Analysis of corporeal characteristics of cervico-vaginal mucus in cows

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Abstract
Present study was conducted among 40 selected cows with an aim to investigate the corporeal characteristics of cervico-vaginal mucus (CVM) in relation to fertility in cows. The CVM samples were collected aseptically on the day of estrus prior to the artificial insemination. Physical characteristics of CVM were studied immediately after collection of cervical mucus. Study reported that per cent of fertile and infertile estrus cows were 57.5 and 42.5, respectively. The physical characteristics of CVM viz., clear colour, thin consistency, typical fern pattern and good spinnbarkeit value (≥12.00 cm) favored the higher conception rate in cows. The alkaline pH (≥8.30) of CVM has favorable effect on the conception rate as compared to less alkaline pH (<7.76). The study concluded that the corporeal characteristics in CVM are helpful to detect the fertility in cows.

Keywords: Cervico-vaginal mucus, corporeal characteristics, fertility, fern pattern, spinnbarkeit value, pH

Introduction
Fertility and breeding efficiency of dairy animals play a vital role in dairy economics. Fertility is one of the key determinants of the lifetime performance of the cow. Cervical mucus quality gives good indication of physiological state of cow during estrus and good quality of mucus results in high conception rates. The physical properties of cervico-vaginal mucus (CVM) have direct relationship with circulatory estrogen-progesterone levels and fertility status of the animals (Rangnekar et al., 2002) as it essentially undergoes certain changes during estrus phase for the passage of spermatozoa.

Cervical mucus is essential to sperm survival and transport. Without the fertile mucus, the sperm would only last for hours in the vagina with minimal chance of mating and fertilization of the ovum (Dunson et al., 1999). Cervical mucus discharge is a mechanical barrier against pathogen of the uterus. The cervical mucus is a hydrogel, which consists of water and a solid component composed of three or more units forming a three dimensional network. The amount and consistency of secretions from the cervix of the cow vary with the stage of the estrous cycle. Cervical mucus is not a fluid in the physical sense and shows unique viscous behavior. Specifically, cervical mucus is watery and has minimal viscosity at the time of ovulation in cattle (Hamana et al., 1971). The important physical characteristics of cervical mucus are colour, consistency, pH, spinnbarkeit and fern pattern are considered as effective laboratory tools to predict the infertility in cattle as they have pronounced effect on the fertilizing capacity of the spermatozoa (Rangnekar et al., 2002). Considering the above facts present study was aimed to study the relationship of corporeal characteristics of cervico-vaginal mucus (CVM) with fertility in cows.

Materials and Methods
Healthy cows free from gross reproductive disorders with regular estrous cycle were selected for the present study. The experimental cows were selected from Bull Mother Experimental Farm (BMEF) and nearby dairy farms. A total 40 animals were selected for the present study. Cows were detected in estrus by using teaser bull, visual observation for signs of estrus. Experimental cows in estrus were confirmed by per-rectal examination and Artificial Insemination was performed by recto-vaginal method by using good quality frozen-thawed semen.
Collection of cervico-vaginal mucus (CVM)
The CVM samples were collected aseptically on the day of estrus prior to the Artificial Insemination by “Pipette and Syringe Method” (Reddy 1974; Panangala et al., 1978) [12, 9]. By inserting broad end of dry and sterilized glass pipette (10 ml capacity) in the anterior vagina and the pointed end of which was connected to a syringe (50 ml capacity) with rubber junction. Physical characteristics of cervical mucus were study immediately after collection of cervical mucus, and remaining sample was transferred into clean, dry and sterilized vials and they were kept at -20°C till the mucus samples were analyzed for biochemical parameters.

Evaluation of Physical Properties of CVM
Following physical properties of estrual mucus were studied immediately after collection of cervical mucus.

Colour
Colour of the CVM was judged by direct examination after collection and classified as per the method described by Deo and Roy (1971) [2]. The classification consisted of (a) Clear and stringy (white of an egg), (b) Turbid (cloudy in appearance) and (c) Dirty (yellowish, grey, red etc.) mucus.

Consistency
The consistency was studied to know the degree of thickness of cervical mucus. A few drops of mucus sample were placed into a grease free glass slide and slide was inclined to 45 for the detection of consistency of the CVM. By observing the movement, the mucus sample was categorized into thin and thick (Deo and Roy 1971) [3].

Fern Pattern (Crystallization or Arborization)
A drop of CVM was smeared on clean, dry and grease free slide. It was air dried and then examined under low power (10x) and high power (40x) microscope for characteristic fern pattern. The types of fern patterns were divided into two type viz., typical, atypical and nil as described by Luktuke and Roy (1967) [1].

Spinnbarkeit
For measuring the Spinnbarkeit value, 2-3 drops of collected mucus sample was taken on a grease free glass slide; then another grease free glass slide was placed over it. The mucus was stretched between two slides by moving the second slide away from the first one, until the mucus breaks. The distance between the two slides just before the breakage of the mucus string was measured through a scale (cm scale). Three to four consecutive readings were taken for each sample and the mean reading was considered as “spinnbarkeit value” of the sample (Panigrahi, 1964) [10].

pH
The pH was measured with the help of pH paper in the range of 0.0 to 14.0. The pH paper was dipped into mucus collected in the bottle; change in the color of pH paper was compared with the standard color of the pH paper strip.

Pregnancy diagnosis
Pregnancy was confirmed by rectal palpation after 60 days of AI in non-returned cows and animals were divided into two subgroups, conceived and non-conceived on the basis of pregnancy diagnosis. Physical characteristics of cervical mucus were compared in conceived and non-conceived animals.

Results and Discussion
Present study revealed that majority 57.5 per cent of cows were found fertile and remaining 42.50 per cent were infertile. The various physical characteristics of estrual cervical mucus were studied in respect to fertility among them.

Colour
Table 1 revealed that per cent incidences of clear, turbid and dirty colour of CVM samples were found to be 86.95, 13.04 and 0, respectively, in conceived cows and the corresponding values in non-conceived cows were found to be 41.17, 35.29 and 23.52 per cent, respectively, with the difference obtained to be highly significant (P<0.05). A considerably higher conception rate (86.95%) was found in cows having clear CVM as compared to those having turbid CVM (13.04%), suggestive of clear CVM providing favorable condition for good conception rate. The present findings are in agreement with the reports of Dev et al. (1997) [3], who reported that clean estrual mucus is conducive for sperm penetration and conception, whereas, turbidity arrests the sperm motility in estrual mucus. Deo and Roy (1971) [2] also opined that the normal cows with clean or turbid and less elastic mucus showing typical fern pattern favoured the conception.

Consistency
The per cent incidences of thin and thick CVM samples were found to be 87.00 and 13.00, respectively, in conceived cows and the corresponding values in non-conceived cows were found to be 58.8 and 41.2 per cent, respectively, with the difference found to be significant (P<0.05) (Table 1). The incidence of thin CVM in conceived cows was found to be comparatively higher as compared to the per cent incidences reported by Rangnekar et al. (2002) [11]; Selvraj et al. (2002) [13], and Modi (2007) [8]. A considerably higher conception rate was found in cows having thin CVM as compared to those having thick CVM, which was suggestive of the fact that the thin CVM contributed positively for good conception rate, whereas, the occurrence of thick CVM was higher in non-conceived cows than conceived cows (41.2% Vs 13.00%).

Fern patterns
Fern pattern of cervical mucus is associated with the changes in the physio-chemical properties of cervical mucus. The per cent incidences of typical and atypical patterns of CVM samples were found to be 82.6 and 17.4, respectively, in conceived cows and the corresponding values in non-conceived cows were found to be 47.1 and 52.9 per cent, respectively, with the difference being significant (P<0.05) (Table 1). A considerably higher conception rate (82.6%) was found in cows having typical fern patterns as compared to those having atypical fern patterns (17.4%), which was suggestive of the fact that typical fern patterns of CVM contributing favorable condition for good conception rate. The present findings are in agreement with the reports of Galhotra et al. (1971) [5], who reported that the typical fern pattern of mucus crystals were indicative of marked ovulatory estrus and animals in such estrus were suitable for service or insemination. Sharma et al. (2008) [14] opined that estrual cervical mucus with classical arborization pattern due to increased salt and organic constituents favoured sperm survival and transport thereby improve the conception rate.
Spinnbarkeit
The mean and range of spinnbarkeit values in CVM of conceived and non-conceived cows were found to be 13.09±0.40 (Range: 9.5-16.5) and 11.41±0.31 (Range: 8.5-13.9) cm, respectively, with the difference being highly significant (P<0.01) (Table 1). The mean spinnbarkeit value (13.09±0.40 cm) in CVM of conceived cows was found to be higher than the values reported by Bennur et al. (2004) [1], 7.38±0.56 cm in cows and Sharma et al. (2008) [14], 10.30±0.93 cm in buffaloes. Spinnbarkeit reaches a maximum value immediately before or during the ovulatory phase. Under the influence of estrogens, cervical mucus becomes abundant, clear and stretchable and somewhat like egg white. Only such mucus appears to be able to be penetrated by sperm. After ovulation, the character of cervical mucus changes and under the influence of progesterone it becomes thick, scant, and tacky.

pH
The mean pH values in CVM of conceived and non-conceived cows were found to be 8.30±0.11 and 7.76±0.12, respectively, with the difference being highly significant (P<0.01) (Table 1). The mean pH value (8.30±0.11) of CVM for conceived cows was found to be very similar to those reported by Vadodaria (1987), 8.36±0.00 in buffaloes; Bennur et al. (2004) [1], 8.13±0.07 in cows and Modi (2007) [8], 8.39±0.17 in Kankrej cows. These observations are suggestive of the fact that alkaline pH of CVM contributed positively for good conception rate, as opined by Rangnekar et al. (2002) [11], who reported that the alkaline pH of cervico-vaginal mucus was more favourable for the progressive motility of spermatozoa and also for the suitable uterine environment.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Conceived</th>
<th>Non-conceived</th>
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<tbody>
<tr>
<td><strong>Colour</strong></td>
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<tr>
<td>Clear</td>
<td>86.95**</td>
<td>41.17</td>
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<td></td>
<td>(20)</td>
<td>(7)</td>
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<tr>
<td>Turbid</td>
<td>13.04</td>
<td>35.29</td>
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<td></td>
<td>(3)</td>
<td>(6)</td>
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<tr>
<td>Dirty</td>
<td>87.00*</td>
<td>58.8</td>
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<td></td>
<td>(20)</td>
<td>(10)</td>
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<td></td>
<td>13.00</td>
<td>41.2</td>
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<tr>
<td><strong>Consistency</strong></td>
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<td></td>
<td><strong>Typical</strong></td>
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<td></td>
<td>82.6 *</td>
<td>47.1</td>
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<td>(19)</td>
<td>(8)</td>
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<tr>
<td>Atypical</td>
<td>17.4</td>
<td>52.9</td>
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<td></td>
<td>(4)</td>
<td>(9)</td>
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<tr>
<td><strong>Fern pattern</strong></td>
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<tr>
<td></td>
<td><strong>Typical</strong></td>
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<td></td>
<td>13.90±0.40*</td>
<td>11.41±0.31</td>
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<td>(20)</td>
<td>(7)</td>
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<tr>
<td><strong>Spinnbarkeit (SBK)Value (cm)</strong></td>
<td><strong>Mean±SE</strong></td>
<td><strong>Range</strong></td>
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<td></td>
<td>8.30±0.11**</td>
<td>7.76±0.12</td>
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</tbody>
</table>

Figures in parentheses indicate the number of animals
*Significant difference (P<0.05) between conceived and non-conceived group
**Significant difference (P<0.01) between conceived and non-conceived group

Conclusion
From the present study concluded that the corporeal characteristics of CVM that is colour, consistency, pH, spinnbarkeit and fern pattern are considered as effective laboratory tools to predict the infertility in cows. The physical characteristics of CVM viz., clear colour, thin consistency, typical fern pattern, good spinnbarkeit value (≥12.00 cm) and alkaline pH favored

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References
11. Rangnekar MN, Dhoble RL, Gacche MG, Inganwale MV, Sawale AG, Jadhav JM. Physical properties of estrual cervical mucus in repeat breeding crossbreed (HF)


