Characteristics Of Human Sperm DNA And Its Relationship With Male Infertility

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Objective: To determine variation in sperm chromatin among different categories of human semen samples.

**Design:** 34 human semen samples including 16 normozoospermic, 15 asthenozoospermic and 3 oligoasthenozoospermic.

Materials and Methods: Assessment of the purity of the DNA samples was performed by spectral characteristic analysis (within a wavelength of 230-290nm) as well as calculation of $A_{260}/A_{280}$ ratio. Estimation of the DNA content, RNA and protein as contaminants were done by diphenylamine method, orcinol method and Lowry’s method respectively. RAPD analyses of sperm DNA samples (categorized on the basis of motility difference) were done using random decamer primer (5’AATCGGGCTG 3’) solely to detect polymorphism within different categories of semen samples. A dissimilarity matrix was prepared for identifying the extent of variation. Band frequencies were also calculated.

Results: Spectral analysis revealed no observable differences for respective DNA samples. The $A_{260}/A_{280}$ ratios (1.98± 0.2) indicated pure DNA samples. Results of DNA content estimation showed a lack of correlation with the nature of the semen samples. RNA was absent in all cases and some of the DNA samples had protein contamination. Identification of the major bands for each category of DNA samples was performed only on higher values of the band frequencies (0.75-1.0). Four major bands were recognized in sperm DNA from samples with normal motility (70-80%), with higher dissimilarity indices of 1.0 and 0.8. Five major bands were identified in sperm DNA with 60-70% motility with low values of dissimilarity indices (0.07-0.53). RAPD of sperm DNA with 50-60% motility also showed 5 major bands with frequency of 1.0. The number of major bands recognized in sperm DNA with a motility percentage of (40-50), (30-40) and (0-30) were 1, 5 and 10, respectively. Sperm DNA in two of the oligoasthenozoospermic samples revealed 100% similarity. RAPD analysis of sperm DNA in different semen categories revealed the lack of existence of even a single band with a band frequency of 1.0.

Conclusion: The present investigation provides a clear view of the genetic variation among different categories of human semen specimens at the level of sperm chromatin.
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