Semen analysis: one year experience with the automated cellsoft system

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Semen analysis is a cornerstone in the diagnosis of male infertility. The sperm concentration and percent motility results of the 813 semen samples examined both with a computerized method, the automated "CellSoft" system (Cryo Resources Inc., Newyork, NY) and the conventional method were compared. The mean sperm concentrations were 36.3242 (SD 28.770) x 10^6 ml and 45.4938 (SD 39.568) x 10^6/ml for "CellSoft" and conventional method respectively. By using the "CellSoft" analyzer the motility was 34.6653% (SD 19.998) while 39.7577% (SD 20.377) by the conventional method. The CellSoft system gave a mean sperm concentration of 0.193x10^6/ml in 54 azoospermic semen samples according to the conventional method. Although the average numbers were quite similar, the results are underestimated by "CellSoft" at each level of sperm concentration except the azoospermic samples, and the manual evaluation performed by trained technicians remains as the gold standart for semen analysis in our laboratory. [Turk J Med Res 1995, 13(4):159-162]

Key Words: Semen analysis, Automated "CellSoft" system

Semen analysis is still the first step and most commonly used procedure in the laboratory investigation of male infertility (1). Determination of sperm concentration, percentage of motile and normally shaped sperm are the main parameters of the analysis (2,3).

Today, sperm velocity, linearity of movement, lateral head displacement and beat frequency are introduced in the semen analysis in addition to classical parameters (4,5,6).

Despite the efforts of World Health Organization (WHO) to standardize the movement analysis of sperm, this paremeter remains highly subjective when interpreted by a technician in the conventional method (7). Semen analysis results show marked variation inter- and intra-individually (8).

The recently developed computerized systems designed for semen analysis, appear to provide more objective results and enable measurement of sperm velocity, linearity and lateral head movements (5,6).

In this article we compared a computerized method, the automated CellSoft system, with the conventional method in terms of sperm concentration and motility percent results of the semen samples.

Received: March 11,1995 Accepted: June 25, 1995

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MATERIALS AND METHODS

Eight hundred thirteen consecutive ejaculates of men attending urology clinic or In Vitro Fertilization-Embryo Transfer (IVF-ET) unit were examined.

All ejaculates were obtained by masturbation and collected in plastic containers after 3-4 days of abstinence. Semen analyses were done within 1 hour of ejaculation after liquefaction at 37°C.

The conventional semen analysis was performed according to the WHO guidelines (7). All semen samples were also analysed by the CellSoft system using the threshold parameters recommended by the company (Table 1). All particles with a size range of 4-25 pixels are accepted as spermatozoa and 10 um/s was set for the threshold velocity.

To assess the possible errors of parameter setting, not only the whole, but also the first hundred ejaculates and the subgroups constitued upon the sperm concentration (SC) results of standard procedure were analysed to determine statistical difference.

The subgroups were: a) azoospermic
b) SC < 20x10^6/ml
c) 20x10^6/ml < SC > 40x10^6/ml
d) 40x10^6/ml < SO 100x10^6/ml
e) SC > 100x10^6/ml

Table 1. Set up of CellSoft system measurement parameters

Number of fra^i ies to analyze		10
Number of frames per secondes		12.5
Minimum sampling	Motile	2
	Velocity	6
Maximum velocity (micron/second)		250
Threshold velocity (micron/second)		10
Pixel scale (micron/pixel)		0.688
Dilution factor		1
Cell size range (pixels)		4-25

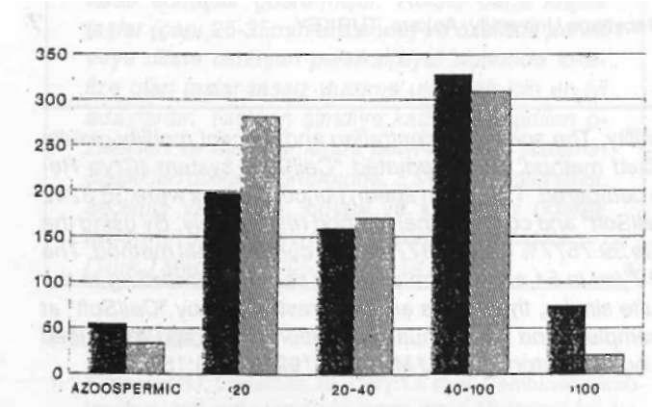

Figure 1. Frequency distribution of sperm concentration in 813 semen samples.

All statistical analyses were performed on an IBM-compatible personal computer using the SPSS+PC+ software. The paired t-test was used to evaluate the results of sperm concentration and percent motility of conventional and automated analysis.

RESULTS

The frequency distribution of sperm concentration in the 813 semen samples determined by the conventional and automated systems are shown in Figure 1.

The CellSoft system gave a mean sperm concentration of 0.193x10^6/ml in 54 azoospermic semen samples according to the conventional method. The results of the whole 813 semen sample analyses and the subgroups described in materials and methods are shown in Tables 2-6.

The results of the first one hundred samples did not show any significant difference from those of the remaining 713 samples in terms of sperm concentration and motility percent results- (Tables 7,8). The sperm concentration and motility percents were underestimated in all groups but the azoospermic samples by the CellSoft system (p<0.001).

DISCUSSION

The CellSoft Automated Semen Analyzer (ASA) is designed to furnish objective and detailed analysis of semen specimens. It is applicable to both veterinary and human investigation (User Manual, CellSoft).

The CellSoft ASA is able to recognize most sperm cells and to distinguish them from other semen constituents based on their size, luminosity and motion (8). These three factors constitute an integral component of the system ability to provide accurate and objective data.

In our study CellSoft system gave a mean sperm count of 0.193x10^6/ml in 54 azoospermic patients. The false record of the motile sperm in azoospermic patients cannot be corrected even when visual correction is made by the adjustment of the gray scale. Following swim-up or washing procedures these false recorded images dissappeared. This finding probably indicates that leukocytes or other seminal particles cannot be distinguished from normal sperm by the CellSoft ASA (9).

Table 2. The mean sperm concentration and motility percent results of 813 samples examined both with CellSoft and conventional method

	Conventional method	CellSoft system	
Sperm concentration (x10^6/ml)	45.4938	36.3242	p<0.001
Motility percent (%)	39.7577	34.6653	p<0.001

Table 3. Mean sperm concentration and percent motility results in samples with a concentration up to 20x10^6/ml by conventional method

	n	Conventional mean±SD	CellSoft mean±SD	
Sperm concentration (x10^6/ml)	198	10.27±5.0	9.35±4.6	p<0.001
Motility (%)	198	35.48±16.0	**31.81*17.1**	p<0.01

Table 4. Mean sperm concentration and percent motility results in samples with a concentration between 20-40x10^6/ml by conventional method

	n	Conventional meaniSD	CellSoft mean+SD	
Sperm concentration (x10^6/ml)	159	28.23±5.6	22.86±6.3	p<0.001
Motility (%)	159	35.00±14.0	29.58±15.1	p<0.001

Table 5. Mean sperm concentration and percent motility results in samples with a concentration between 40-100x10⁶/ml by conventional method

	n	Conventional mean±SD	CellSoft mean±SD	
Sperm concentration (x10⁶/ml)	328	63.13± 15.6	53.38±15.8	p<0.001
Motility (%)	328	45.87±15.7	39.68±15.8	p<0.001

Table 6. Mean sperm concentration and percent motility results in samples with a concentration greater than 100x10⁶/ml by conventional method

	n	Conventional mean±SD	CellSoft mean±SD	
Sperm concentration (x10⁶/ml)	74	131.82140.9	88.18±23.7	p<0.001
Motility (%)	74	63.31 ±19.9	56.24±21.9	p<0.001

Table 7. The results of the first 100 samples analyzed

	Conventional method	CellSoft system	
Sperm concentration (x10⁶/ml)	54.5150	30.6780	p<0.001
Motility percent (%)	44.7200	34.4730	p<0.001

Table 8. The results of the last 713 samples analyzed

	Conventional method	CellSoft system	
Sperm concentration (x10⁶/ml)	44.2286	37.1161	p<0.001
Motility percent (%)	39.0617	34.6923	p<0.001

The same problem was announced by several investigations in semen samples of up to 80x10⁶/ml correlation (8,9). However, the results were more comparable in sperm densities of 20 to 80x10⁶/ml (10).

Underestimation of actual sperm concentrations in all groups by the CellSoft is not a usual finding in many similar studies (8,9,10). It seems crystal clear that it is not only the CellSoft system which is responsible for this outcome. Probably failure in parameter setting and errors in operating the system are further causes of these unexpected results. Again, it is very hard to explain our underestimated motility results by the CellSoft. We don't try to find a scientific excuse for this.

This study has clearly demonstrated that manual evaluation performed by trained technicians remains as the present gold standard for semen analysis and the CellSoft system has severe limitations as an automated semen analyser in our routine laboratory setting. It certainly needs additional modules to determine sperm velocity, linearity and lateral head displacement.

Semen analizi: Bilgisayarlı "Cellsoft" sistemle bir yıllık deneyim

Semen analizi erkek infertilltesi tanısında çok önemli bir yere sahiptir. Çalışmamızda 813 semen örneği konvansiyonel yöntemle ve bilgisayarlı "CellSoft" sistemi (Cryo Resources Inc., Nevvyork, NY) ile değerlendirilmiş; sperm konsantrasyonları ve motil sperm yüzdeleri karşılaştırılmıştır. Ortalama sperm konsantrasyonları "CellSoft" ve konvansiyonel yöntemler için sırasıyla 36.3242 (SD 28.770) x 10⁶/ml ve 45.4938 (SD 39.568) x 1(r/ml bulunmuştur. "CellSoft" cihazı ile %34.6653 (SD 19.998) motil sperm saptanırken, konvansiyonel yöntemle bu oran %39.7577 (SD 20.377) olarak hesaplandı. Bilgisayarlı "CellSoft" sistemi, konvansiyonel yöntemle azoospermik olarak saptanan semen örnekleri için ortalama 0.193x10⁶/ml sperm konsantrasyon değeri vermiştir. Ortalama rakamlar benzer olsa da, sonuçlar her sperm konsantrasyon düzeyinde (azoospermik örnekler dışındı) "CellSoft" ile daha düşük bulunmuştur. Laboratuvarımızda semen analizi için deneyimli teknisyenlerin uyguladıkları konvansiyonel yöntem halen altın standart olma özelliğini korumaktadır.
[TurkJMedRes 1995, 13: (4): 159-162]

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