritubal adhesions.

In the PCOS group, hormonal parameters were measured on cycle days two to five via phlebotomy one to three months before the operation. All examined serum parameters were determined in the ISO-certified central laboratory of the General Hospital of Vienna, Vienna, Austria using commercially available assays. Radioimmunoassays were used to determine serum levels of luteinizing hormone (LH; Autodelphia; Wallac Oy, Turku, Finland), follicle stimulating hormone (FSH; Enzymun ES700; Böhringer Mannheim, Mannheim, Germany), testosterone (Immunotech, Westbrook, ME, USA), androstenedione (Immunotech, Westbrook, ME, USA), and dehydroepiandrosterone (DHEAS).

2.4 Statistical analysis

Statistical analyses were performed with the SPSS software package, version 24.0 (SPSS, Chicago, IL, USA). Nominal variables are reported as mean and standard deviations and continuous variables with median and range. Differences in numerical and categorical parameters between groups were calculated using the unpaired *t*-test and the chi-square test/Fisher's exact test, respectively. Differences were considered significant if $p < 0.05$.

3. Results

No difference was observed for mean patient age at the time of surgery between women of the PCOS and women of the control group (28.3 ± 4.9 versus 29.2 ± 4.1 years, respectively; $p = 0.226$). PCOS women were found to have a statistically higher BMI, consistent with greater risk for the metabolic syndrome (PCOS: 25.3 ± 4.5 kg/m²; $p = 0.022$, controls: 23.3 ± 4.3 kg/m²). No participants in either the PCOS or in the control group had undergone previous abdominal surgery. In PCOS women, the mean LH:FSH ratio was 2.3 ± 1.3 . Mean serum testosterone, androstenedione and DHEAS levels were 0.5 ± 0.2 ng/mL, 3.7 ± 2.3 ng/mL, and 2.6 ± 1.2 µg/mL, respectively.

Regarding laparoscopic chromopertubation, PCOS patients had greater need for high pressure chromopertubation to demonstrate bilateral tubal patency to an extent that statistically was significantly more often than controls (12.4% versus 2.1%; $p = 0.036$). There were no other differences between the groups, which also includes the total number of occluded tubes and the total number of patent tubes achieved by high chromopertubation pressure only ($p > 0.05$; Table 2).

We also substratified PCOS women by comparing those having bilateral patency with low pressure ($n = 172$) to those with high pressure patency on chromopertubation ($n = 25$; Table 3). The latter revealed higher total testosterone levels (0.66 ± 0.17 versus 0.47 ± 0.24 ng/mL; $p = 0.001$) and higher DHEAS levels (3.57 ± 1.07 versus 2.51 ± 1.13 µg/mL; $p < 0.001$), whereas there were no differ-

ences in the LH:FSH ratio or in androstenedione levels, or in estradiol levels.

4. Discussion

In this retrospective case-control study, about seven percent of all Fallopian tubes were occluded. Of note, it is rare to see laparoscopic chromopertubation control groups where no participants have immediate procreative goals. Given that tubal factor accounts for up to thirty percent of subfertility and is one of the most common etiologies [1], it is important to know that only about four percent of women without subfertility have bilateral occlusion, emphasizing the close link between tubal disease and nonconception [1].

No differences were noted between women with PCOS or with ovarian cysts for the total number of occluded tubes or the number with bilateral blockage (Table 2). Accordingly, given the typical difficulty in obtaining control groups without subfertility, it would be reasonable to consider using clomiphene- or letrozole-resistant PCOS women without known PID, hydrosalpinges, or endometriosis as a laparoscopic control group for chromopertubation studies. However, it was more common in PCOS patients to require high chromopertubation pressure in order to achieve patency (12.4% versus 2.1%; $p = 0.036$; Table 2), suggesting that the Fallopian tubes may be inherently different for patients with PCOS. With multiple studies showing testosterone to influence Fallopian tube function [2,3,11], it seems reasonable to find differences.

Given that differences in testosterone levels do not seem to increase occlusion, two main factors may explain why PCOS patients require higher chromopertubation pressures to demonstrate patency. First, they have higher BMI, where increasing intraabdominal pressures (accentuated by abdominal adiposity) lead to a need for greater pressure to demonstrate patency. However, with a mean BMI of 25.3 relative to 23.3 in ovarian cyst controls, a two-point shift in BMI is unlikely to account for a six-fold increase in needing additional pressure. Moreover, standardization of intraabdominal insufflation pressures (12 mm Hg) may lead to less space with pneumoperitoneum in patients with a higher BMI means that the amount of pressure on the tubes still remains fifteen millimeters of mercury.

If neither obstruction nor obesity are reasons for needing higher pressures to achieve patency in PCOS women, then this may derive from shifts in the tubal lumen from hyperandrogenism. As noted in the introduction, PCOS women have slower flagellation of cilia associated with testosterone exposure [2]. Also, there is increased intraluminal accumulation of secretions and cell debris for the Fallopian tubes of testosterone-pretreated transmen [3]. Admittedly, the supraphysiologic testosterone levels in transmen do not reflect those in PCOS women, consistent with PCOS patients in our study having a mean testosterone level of 0.5 ng/mL. However, plugging of the tubes through intraluminal debris has been postulated as a reason for sub-

Table 2. Results of laparoscopic chromopertubation in the PCOS and the control groups.

	PCOS group	Control group	<i>p</i>
Number of patients	202	48	-
Bilateral tubal occlusion#	5 (2.5)	2 (4.2)	1.000
Bilateral tubal patency achieved with high chromopertubation pressure only#	25 (12.4)	1 (2.1)	0.036
Bilateral tubal patency achieved with normal, i.e., low chromopertubation pressure#	172 (85.1)	45 (93.8)	0.154
Number of Fallopian tubes	404	96	-
Number of occluded Fallopian tubes*	30 (7.4)	6 (6.3)	0.828
Number of tubes with patency achieved with high chromopertubation pressure only*	56 (13.9)	10 (10.4)	0.502
Number of tubes with patency achieved with normal, i.e., low chromopertubation pressure*	318 (78.7)	80 (83.3)	0.398

Data are provided as numbers and frequencies. Findings are stratified by #per patient or *per Fallopian tube.

Table 3. Hormone levels in PCOS patients with bilateral tubal patency according to chromopertubation pressure.

	Low pressure (n = 172)	High pressure (n = 25)	<i>p</i>
Total testosterone (ng/mL)	0.47 ± 0.24	0.66 ± 0.17	0.001
DHEAS (µg/mL)	2.51 ± 1.13	3.57 ± 1.07	<0.001
LH:FSH ratio	2.2 ± 1.3	2.4 ± 1.2	0.288
Androstenedione (ng/mL)	3.7 ± 2.4	3.7 ± 1.3	0.899
Estradiol (pg/mL)	77.5 ± 71.0	71.3 ± 45.7	0.686

Data are provided as mean ± standard deviation.

fertility. Notably, it was found that women with high pressure patency was associated with significantly higher testosterone levels. Though high testosterone levels are associated with both accumulation of intraluminal debris [3] and high-pressure patency, future studies will have to directly correlate these issues, as we did not specifically track which patients had increased intraluminal contents. Moreover, this debris does not invade the tubal lumen so one would not expect a meaningful increase in pressure to be required for displacement. These findings need to be compared to fecundity, as high pressure patency has been linked with lower pregnancy rates [12].

Study limitations which need to be mentioned include the retrospective study design as well as the sample size in the control group. In detail, given a prevalence of bilateral Fallopian tube occlusion of 6.3% in the control group, a minimum sample size of 47 patients per group would have been sufficient to show a significant difference between the two groups, if the rate for bilateral occlusion was 25% in PCOS women (power 80%, alpha 0.05, after Fleiss correction). This underscores the fact that a larger sample size would be needed to completely rule out minor differences between the groups. However, 48 patients undergoing chromopertubation without subfertility is a larger control group than is readily found in the literature for chromopertubation in non-subfertile women. Another opportunity for improvement is that the pressure needed to achieve tubal patency was only measured subjectively; however, most research on tubal patency doesn't account at all for the degree of pressure required. Studies which use a direct

measurement of chromopertubation pressure are underway. Also, subjectively measured pressures may have influenced by confounding variables other than tubal function. However, since many of these possible variables were applied as exclusion criteria and since only experts in tubal surgery were responsible for chromopertubation, we consider this a minor limitation only. Additionally, for generalizability, PCOS participants were medication resistant to the point of undergoing ovarian drilling, which doesn't reflect the majority of women with PCOS. Also, hormone levels were not available for the control group. However, since none of the women in the control group met the revised Rotterdam criteria, we would expect normal testosterone profiles even though this was not directly confirmed. Additionally, for many women hysteroscopy was performed prior to laparoscopy. Were there longer operative hysteroscopies, myometrial edema could have created a false positive for occlusion with chromopertubation when the tubes would normally be patent. However, access to the uterine cavity was easy in all patients and operative hysteroscopies had brief duration, making this consideration unlikely to meaningfully shift results. Last but not least, though pelvic inflammatory disease was an exclusion criteria, medical history about other sexually transmitted infections (STI), which could have resulted in tubal adhesions and blockage, were not available. However, when most women who have had an STI are unaware of it, not accounting for this factor may actually enhance generalizability.

5. Conclusions

This retrospective case-control study adds three central findings to the literature. First, for women without subfertility and without known risk factors for tubal disease, six percent of all Fallopian tubes are occluded and four percent of women have bilateral blockage. Second, women with medication-resistant anovulatory PCOS requiring drilling have similar rates of tubal occlusion relative to patients without subfertility, making these women a reasonable but more available control group for laparoscopic chromopertubation studies. Third, higher total testosterone and DHEAS levels were found in enrolled PCOS patients and this was associated with a need for higher chromopertubation pressures to demonstrate bilateral patency. Future studies should directly correlate accumulation of tubal debris with high pressure patency and on quantifying chromopertubation pressure objectively rather than subjectively.

Author contributions

DM—protocol and project development, data collection and management, manuscript writing; JPP—protocol and project development; MH—data collection and management, manuscript editing; RM—data analysis; JO—protocol and project development, data analysis, data collection and management, manuscript writing. All authors read and approved the final manuscript.

Ethics approval and consent to participate

This study was approved by the Institutional Review Board (IRB) of the Medical University of Vienna (IRB number 2371/2020). Data in this retrospective study was anonymized; thus, the requirement for informed consent was waived.

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Conflict of interest

The authors declare no conflict of interest. JO is serving as one of the Editorial Board members and Guest editors of this journal. We declare that JO had no involvement in the peer review of this article and has no access to information regarding its peer review. Full responsibility for the editorial process for this article was delegated to MD.

Availability of data and material

The datasets generated and analyzed during the current study are not publicly available, since the dataset will be used for other retrospective analysis. The data are available from the corresponding author upon reasonable request.

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