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Does the salivary fern pattern determine fertile period in reproductive female?

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ABSTRACT

Introduction: Assessment of the basal body temperature (BBT) is generally used to predict the fertile period. On the other hand, it is well documented that the fern pattern appears before ovulation due to increased estrogen level from the cervical mucus. However, the collection of cervical smear is cumbersome. Delta change in the BBT serves as ovulatory indicator whereas fern pattern has a temporal specificity from 11th to the 16th day. Since the saliva also mirrors the cervical fern pattern and easy to collect, it can be used to assess the fertile status. So the present study was done to compare the BBL with salivary smear for fern pattern to determine the fertile period.
Method: ology: The present study was carried out in 23 healthy female volunteers in the age group of 18–21 years with regular menstrual cycles of 28 days. Ethical clearance was obtained from the institute ethics committee. The basal body temperatures were measured and salivary smears were observed under low power objective (10X) for fern pattern analysis from 11th to 16th day of the menstrual cycle.
Result: On any given day from 11th to 16th day fern pattern appeared with an average of 53.7% as a fertility indicator in all the subjects. The delta change in basal body temperature varied from 0.2 to 1.4° Fahrenheit over a span of 6 days.
Conclusion: Salivary fern pattern and basal body temperature together might serve as better indicators of fertility status than anyone individually taken which is a non-invasive and cost-effective method to increase the chances of conception.

1. Introduction

Ovulation is the release of a secondary oocyte from any one of the ovaries in reproductive females during each menstrual cycle. Ovulation occurs 14 days before the onset of menstruation in a woman who has a normal duration of the menstrual cycle.[1] The ovulation is caused by LH surge which in turn elevates oestradiol in later ovulatory phase. However, it does not occur at the midpoint of the menstrual cycle when the cycle is irregular.[2] The day of ovulation is important in order to get awareness for a female either for conception or contraception.[3] It is very difficult for people to determine the day of ovulation who don't have the knowledge on their reproductive behaviour. They can follow the calendar for length and midpoint of the menstrual cycle which helps to determine the ovulation.[4] The conventional methods to evaluate the ovulation time are the estimation of plasma and urine levels of sex hormones, basal body temperature and the pattern of cervical secretions.[5–9] However, the sex hormones measurement is cumbersome and requires to collect the body fluids from the subjects. They are also expensive diagnostic protocols and interpretation requires orientation on the entire hormonal fluctuations of the menstrual cycle of that individual. Cervical sampling smear is often associated with procedural difficulties and non-compliance of participant's especially healthy young volunteers.[10,11] To overcomes the above-mentioned difficulties, it has been replaced with saliva which is conveniently available biological fluid. The blood born substances have also been detected in saliva, which serves as an essential, non-invasive medium for diagnosis of several vital parameters. In the past few decades, researchers have shown more interest in saliva for the estimation of cortisol, amylase and blood glucose due to its composition contains the multiple organic substances secreted from salivary glands.[12]

Saliva is a body fluid which has multiple advantages to use as a biological sample compared to serum. Saliva acquisition is an easy and noninvasive procedure and has higher sensitivity even with a smaller amount of sample.[13] The collection of saliva does not require special equipment and trained technicians and can be collected anywhere. The saliva processing is cost-effective and can be used to screen a large

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population. The pattern of salivary secretion is influenced by various hormones especially sex hormones which can change its composition. Since the saliva also mirrors the cervical fern pattern and is easy to collect, it can be used to assess the fertility status. The fern pattern of mucus in cervical and vaginal smears can also be seen in saliva which is a relative indicator of ovulation. There is a great demand to promote cost-effective, accurate and surrogate method for the detection of ovulation to reduce the economic burden. So the current study was aimed to determine ovulation through fern pattern observed from the smears prepared with saliva.

2. Materials and methods

2.1. Study population

The entire protocol was reviewed and approved by the Institutional ethics committee of host institution. 23 healthy volunteers aged between 18 and 21 years with no other pathology and with regular menstrual cycle were included in the study. Detailed clinical history of the participants was taken, lifestyle behaviour and pattern of menstrual cycle. The females with the irregular menstrual cycle, usage of contraceptive pills were excluded from the study. The subjects were recruited through non – random sampling technique (Convenient sampling). The subjects were medical students and other technical staff of the hospital premises who were acquainted with the female investigators. Written informed consent was obtained from all the participants in accordance with the Declaration of Helsinki (1975) after explaining the complete nature of the experiment.

2.2. Preparation of the subjects

Participants followed few specific guidelines before the data acquisition. They were directed to take normal food, to maintain good sleep pattern and to abstain centrally acting beverages like caffeine that may influence the measures carried out. All the examinations were performed in early morning hours in the Research lab, Department of physiology in Narayana Medical College, Nellore.

2.3. Basal body temperature

The volunteers were instructed to record the basal body temperature (oral temperature) in early morning hour before waking up from bed with digital thermometer provided to them. Modern digital thermometers were used since traditional glass thermometer are only 1/10th accurate.

2.4. Collection of saliva

After waking up from the bed without brushing the teeth/having breakfast/water, saliva sample was taken with the fingertip by placing it under the tongue and smeared on the glass slide provided to them. This smear was examined under low power objective 10x, (10× objective and 10x eye piece total of 100X) for fern pattern analysis. The different fern patterns were shown in Image −1.

3. Statistical analysis

Statistical analysis was carried out using graph pad prism & data was represented as mean ± SD and sensitivity. Normality of data was tested using the Kolmogorov-Smirnov test. A *p*-value of > 0.05 indicated normal Gaussian distribution. As the data sets were skewed, the Spearman correlation was done to find out associations.

4. Results

A ferning pattern was reported in all the participants during the typical preovulatory phase of the menstrual cycle. One subject could not perform smear as per the protocol. On any given day from 11th to 16th day, fern pattern appeared with an average of 53.7% sensitivity. The average delta change in BBT in 22 subjects was 0.60° F (Table – 1). The fern pattern from 11th day to 16th day had wide variations with different intensities ranging from no fern, maximum fern to disintegrated fern. (Table – 2, Graph −1). There was a weak positive correlation (r0.05), (P > 0.05) between the salivary fern pattern and basal body temperatures recorded from 11th to 16th day.

5. Discussion

The present study was aimed to introduce a novel approach for the determination of the ovulatory period in reproductive-age women. The ferning pattern identification found in preovulatory cervical mucus is a biological indicator, but relatively underutilized method due to its inconvenience of ovulation testing. The basal body temperature of the female is constant within a range throughout the menstrual cycle. There is a raise in the temperature during the ovulation which helps to indicate the fertility period. The saliva is the biological fluid which shows the typical fern pattern during the pre-ovulatory phase of the menstrual cycle. Before ovulation, there are two hormones-estrogen and ACTH that are known to stimulate the Aldosterone, which in turn helps in regulating the electrolytes and also the levels of fluid in the human body. Estrogen rushes during the fertile phase of the cycle and this causes Sodium Chloride crystals to form in the shape of fern leaves in both saliva and cervical mucus. It is the crystallization of Sodium Chloride (NaCl) that produces the ferning appearance in saliva. Therefore, saliva fern pattern is one of the best non-invasive technique to evaluate ovulation. These patterns are absent during the infertile phase of the cycles. As ovulation starts to set in, there is a transitional pattern of ferns that appear sparsely. But denser and thicker fern patterns are captured as ovulation is about to start and during. The Estrogen hormone levels correspond to the saliva patterns and the fertility stage itself. As ovulation is nearing, estrogen levels increase and cause a rise in the levels of sodium present in the body. The increasing salinity in the saliva during the ovulatory phases is a result of the changes in the chemical composition and this can be noticed while the saliva samples are allowed to dry. The crystallization patterns or ferning pattern are formed as a result of the higher levels of salt content in the dried saliva sample. The same patterns are detected in both saliva and cervical mucus samples. But as mentioned above, saliva is preferable. Since several studies were published which support the use of salivary testing for a ferning pattern as an indicator of the period of fertility in the menstrual cycle.[14–16]

The current study results suggest that saliva shows alteration in fern pattern and physiochemical changes during different phases of the menstrual cycle. This would assist to set a recommendation for the determination of fertile period in reproductive females.[17] These results favour the use of saliva fern pattern technique as non-invasive indicator of ovulation. However, further studies are warranted with higher sample size. The project will be planned to extend with the comparison of salivary fern with cervical fern and hormonal estimation.

6. Limitations

The current study has some limitations where we did not compare the salivary pattern with a cervical fern pattern. The hormonal levels also did not measure which could provide more valuable information to increase the strength of study results.

A- No fern
B- Mild Fern
C- Moderate fern
D- Maximum fern
E- Disintegrated fern
F- No fern

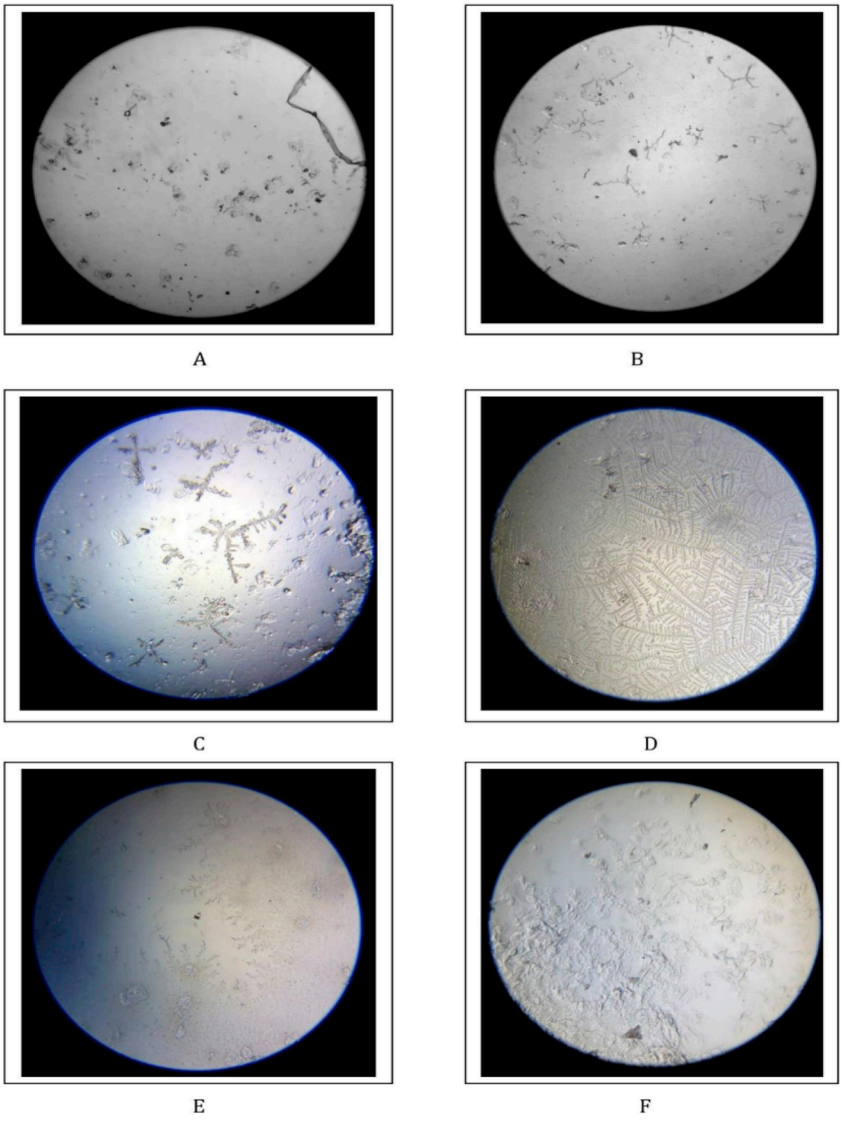


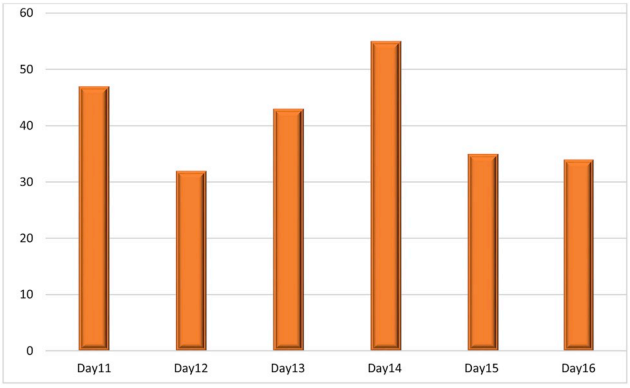
Image 1. Shows the pattern of fern from the saliva.

Table 1
Shows the salivary fern pattern and BBT from 11th to 16th day of the menstrual cycle.

S.No	Day of Menstrual cycle	Sensitivity of fern pattern	BBT Mean ± SD	CI of BBT
1	11	12 (54.54%)	97.28 ± 0.64	96.1–98.8
2	12	13 (59%)	97.20 ± 0.53	96.3–98.6
3	13	13 (59%)	97.45 ± 0.56	96.6–99
4	14	14 (63.63%)	97.57 ± 0.46	97.0–98.8
5	15	11 (50%)	97.53 ± 0.68	96.2–99
6	16	10 (45.45%)	97.51 ± 0.72	96.2–99

Table 2
Shows the cumulative frequency of different types of fern pattern.

Fern Pattern	Frequency	Per cent
Not Available	22	16.1
Full fern	30	21.9
Mild fern	42	30.7
No fern	33	24.1
Disintegrated fern	10	7.3
Total	**137**	**100**



Graph 1. Shows the frequency distribution of the fern pattern.

7. Conclusion

Salivary fern pattern and basal body temperature together might be a better indicator of fertility status than anyone individually taken. It can be done at home with a handheld microscope. It is a non-invasive and cost-effective method. However, it cannot be used for contraception due to low sensitivity.

Author contribution

Sruthi Priya B: Data collection.

Pushpaja M: Data collection.

A.V. Siva kumar: Conceptualization of study design and manuscript preparation.

K. N Maruthy: Guided whole project including manuscript preparation.

Ethical consideration

The current study protocol was reviewed and approved by Institutional Ethics Committee, Narayana Medical College, and Nellore.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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References

1. Guida M, Barbato M, Bruno P, Lauro G, Lampariello C. Salivary fern ing and the menstrual cycle in women. *Clin Exp Obstet Gynecol.* 1993;20(1):48–54.
2. Svanes C, Real FG, Gislason T, et al. Association of asthma and hay fever with irregular menstruation. *Thorax.* 2005 Jun 1;60(6):445–450.
3. Farris EJ. A test for determining the time of ovulation and conception in women. *Am J Obstet Gynecol.* 1946 Jul 1;52(1):14–27.
4. Venturoli S, Porcu E, Fabbri R, et al. Menstrual irregularities in adolescents: hormonal pattern and ovarian morphology. *Horm Res Paediatr.* 1986;24(4):269–279.
5. Guida M, Tommaselli GA, Palomba S, et al. Efficacy of methods for determining ovulation in a natural family planning program. *Fertil Steril.* 1999 Nov 1;72(5):900–904.
6. Schiphorst LE, Collins WP, Royston JP. An estrogen test to determine the times of potential fertility in women. *Fertil Steril.* 1985 Sep 1;44(3):328–334.
7. Farris EJ. A test for determining the time of ovulation and conception in women. *Am J Obstet Gynecol.* 1946 Jul 1;52(1):14–27.
8. Collins WP. Hormonal indices of ovulation and the fertile period. *Adv Contracept.* 1985 Dec 1;1(4):279–294.
9. Roney JR, Simmons ZL. Hormonal predictors of sexual motivation in natural menstrual cycles. *Horm Behav.* 2013 Apr 1;63(4):636–645.
10. McSweeney DJ, Sbarra AJ. A new cervical mucus test for hormone appraisal. *Am J Obstet Gynecol.* 1964 Mar 15;88(6):705–709.
11. MacDonald RR. Cyclic changes in cervical mucus. *BJOG An Int J Obstet Gynaecol.* 1969 Dec 1;76(12):1090–1099.
12. Pullaganti M, Kumar SA, Maruthy KN, Gurja JP, Chintala KK. Association of stress with heart rate variability in different phases of the menstrual cycle. *Natl J Physiol Pharm Pharmacol.* 2019;9(3):256–260.
13. Beerda B, Schilder MB, Janssen NS, Mol JA. The use of saliva cortisol, urinary cortisol, and catecholamine measurements for a noninvasive assessment of stress responses in dogs. *Horm Behav.* 1996 Sep 1;30(3):272–279.
14. Melnick H, Goudas VT. The detection of a salivary ferning pattern using the knowhen ovulation monitoring system as an indication of ovulation. *J Women's Health Care.* 2015;4(235) 2167-0420.
15. Biel Casals JM. Descripcion de un nuevo test de ovulacion y analisis de sus resultados. *Med Clínica.* 1968;50:385–392.
16. Braat DD, Smeenk JM, Manger AP, Thomas CM, Veersema S, Merkus JM. Saliva test as an ovulation predictor. *Lancet.* 1998 Oct 17;352(9136):1283–1284.
17. Ersyari RM, Wihardja R, Dardjan M. Determination of ovulation in women using saliva ferning test. *Padjadjaran J Dent.* 2014 Nov 30;26(3).