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Original Research Article

## Seasonal variation in semen quality: effect of temperature: a study in North India

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### ABSTRACT

**Background:** Variable temperature in different seasons may affect the quality of semen. Most of the studies are from west barring a single large Chinese study with variable results; however, no such study focusing on temperature alone has been published from Indian subcontinent. Present study was undertaken to observe any seasonal variation affecting the semen quality at different times of year.

**Methods:** The study was conducted retrospectively over one year period (2014-2015) at a tertiary care hospital in North India. 815 semen samples referred from infertility clinic were analysed for Sperm concentration, functional sperm concentration, motile sperm concentration and sperm motility index by an automated semen analyser system (SQAIIC-P, Medical Electronic Systems, Los Angeles, CA, USA); and morphological assessment by Papanicolaou stained smears. Statistical analysis was done using SPSS 24 software (SPSS Inc, Chicago, IL, USA). The ANOVA test was used to assess differences in semen parameters,  $p < 0.05$  was considered as statistically significant.

**Results:** Depending on the average highest temperature (AHT) in different seasons, samples were categorized in four groups: winter AHT  $< 10^{\circ}\text{C}$ , spring AHT  $< 20^{\circ}\text{C}$ , autumn/rainy season AHT  $20\text{-}30^{\circ}\text{C}$  and summer AHT  $> 30^{\circ}\text{C}$ . There was significant variation on different parameters e.g. sperm count, motility, FSC, MSC, SMI, abnormal sperms and morphological defects when compared in different groups using ANOVA test ( $p < 0.05$ ).

**Conclusions:** Present study observed that AHT of  $20\text{-}30^{\circ}\text{C}$  (autumn) has a favourable effect on semen quality; however, larger and continuous data in the form of longitudinal study is needed for better correlation.

**Keywords:** Semen quality, Male infertility, Temperature

### INTRODUCTION

Infertility is a global public health concern. It is defined as the inability of a reproductive age couple cohabiting together to conceive after one year of unprotected intercourse. Approximately 15% of couples are infertile worldwide and, around 20-70% of these are attributed to male infertility factors.<sup>1,2</sup> The extensive published data suggest that semen quality is declining globally.<sup>3,4</sup> Of the multitude of variables studied so far, temperature is most important yet least studied parameter. The present study aimed to observe the effect of temperature on semen quality in different seasons in a year.

### METHODS

This retrospective study was conducted over the period of one year (July 2014 to June 2015) at a tertiary care hospital of North India. 815 semen samples referred from the infertility clinic were analysed. The donors ranged in age from 20 to 52 years, sexual abstinence of 3-7 days was followed. The samples were collected in the laboratory and processed within 30 minutes of collection as per the standard protocol. Depending on the average highest temperature (AHT) in different seasons, the samples were categorized in four groups: winter AHT

<10°C, spring AHT <20°C, autumn/rainy season AHT 20-30°C and summer AHT >30°C (Table1).

**Table 1: Distribution of semen samples under various groups as per AHT.**

Groups	AHT*_(°C)	N	Months
Winter	≤10	200	November-January
Spring	10.1-20.0	186	February-April
Autumn/ rainy	20.1-30	197	August-October
Summer	>30	232	May-July



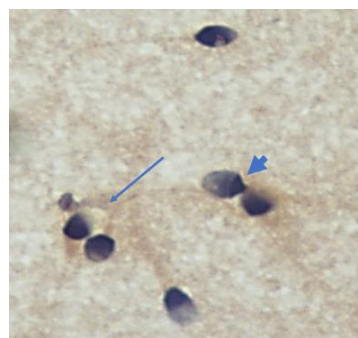
**Figure 1: Automated sperm analyser, Sysmex-SQIIC.**

For each sample, sperm concentration (SC), motility (SM), functional sperm concentration (FSC), motile sperm concentration (MSC) and sperm motility index (SMI) were conducted as per WHO guideline 2010<sup>5</sup> on SQAIIIC-P (Medical electronic systems, Los Angeles, CA, USA). Sperm morphology was assessed on Papanicolaou stained (Pap) smear after centrifugation of the remaining sample; two smears were prepared from each sample and a minimum of 200 spermatozoa were counted. In every sample, number of sperms with normal morphology (NM), head defect (HD), midpiece defect (MPD), cytoplasmic defect (CD) and tail defect (TD) were recorded following Tygerberg Kruger strict morphological criteria. Results were expressed as mean, median and standard deviation. Various groups were created as per season, as per average highest temperature. Statistical analysis was done using SPSS 24 software (SPSS Inc, Chicago, IL, USA). The ANOVA test was used to assess differences in semen parameters, p<0.05 was considered as statistically significant.

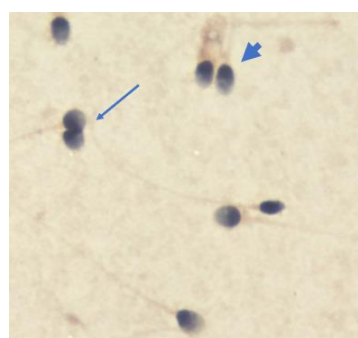
**RESULTS**

Donors ranged in age from 20-52 years (median age-28 years). Volume of semen ranged from 0.5- 2.5mL. Sperm concentration ranged from 1-189 million/ml, with a mean of 37.48±32.40 million/ml. The highest SC was observed

in autumn, followed by summer and spring (AHT >10°C), while winter season showed lowest (AHT<10°C). Percentage of SM ranged from 1-83 (mean 32.26±15.67). Lowest SM were found in winter (AHT<10°C) and maximum in autumn followed by summer and spring (AHT >10°C). A similar pattern was noted in FSC, MSC & SMI with highest concentrations in autumn followed by summer, spring, and winter. A significant effect of AHT variation was seen on all semen parameters (p=0.000). SC and SM were significantly lower in winter (AHT <10°C) than in other seasons (p=0.000). The mean of sperms with NM was lowest in spring (AHT=10.1-20°C) and highest in autumn (AHT=20.1-30°C) followed by summer (AHT >30°C). The mean HDs decreased significantly in autumn and winter and, increased significantly in spring followed by summer (p=0.000). The mean of MPD was maximum in summer and winter while it was lowest in autumn (p=0.000). The lowest mean of TD was observed in autumn and, highest in winter (p=0.000). CD were maximum in winter and minimum in summer (p=0.000) (Table 2-4).



**Figure 2: Abnormal sperms showing tapered head (arrowhead), amorphous head (arrow), Papanicolaou stain, X1000.**



**Figure 3: Abnormal sperms showing small round double heads (arrow), mid piece defect (arrowhead), Papanicolaou stain, X1000.**

**DISCUSSION**

The initial evaluation in a case of male factor infertility relies on semen analysis and various parameters are assessed on semen analysis i.e. volume, SC, SM

(progressive & non progressive), sperm morphology, pH and SMI. along with newer automated parameters like FSC, MSC

**Table 2: Age distribution with semen parameters.**

Semen parameter	Mean	(SD)	Minimum	Maximum
<b>Age</b>	29.73	5.77	20	52
<b>Concentration* (sperm/ml)</b>	37.48	32.40	1	189
<b>Motility (%)</b>	32.26	15.67	1	83
<b>Normal morphology† (%)</b>	76.37	35.88	2	198

\*Number of sperm X 10<sup>6</sup>, †Tygerberg Kruger strict morphology<sup>16</sup>

**Table 3: Seasonal variations of semen parameters.**

Semen parameter		Winter‡	Spring‡	Summer‡	Autumn/rainy‡	F value	P value
<b>Sperm concentration* (sperm/ml)</b>	Median	22.5	24	27	36	16.243	0.000
	Mean	29.82	31.91	37.80	50.13		
	SD	23.21	26.05	32.33	41.13		
<b>Motility (%)</b>	Median	27	30	32	36	10.156	0.000
	Mean	29.04	29.82	33.01	36.94		
	SD	13.20	14	15.87	17.95		
<b>FSC</b>	Median	2.1	2.35	2.8	4.7	17.876	0.000
	Mean	5.64	6.80	9.54	16.91		
	SD	8.85	11	15.56	23.34		
<b>MSC</b>	Median	5.7	6.7	8.3	12.9	19.003	0.000
	Mean	11.11	12.67	17.01	26.25		
	SD	13.82	11	22.01	30.49		
<b>SMI</b>	Median	65	70	76.5	95	16.100	0.000
	Mean	84.94	92.08	105.76	138.19		
	SD	60.15	67.96	82.22	106.79		

\*Number of sperm X 10<sup>6</sup>, ‡ number of subjects for sperm concentration, motility, FSC, MSC, SMI (winter=200, spring=186, summer=232, autumn/rainy=197)

**Table 4: Seasonal variations in sperm morphology (Papanicolaou smears on 200 spermatozoa count).**

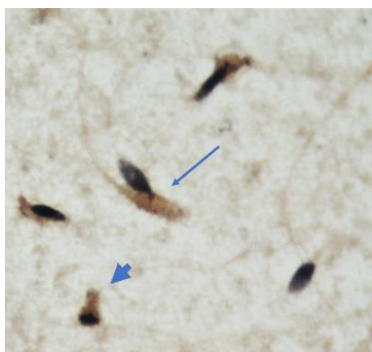
Morphology*		Winter‡	Spring‡	Summer‡	Autumn/rainy‡	F value	P value
<b>Normal</b>	Median	70	67	64.5	104	63.962	0.000
	Mean	67.87	64.97	69.48	103.86		
	SD	31.49	25.51	37.73	32.17		
<b>Head defect</b>	Median	102.5	124.5	128.5	70	104.555	0.000
	Mean	104.8	125.67	123.73	74.82		
	SD	32.10	27.05	38.42	29.04		
<b>Midpiece defect</b>	Median	27	15	33.5	12	43.283	0.000
	Mean	29.07	21.40	38.67	16.17		
	SD	19.81	19.77	26.10	14.22		
<b>Tail defect</b>	Median	24	16	19.5	11	15.959	0.000
	Mean	29.35	21.51	25.07	14.76		
	SD	21.63	19.31	20.17	12.87		
<b>Cytoplasmic defect</b>	Median	70.5	50	5	30	131.877	0.000
	Mean	70.83	54.52	12.81	40.06		
	SD	36.34	26.64	16.08	26.51		

\*Tygerberg Kruger strict criteria, ‡number of subjects for normal morphology, head defect, midpiece defect, tail defect, cytoplasmic defect (winter=200, spring=186, summer=232, autumn/rainy=197).

Seasonal variation in semen parameters has been studied in the past with controversial results;<sup>6-10</sup> some observed significant variations in parameters while others showed

no effect of seasonal variation on semen quality whatsoever.<sup>9,11</sup> To the best of literature search no such study has been published from Indian subcontinent.

The present study observed a significant effect of seasonal variation on all semen parameters.



**Figure 4: Abnormal sperms showing tapered head and bent neck (arrow), mid piece defect and absent tail (arrowhead), Papanicolaou stain, X1000.**

The lowest SC was noticed in winter (AHT <10°C), and the highest in Autumn (AHT 20.1-30°C). The correlation of SC in both these seasons was highly significant (p=0.000); however, Carlsen et al and Chen et al reported no seasonal change in SC.<sup>9,12</sup> SM also showed similar variation with minimum in winter and maximum in autumn (p=0.006); these observations were different from earlier reports by Yogev et al and Chen et al that showed that SM did not change with seasonal variations.<sup>8,12</sup> Automated semen parameters (FSC, MSC and SMI) have not been previously studied earlier. In the present study highest levels of functional and motile sperms were observed in autumn and lowest in winter with a significantly high p value (p=0.000) in each. Mature sperm/spermatozoon is an actively motile, highly specialized, free swimming cell; with various parts (head, neck, middle piece and a principal piece or tail. An axial filament passes through the middle piece and extends into the tail. Mature sperm measures approximately 60 µm in length.<sup>13</sup> Therefore any variations from normal will make spermatozoa abnormal morphologically and or functionally, thus it is imperative to look into morphology as well in investigating a case of male infertility. In the present study, sperms with NM were maximum in autumn followed by spring, winter and lowest in summer (p=0.000). HD, MPD and TD were significantly lower in autumn while CD were lowest in summer followed by autumn (p=0.000). HD were maximum in spring (AHT: 10.1-20°C) (p=0.000); MPD in summer (AHT>30°C) (p=0.000) while CD and TD were maximum in winter (AHT<10°C) with p=0.000. Changes in testicular temperature could impact synthesis and repair of DNA in sperm head which can be the reason for the seasonal variation of HD.<sup>14</sup> A positive correlation between HD and DNA abnormality was observed by Varghese et al.<sup>15</sup>

#### Limitations

Due to smaller sample size and absence of follow up of same patients in different seasons, more elaborate and

consistent longitudinal study is warranted to further improve the knowledge about the effects of AHT on semen parameters.

#### CONCLUSION

The present study observed significant variations on sperm quality under different temperature and; autumn season was the best (AHT-20.1-30°C) with all parameters including calculated values through analyzer (SC, SM, FSC, MSC, SMI) and morphological parameters on Pap smears (NM, HD, MPD, TD). However, CD did not show any consistent pattern with changing temperature.

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*Ethical approval: The study was approved by the Institutional Ethics Committee*

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