WITHDRAWN: The role of maintaining the integrity of sexual response cycle in the improvements of ovulation rate and pregnancy outcomes

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Research Article

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EDITORIAL NOTE:

The full text of this preprint has been withdrawn by the authors while they make corrections to the work. Therefore, the authors do not wish this work to be cited as a reference. Questions should be directed to the corresponding author.

Abstract

Background

Previous studies reported that the timing and frequency of intercourse were associated with the menstrual cycle and immune response, with fewer research on the correlation between sexual response cycle and ovulation rate or pregnancy outcomes.

Methods

We performed a prospective cohort study of 150 patients undergoing ultrasonic ovulation monitoring, with guidance on sexual behavior, psychological counseling, alkalizate on of the vaginal environment conducted for the experimental group(n = 75).

Outcomes:

The overall score of FSFI questionnaire, the score of sexual satisfaction degree, the incidence of orgasm in coitus, the pre-pregnancy cycle of ovulation monitoring, periodic ovulation rate, clinical pregnancy rate, biochemical pregnancy rate, live birth rate and abortion rate of the two groups were analyzed.

Results

After intervention, the incidence of FSD in the experimental group was decreased, with an improvement in sexual satisfaction degree and a higher incidence of orgasm. The periodic ovulation rate, clinical pregnancy rate and live birth rate in the experimental group were significantly higher, with shorter average pre-pregnancy cycle of ovulation monitoring. Furthermore, there were no differences in biochemical pregnancy rate and abortion rate.

Conclusion

Taken together, guidance on sexual behavior not only enhance the relationship between couples and alleviate sexual dysfunction, but also have crucial reproductive significance.

1 Introduction

From puberty to menopause, ovarian morphology and function show periodic changes in reproductive-aged women. Currently, it is generally accepted that, unlike some mammals that require mating stimulation to ovulate, human female ovulation is spontaneous and not affected by sexual behavior. However, relevant studies[1–3]pointed out that there were complex interactions of sexual behavior,

menstrual cycle phase, and humoral immunity, whose findings supported the hypothesis that shifts in humoral immunity caused by sexual activity across the menstrual cycle were associated with reproductive effort. Correspondingly, from the perspective of the integrity of sexual response cycle of the human female, our researchers were committed to figuring out whether the ovulation rate and pregnancy outcomes in women of childbearing age could be improved. The complete sexual response cycle includes sexual desire phase, sexual arousal phase, sexual plateau phase, sexual orgasm phase and sexual resolution phase, in which an unusual abnormality in one or more phases may lead to female sexual dysfunction (FSD)[4], the overall incidence of which reaches as high as 40%[5]. Ovulation is the basis condition of fertilization, and sexuality is also closely associated with natural conception, with the incidence of FSD in infertile women up to 87.1%[6]. Accordingly, in our study, systematic guidance on sexual behavior and psychological counseling were provided for patients in the experimental group, in order to observe the differences of periodic ovulation rate, clinical pregnancy rate, live birth rate and the occurrence of adverse pregnancy events between the two groups.

2 Subjects And Methods

2.1 Subjects

A total of 236 patients undergoing ovulation monitoring in the outpatient department of reproductive endocrinology of West China Second Hospital of Sichuan University between March 2021 and March 2022 were selected and screened according to the inclusion and exclusion criteria.

The inclusion criteria were as follows: (1) women of childbearing age between 20–35 with fertility requirements; (2) no organic lesion of uterus or bilateral adnexa; (3) serum anti-Müllerian hormone (AMH) > 1.1 ng/ml[7]; (4) the husband's semen was normal.

The exclusion criteria were as follows: (1)body mass index (BMI) ≥ 28 kg/m^2 [8]; (2) chromosomal abnormalities in either spouse; (3) suffering from acute genitourinary infectious diseases or sexually transmitted diseases; (4) suffering from serious mental illness; (5) merge with other endocrine system diseases (such as diabetes, thyroid diseases, adrenal diseases, etc.); (6) allergic reaction to human chorionic gonadotropin (HCG) or combined with other contraindications.

The sample size of this study was determined based on the previous studies by Tierney Lorenz[1–3], which also investigated the interaction between sexual behavior, reproduction and immunity. Finally, 150 subjects were screened on the basis of the predetermined inclusion and exclusion criteria described above, and all the couples included had no religious beliefs. Our study was approved by the ethic committee of West China Second University Hospital, and all subjects signed an informed consent. All subjects underwent ovulation monitoring, while they were divided into experimental group (75 cases) and control group (75 cases) according to whether they received guidance on sexual behavior, psychological counseling and treatment of alkalizing the vaginal environment during the periovulatory period. The general information of all subjects was recorded in detail, including age, BMI, the education level of both

spouses, menstrual cycle, pregnancy and childbirth history, serum AMH level, endometrial thickness on HCG injection day.

2.2 Methods

2.2.1 The follicular monitoring and ovulation triggering.

All patients underwent transvaginal ultrasonography (Philips Affiniti70) to closely monitor endometrial thickness (no difference) and follicular development from the 10th day of menstrual cycle. 10000 IU of HCG (Lizhu Pharmaceuticals, Zhuhai, China) was injected intramuscularly to trigger ovulation when the mean maximum follicular diameter (MFD) ≥ 18 mm. If transvaginal ultrasound indicated that there were more than 3 dominant follicles, of which the diameter ≥ 14 mm, this treatment cycle should be cancelled[9]. Besides, subjects were informed to intercourse on the same and next day of HCG injection, and ultrasonography was re-examined to observe whether ovulation occurred 48 hours after injection.

2.2.2 Sex education and psychological counseling were provided for the experimental pairs

The systematic and comprehensive knowledge of sex would be imparted to couples of the experimental group before ovulation monitoring, which mainly included the following aspects:

1. Anatomical study of the female reproductive system: A series of pictures and videos of female genitalia (which included vagina, uterus, fallopian tube, ovary, labium majus, labium minus, external orifice of urethra, vaginal orifice and clitoris, etc.) were shown to the spouses in order to help them understand the anatomical and physiological structure of women, which facilitated the identification of the erogenous zones. Erogenous zones are areas of the body which arouse sexual excitement when stimulated, which generally include lip, ear, neck, breast, nipple, clitoris, labium minus, lower abdomen and inner thigh.
2. The sexual response cycle: The definition and phases of sexual response cycle were explained, which included sexual desire phase, sexual arousal phase, sexual plateau phase, sexual orgasm phase and sexual resolution phase.
3. Appropriate environment for sexual intercourse and sexual hygiene: The dark, comfortable circumstance is favorable for intercourse, which helps the intimate partner feel safe and relaxed. Then we should explain to patients the significance of stimulating erogenous zones before sex behaviors. Sometimes, fresh environment or erotic videos could also be conducive to stimulating sexual desire. Additionally, taking a bath before sex is an effective way to improve sex experience and prevent genital tract infection.
4. Psychological factors on sexual function: Counseling and psychological services were provided with the purpose of getting over nervousness and pressure, which help the couples realize that sex is a mutual need and pay attention to communication during sex.

2.2.3 Sexual behavior inquiry

All subjects were required to fill in the female sexual function index (FSFI) questionnaire[10] when they were enrolled, and the experimental group were asked to fill out the questionnaire again after receiving sex education.

2.2.4 Detection of serum sex hormones

In order to determine the time of ovulation triggering more accurately, the level of estradiol (E2) and luteinizing hormone (LH) in serum were detected separately with Estradiol ELISA Kit (Estradiol Enzyme-Linked ImmunoSorbent Assay Kit) and Luteinizing Hormone Human ELISA Kit (ab178658).

2.2.5 Observation of the characteristics of cervical mucus and alkalization of the vaginal environment

During the peri-ovulation period, the pH of cervical mucus (CM) was measured by pH indicator strips (Guangzhou Ikeme Technology Co., Ltd.), with the structure of CM observed under the microscope (Nikon, TiU). Additionally, to improve sperm survival and motility, corresponding treatment measures were taken including removing the sticky leucorrhea covered on the cervical surface with cotton swabs and wiping the cervix and vagina walls with sterile soap solution to alkalinize the vaginal environment.

2.2.5 Diagnosis of ovulation and pregnancy

Serum sex hormone levels were detected to determine whether ovulation or pregnancy 10 days after the re-examination of ultrasound. The following signs indicate ovulation: (1) serum progesterone (P) >3 ng/ml; (2) a decrease in the number of follicles; (3) occurrence of luteal echotexture. The diagnosis criteria of clinical pregnancy are that the β-human chorionic gonadotrophin (β-HCG) was positive with gestational sac found in the uterine cavity by ultrasonography[11]. The definition of biochemical pregnancy is that the β-HCG in blood or urine is positive, but terminates without visualization of a gestational sac through ultrasonographic check[12]. The pregnancy outcomes of the two groups were followed up. If pregnancy did not occur, continuous monitoring of ovulation with HCG therapy should not exceed three cycles.

2.3 The outcome measures

The incidence of FSD (FSFI-score ≤ 26.55), the score of sexual satisfaction degree, the incidence of orgasm in coitus, the pre-pregnancy cycle of ovulation monitoring, periodic ovulation rate, clinical pregnancy rate, biochemical pregnancy rate, live birth rate and abortion rate of the two groups were calculated.

2.4 Statistical analysis

The statistical analysis was conducted using SPSS version 23.0 software (SPSS Inc: Chicago, IL, USA). The quantitative data coincident with normal distribution were described as the mean ± standard deviation (SD) and were analyzed by the t-test for comparison between groups, while the qualitative data

were expressed as numbers and percentages (%) and the analysis of data was done using Chi-square test. P-value ≤ 0.05 was considered statistically significant.

3 Results

3.1 Baseline data of the two groups

There was no significant difference in age, BMI, menstrual cycle, pregnancy and childbirth history, serum AMH level, endometrial thickness on the day of triggering ovulation between the experimental group and the control group (all P > 0.05). The analysis of the education level of both spouses was done by Mann Whitney U-Test, and there were no statistical differences in wife's educational level (Z = 0.680, P = 0.496) and the husband's (Z = 0.255, P = 0.799) separately between the two groups, which indicated that all baseline characteristics were comparable (Table 1).

Table 1

Comparison of the baseline data of the two groups ($\bar{x} \pm s$)

	Experimental Group	Control group	*t*	*P*
Age (year)	28.24 ± 2.95	28.53 ± 3.05	0.599	0.550
BMI (kg/m^2)	22.55 ± 2.71	22.44 ± 2.51	0.264	0.792
Menstrual cycle (day)	29.96 ± 3.09	30.08 ± 3.18	0.235	0.815
Pregnancy history	2.03 ± 1.39	2.00 ± 1.20	0.126	0.900
Childbirth history	0.56 ± 0.55	0.59 ± 0.59	0.285	0.776
AMH (ng/ml)	4.31 ± 2.29	4.26 ± 2.63	0.125	0.901
Single-layer endometrial thickness (cm)	0.41 ± 0.14	0.41 ± 0.13	0.155	0.877
Abbreviations: BMI, Body Mass Index; AMH, anti-Müllerian hormone.				

3.2 The incidence of FSD

There was no statistical difference in the incidence of FSD between the experimental group (56.00%, 42/75) and the control group (52.00%, 39/75) when subjects were enrolled (χ^2=0.242, P = 0.623). After receiving sex education, the incidence rate of FSD in the experimental group decreased to 25.33% (19/75), and there was a significant difference (χ^2=11.244, P 0.001) compared with the control group (Table 2).

3.3 The proportion of patients reaching orgasm and the score of sexual satisfaction degree

85.33% (64/75) of the patients with anatomical education and sexual behavior guidance in the experimental group reached orgasm, while the incidence of orgasm in the control group was only 56.00% (42/75), the difference was statistically significant (χ^2=15.566, P 0.001) (Table 2). Furthermore, the score of sexual satisfaction degree in the experimental group and in the control group was 4.17±1.15 and 3.13±1.26 respectively, with significant difference (t=5.270, P 0.001) (Table 3).

Table 2
Comparison of the quality of sexual life between the experimental group (after intervention) and the control group % (n)

	Experimental Group	Control group	χ^2	*P*
incidence rate of FSD	25.33 (19/75)	52.00 (39/75)	11.244	<0.001
incidence of orgasm	85.33 (64/75)	56.00 (42/75)	15.566	<0.001
Abbreviation: FSD, female sexual dysfunction.				

Table 3
Comparison of the score of sexual satisfaction and average treatment cycle between the two groups ($\bar{x} \pm s$)

	Experimental Group	Control group	*t*	*P*
score of sexual satisfaction	4.17 ± 1.15	3.13 ± 1.26	5.270	<0.001
average treatment cycle	1.89 ± 0.76	2.21 ± 0.72	2.637	0.009

3.4 Comparison of ovulation and pregnancy outcomes between the two groups

The average pre-pregnancy cycle of ovulation monitoring in the experimental group and in the control group was 1.89 ± 0.76 and 2.21 ± 0.72 respectively, with significant difference (t = 2.637, P = 0.009) (Table 3). After ultrasonic ovulation monitoring treatment, the periodic ovulation rate of the experimental group was 88.03% (125/142), which was significantly higher (χ^2 = 4.529, P = 0.033) than that of the control group 78.92% (131/166) (Table 4).

The clinical pregnancy rates of the experimental group and the control group were 68.00% (51/75) and 49.33% (37/75) respectively, with statistical significance (χ^2 = 5.389, P = 0.020). The live birth rate in experimental group (62.67%, 47/75) were significantly higher (χ^2 = 7.712, P = 0.005) than that in control group (40.00%, 30/75). In the two groups, biochemical pregnancy rate and abortion rate had no statistically significant differences (χ^2 = 0.174, 0.883; P = 0.677,0.347) (Table 4).

Table 4
Comparison of ovulation and pregnancy outcomes between the two groups % (n)

	Experimental Group	Control group	χ^2	*P*
periodic ovulation rate	88.03 (125/142)	78.92 (131/166)	4.529	0.033
clinical pregnancy rate	68.00 (51/75)	49.33 (37/75)	5.389	0.020
live birth rate	62.67 (47/75)	40.00 (30/75)	7.712	0.005
biochemical pregnancy rate	2.67 (2/75)	5.33 (4/75)	0.174	0.677
abortion rate	5.33 (4/75)	9.33 (7/75)	0.883	0.347

4 Discussion

Generally, conception refers to the successful combination of eggs and sperm, with the formation of fertilized eggs, thus it is essential to have sex at the appropriate time. Nevertheless, Lorenz[1-3] found that regular sex in the menstrual cycle, even not during the ovulation period, might increase the chances of conception, the mechanism of which could be that shifts in immunity response caused by sexual activity were conducive to reproductive activities. Accordingly, our researchers explored the role of maintaining the integrity of sexual response cycle in the improvements of ovulation rate and pregnancy outcomes, whose findings indicated that the incidence rate of FSD in experimental group decreased after intervention, with the sexual satisfaction degree, periodic ovulation rate, clinical pregnancy rate, and live birth rate higher than those in the control group, while the biochemical pregnancy rate and abortion rate had no differences.

As is known to all, ovulation function, the quality of ovum, the quality and number of sperm and the patency status of female genital tract are closely related to the success rate of pregnancy [13]. Although it is generally accepted that ovulation occurs spontaneously in female human, which is different from animals such as the rabbits, whose ovulation rely on mating, whether ovulation can be accelerated by coitus remains controversial. For instance, some researchers maintain that a second ovulation can be induced by intercourse after spontaneous ovulation [14]. At present, many studies have reported that the timing and frequency of intercourse relative to ovulation are closely associated with pregnancy [15, 16], lacking of research on the correlation between sexual behavior guidance and ovulation or pregnancy outcomes. On the basis of previous researches, our study found that the periodic ovulation rate and clinical pregnancy rate were significantly higher than those in the control group, with the pre-pregnancy cycle of ovulation monitoring shortened. Moreover, our research revealed that, through observation of cervical mucus under the microscope, it was more likely to found centrally distributed fern-like crystals with typical morphology in the experimental group, while less or no fern-like crystals existed and the glycoprotein distributed in a reticular pattern in the control group (Figure1-2). Accordingly, we hypothesized that sexual behavior could affect the endocrine hormone levels in patients to some extent. Related studies have also suggested that sexual excitement can induce the hypothalamus to secrete

gonadotropin-releasing hormone (GnRH) [17]. Combined with the positive feedback effect of estrogen, serum luteinizing hormone (LH) level is easier to reach peak, which is a favorable condition for ovulation.

Currently, with the adjustment and improvement of the fertility policy, the rising problem of infertility is particularly prominent. Furthermore, relevant studies have shown that there is a close association between sex function and conception[18], with the incidence of FSD in infertile women reported by epidemiologic studies up to 87.1% [6], of which the occurrence is closely related to psychosocial factors[19], such as anxiety and depression, which may also lead to dysfunction of the hypothalamic-pituitary-gonadal axis (HPGA), abnormal hormone levels, menstrual disturbance and ovulation disorders. The complete sexual response cycle includes sexual desire phase, sexual arousal phase, sexual plateau phase, sexual orgasm phase and sexual resolution phase. FSD may occur due to obstacles in any phases of female sexual response cycle, while the correlation research indicates that the female sexual desire phase is the first stage as well as an essential link of the sexual response cycle, which can be triggered through touching erogenous zones [20, 21]. In addition, the relevant findings provide evidence that genital stimulation activated widespread brain regions in differential temporal patterns in the approach to, during, and after orgasm [22, 23]. However, some researchers claim that there are no definitive explanations for what triggers orgasm as yet [24]. In our study, the couples in the experimental group who received guidance on sex behavior and psychological counseling were better at overcoming negative emotions, such as anxiety and fear, and effectively stimulating sensitive areas with correct techniques, with the Bartholin's gland promoted to secrete sufficient fluid to lubricate the vagina, which is conducive to achieving sexual arousal or even reaching orgasm.

It's easy to understand that sexual dysfunction will reduce the probability of pregnancy. Furthermore, relevant studies [25, 26] have shown that, sexual excitement and orgasm are of positive significance to improve the clinical pregnancy rate, which is consistent with our findings. Its mechanism may be related to the oxytocin (OT) released by pituitary during orgasm, which has direct effects on female and male sexual behavior [27], and the intensity of orgasm is positively correlated with the concentration of OT in the peripheral blood. Additionally, it has been proved that OT is released by the posterior pituitary during male orgasm, which is supposed to participate in the ejaculatory process [28]. As a neuromodulator, OT plays a crucial role in affecting the brain's cognitive and perception of orgasm, increasing the sensitivity of brain neurons related to the contraction of pelvic floor striated muscle, and promoting the orgasmic contraction of uterine and vaginal smooth muscle to support sexual orgasm.

Although some researchers consider that the purpose of female sexuality is not necessarily to attain orgasm and the absence of orgasm in sex activity can also be regarded as a complete sexual response cycle[29]. In fact, the related changes of female genitalia during orgasm create favorable conditions for fertilization. During and after climax, the pressure of uterine cavity undergoes a dramatic change from positive to negative pressure, which is conducive to absorbing sperm into the uterus [30, 31]. In addition, the cervix open slightly and uterine body lift upward at orgasm, with the external orifice of the cervix closer to the semen pool, facilitating the entrance of sperm to the uterine cavity. Moreover, studies [32]

have reported that the blood flow of female reproductive organs elevates during excitement resulting in the increasement of cervical and vaginal secretions, with the vagina well-lubricated and the vaginal acidity buffered (the vaginal pH can rise to 7.2 and maintain for 6 ~ 8 hours after sexual intercourse[33-35]), which is also beneficial for sperm survival.

When the ovum is discharged out of the follicle, deterioration will occur in the ovulated mature oocyte if fertilization does not happen for a prolonged period in vivo or in vitro, which is called post-ovulatory aging POA . The correlation researches [36, 37] indicate that POA is closely linked to lower chance of conception, poor embryo quality and pregnancy loss. After sexual arousal, the peristalsis of fallopian tube enhances to pick up ovum released from the ovary, which creates favorable conditions for fertilization. Nevertheless, our results revealed that although the live birth rate was higher in the experimental group, there was no difference in the rates of biochemical pregnancy or miscarriage between the two groups. Consequently, we speculate that the clinical sample size should be expanded in the future so as to verify whether there is a correlation between sexual behavior and pregnancy outcomes.

In conclusion, on the basis of ovulation monitoring, the reproductive significance of maintaining the integrity of sexual response cycle is definitely apparent. Additionally, further detailed study is required to investigate the effect of sexual behavior on female endocrine and pregnancy outcomes. Notwithstanding, the clinical management of FSD is hampered to some extent by the sensitivity of sexual issues due to differences in region culture and religious beliefs. As a matter of fact, sex should not be considered as a taboo subject. Reproductive physicians should pay close attention to the sexual health of their patients so as to shorten the pre-pregnancy cycle of ovulation monitoring, improve the ovulation rate and clinical pregnancy rate, and reduce the occurrence of adverse pregnancy events.

Declarations

Ethics approval and consent to participate

The experimental protocol was established, according to the ethical guidelines of the Helsinki Declaration and was approved by the Human Ethics Committee of West China Second University Hospital. Written informed consent was obtained from individual or guardian participants.

Consent for publication

Not applicable.

Availability of data and materials

The datasets used or analysed during the current study are available from the corresponding author on reasonable request.

Competing interests

The authors declare that they have no competing interests.

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Not applicable.

Authors' contributions

Jiexin Tang conducted the experiments and drafted the main manuscript text. Danhua Lu supervised the research. Caixia Jiang and Zhilin Wang collected the data, and Yucen Xie prepared Tables 1-4. Dan Zhang supervised the research and revised the manuscript. All authors read and approved the final manuscript.

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Figures

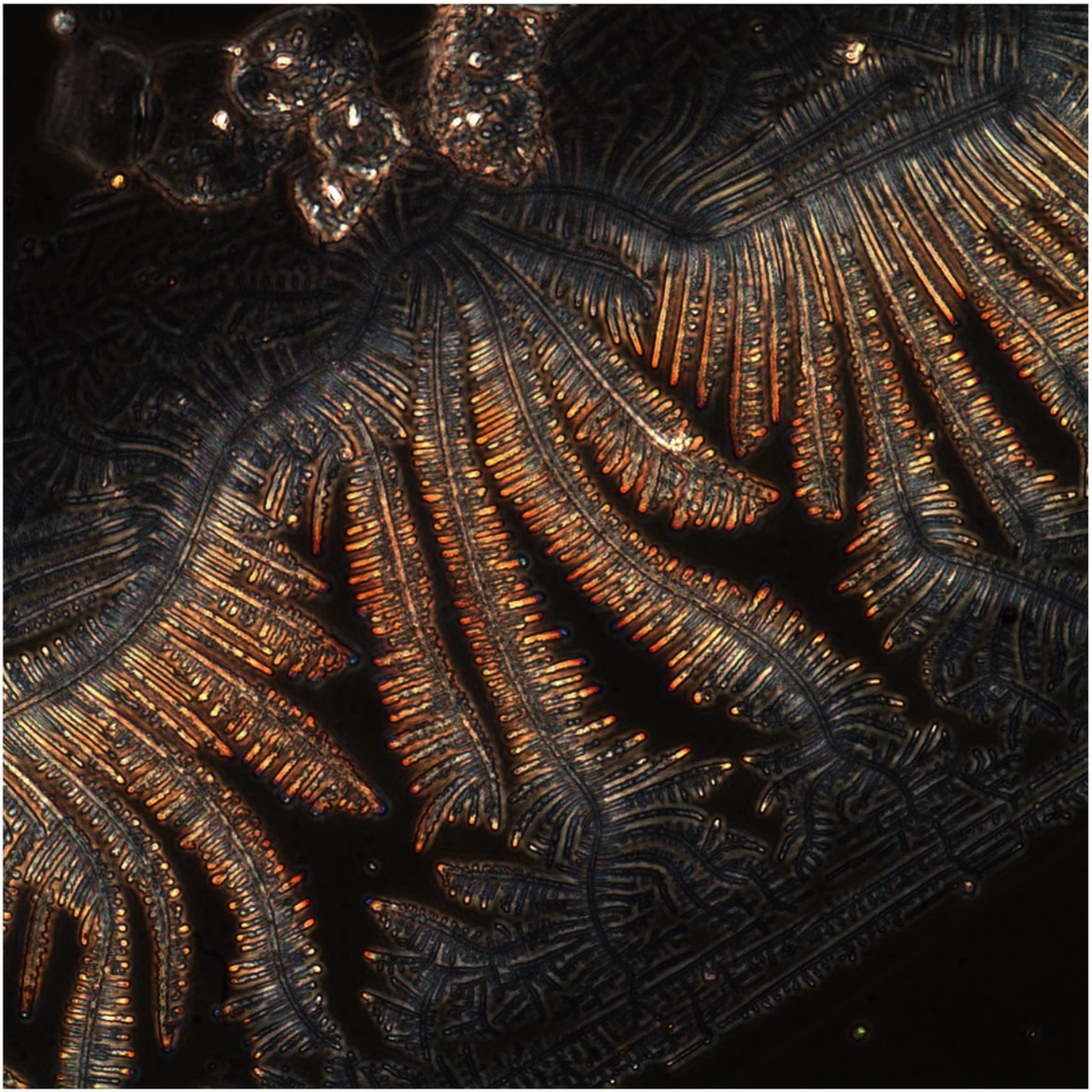

Figure 1

Cervical mucus of the experimental group under microscope. Graph showing centrally distributed fern-like crystals with typical morphology.

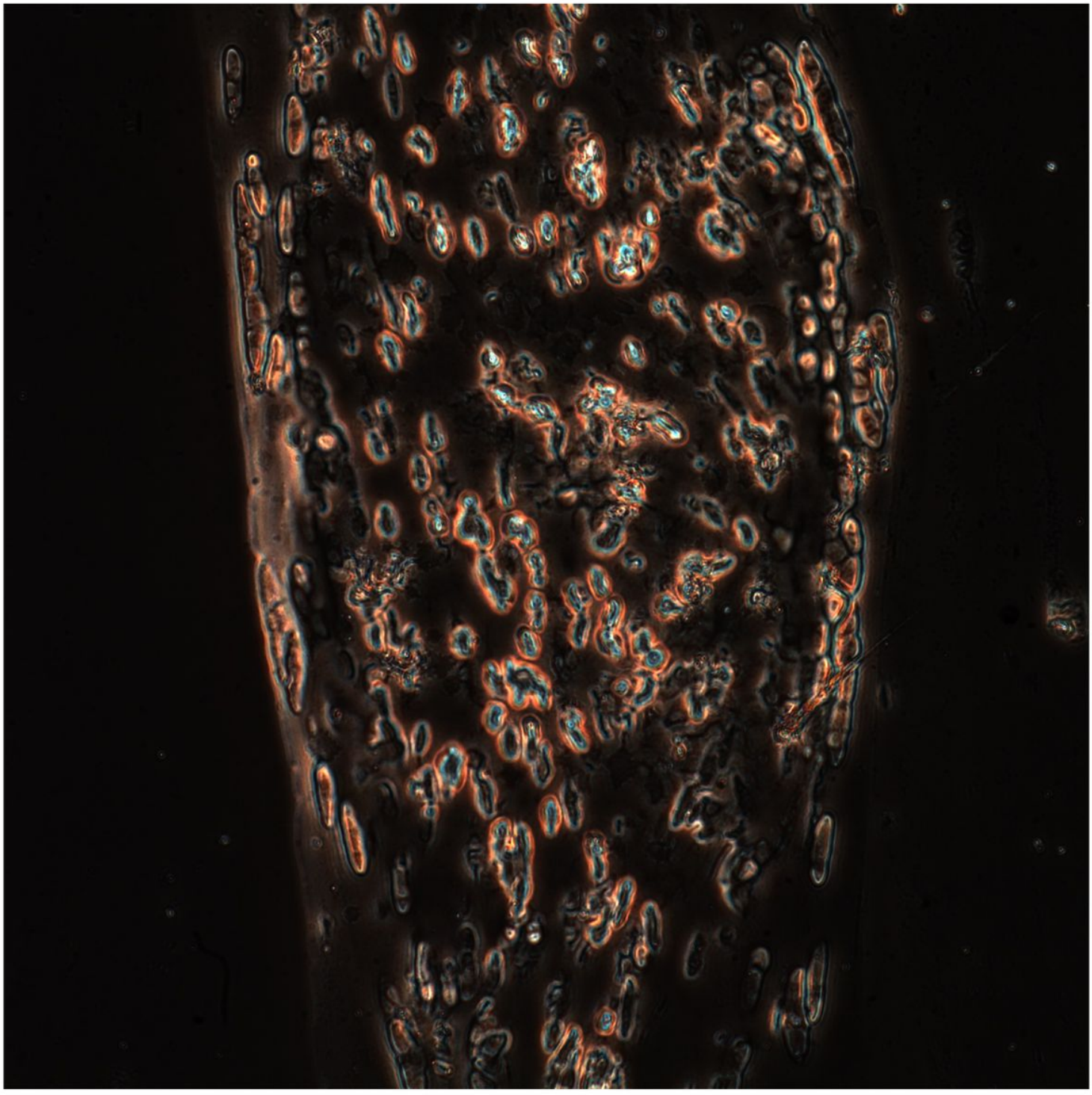

Figure 2

Cervical mucus of the control group under microscope. Graph showing distribution of glycoprotein with no fern-like crystals.