

Calcium and magnesium in male reproductive system and in its secretion.

Part II* within-subject variability in human seminal plasma and spermatozoa

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ABSTRACT: *This study was undertaken to find out what, if any, within-subject variability exists in semen parameters, and calcium and magnesium levels in seminal plasma and spermatozoa. Significant changes ($p < 0.05$) were found in semen volume and in the percentage of sluggish motile spermatozoa. No other significant changes were seen in semen parameters or in calcium or magnesium in spermatozoa and seminal plasma. (Urologia 2008; 75: 94-6)*

KEY WORDS: *Abstinence, Calcium, Magnesium, Semen, Seminal plasma, Variability*

PAROLE CHIAVE: *Calcio, Magnesio, Sperma, Variabilità*

Introduction

The variability of different substances present in fluids and tissues of the human body is well known. Therefore, values are given in ranges. Example, the normal value for blood glucose in adults is 80-120 mg/dL (1). This might also be applicable in the case of human semen and its composition. The present study was designed to evaluate the within-subject variability of seminal parameters and of the level of calcium and magnesium in seminal plasma and spermatozoa.

Materials and Methods

A total of 24 clinically healthy adults (age group: 20-28 years) participated in this study. They submitted their semen samples on 2 different occasions by following the instructions given below:

i. Participants were to practice abstinence for 3 to 5 days prior to semen collection. This time is sufficient to bring all major parameters and chemical substances of semen back to normal range (2).

ii. The sample was to be submitted to the laboratory immediately after collection. This was essential to enable us to establish the liquefaction time of the sample.

iii. Collection was to be done by masturbation. This method enables collection of the full sample without any loss. Other methods of collection were found to yield differences in the value of semen composition (3). During collection by means of coitus interruptus chances were high that a portion of semen would be lost and also that it would be mixed with vaginal cells and secretions.

iv. Well-cleaned, wide-mouthed containers were provided for sample collection. Cleanliness is important for preserving sperm motility. Wide-mouth containers

helped to avoid the loss of any portion of the ejaculation. Each portion of sample was important as the seminal plasma, as well as the semen chemical parameters, is not constant in all portions of sample (4, 5). In the present study whole semen was analyzed.

v. Collection was to be completed between 8:00 and 10:00 am. This was to exclude the effect of time on the study parameters. We have seen difference existing in seminal parameters when the collection was carried out at different times during the day (Valsa, Skandhan, Amtih, Avni 2007).

Semen evaluation was done according to WHO criteria (6). Semen was centrifuged (2.000 rpm * 20 minutes) to separate spermatozoa into pellet. Calcium was estimated by the Clark-Collip method; magnesium was determined employing the colorimetric method with titan yellow (7).

Results

The results of the present study are given in Table I.

Discussion

Eliasson (8) reported the results of 3 samples collected in 3 different weeks, which allowed consideration to be given to any individual variations. The present study was designed to clarify the details of within-subject variability of semen parameters and levels of calcium and magnesium in seminal plasma and spermatozoa.

All factors such as abstinence time prior to semen collection, collection time, collection method and the container used for collection allowed us to manage any changes which could otherwise be present among the study parameters (2).

Table I shows significant changes ($p < 0.05$) in semen volume and percentage of sluggish motile spermatozoa between samples I and II. In earlier studies, Heychel et al (9) and Schwartz et al (10) reported an increase in semen volume by permitting their patients to maintain abstinence for 1-5 days before collection. They reported a highly significant correlation ($r = 0.41$, $p < 0.001$) between the volume and length of abstinence. Mortimer et al (11) observed that the ejaculate volume increased by 1.0 mL between the first and second day, and by approximately 3 ml/day thereafter until day 5. The present study (Tab. I) clearly shows that there was a variability in volume ($p < 0.05$). Mallidis et al (12) also observed within-subject variations of semen volume.

Skandhan and Mazumdar (13) reported total sperm count and percentage of sperm motility as the 2 main parameters in semen evaluation. We did not find any significant differences in the values of sperm count (Tab. I). This study showed that the sperm count remained at a steady level if abstinence could be maintained before collection of the sample. Normal sperm production in man was reported to be constant at 4.25×10^6 /g weight of testis (14). Many authors have reported total sperm production to vary from 250×10^6 to 1.000×10^6 sperm/day (15). Some studies have shown a difference in count when the abstinence period was changed (10, 11). However, one author hypothesized that the more frequent the ejaculation, the more often the sperm count was within physiological limits (16). This was supported by an experimental study on dogs (17). A study conducted on semen of impotent patients showed that, whenever a second ejaculation was made in a 1-week period, the quality of semen improved in terms of volume, liquefaction time and sperm count (18).

TABLE I - RESULTS OF SEMEN EVALUATION AND CALCIUM AND MAGNESIUM LEVELS IN SEMINAL PLASMA AND SPERMATOZOA IN SAMPLES I & II

Samples (no.)	Volume, mL	Motility percentage				Seminal plasma		Spermatozoa	
		Count, millions/mL	Total	Active	Sluggish	Calcium	Magnesium	Calcium	Magnesium
I (24)	2.1±0.12* (1.1-3.5)	99.04±5.23 (56-156)	72.31±1.74 (60-90)	50.38±2.46 (30-78)	21.92±1.35* (12-37)	51.5±6.64 (16.0-76.0)	26.98±2.63 (7.12-51.48)	28.30±4.57 (1.12-19.59)	8.10±1.41 (1.12-19.59)
II (24)	2.45±0.13 (1.5-4.5)	92.75±4.00 (57-129)	77.71±1.9 (65-90)	51.21±1.89 (35-65)	26.5±1.47 (10-45)	45.67±3.7 (12-80)	28.37±2.63 (5.66-51.48)	18.39±2.41 (3.67-46.15)	19.5±1.49 (1.30-24.46)

Samples I and II refer to the 2 occasions on which samples were taken by each participant. Values are means ± SE; numbers in parenthesis indicate ranges.

*Significant difference ($p < 0.05$) between samples I and II.

Differences in the motility of spermatozoa in terms of pattern and velocity were observed. Accordingly, motility patterns, defined as either active or sluggish, were studied separately, and the results are presented in Table I. No significant difference in active motile spermatozoa was seen between groups. However, a significant difference ($p < 0.05$) was observed in the case of sluggish spermatozoa. Mallidis et al (12) reported within subject variability in percentage of sperm motility on different days in same subjects. The motility of spermatozoa was influenced by the different substances which were present. The increased number of abstinence days was responsible for poor sperm motility in many cases (19). As all the conditions, including abstinence time, were kept unchanged for the 2 collections, the differences observed here must be considered a normal outcome.

Calcium and magnesium levels present in seminal plasma and spermatozoa did not show any difference between ejaculation I and II. Calcium and magnesium present in seminal plasma are mainly secreted by the prostate gland. Cell membrane prevents the entry of

calcium and magnesium into spermatozoa. Our study employing X-ray diffraction on the human genital system showed that calcium was present the entire length of it (Skandhan, Skandhan, Avni 2007).

In conclusion, the present study showed 2 significant changes in seminal parameters: volume and percentage of sluggish motile spermatozoa. There was no change in the values for calcium and magnesium in seminal plasma or spermatozoa.

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