Frequency of subnormal hypoosmotic swelling tests increase with advancing age of the male

J.H. Check, D. Kramer, A. Bollendorf, C. Wilson

The University of Medicine and Dentistry of New Jersey, Robert Wood Johnson Medical School at Camden, Cooper Hospital/University Medical Center, Department of Obstetrics and Gynecology, Division of Reproductive Endocrinology & Infertility, Camden, NJ (USA)

Summary

Purpose: To determine if there is an increased frequency of abnormal hypoosmotic swelling (HOS) tests when testing semen in males of advanced reproductive age. *Methods:* Ten-year retrospective review according to six age groups (\leq 29.9, 30-34.9, 35-39.9, 40-44.9, 45.0-49.9, and \geq 50). *Results:* Compared to males < 35 years old, the frequency of HOS scores < 50% doubles in males 45-49.9 and quadruples in males \geq 50. *Conclusions:* Males with low HOS scores despite having no problem with fertilization of the oocytes rarely achieve a pregnancy because of embryo implantation defects unless intracytoplasmic sperm injection (ICSI) is performed. Thus at least one abnormality seen in sperm of males of advanced reproductive age is correctable by performing in vitro fertilization with ICSI.

Key words: Advanced reproductive age; Hypoosmotic swelling test; Embryo implantation defect.

Introduction

Despite the demonstration that when evaluating various semen parameters a subnormal HOS test score of < 50% far better predicts low implantation potential of embryos achieved by conventional insemination compared to low motile density and morphology, this simple inexpensive test is rarely evaluated by most physicians treating infertility [1, 2]. This is probably related to the fact that some studies found the HOS test to be the least valuable semen parameter to predict poor fertilization rates [3-6].

However most physicians treating infertility are not aware that certain male factor abnormalities, e.g., a low HOS test score can produce normal appearing embryos that rarely implant [7-10]. The aim of the present study was to see if the percentage of abnormal HOS test scores, presumably related to a toxic factor possibly in the ejaculatory system, may increase with advancing age.

Materials and Methods

A retrospective evaluation of the HOS test scores was performed on all initial semen analyses performed in our office over a 10-year time period. The HOS scores were sorted into six groups based on ranges of age: \leq 29.9 years, 30.0-34.9 years, 35.0-39.9 years, 40.0-44.9 years, 45.0-49.9 years and \geq 50.0 years.

The HOS test was performed by combining 0.1 ml of ejaculate with 1.0 ml hypoosmotic solution (fructose/sodium citrate) following precisely the technique described by Jeyendran et al [11]. After incubation of the mixture for at least 30 min at 37°C, 100 spermatozoa were observed with a phase-contrast microscope for tail swelling changes typical of a reaction in the HOS test. The HOS tests were performed on unprepared specimens

during standard semen analysis. An HOS test score < 50% was considered abnormal.

Results

A total of 4,309 patients were evaluated. The frequency of low HOS test scores according to age is seen in Table 1. The percentage of subnormal levels gradually increase with each age group with an acceleration in the two oldest groups such that abnormal HOS test scores were twice as high in males age 45-49.5 as males < 35 and were more than 4-fold higher in males aged ≥ 50 .

Conclusions

The HOS abnormality is correctable by IVF with ICSI [12] and to a lesser extent by first treating the sperm with the protein digestive enzyme chymotrypsin [13, 14]. Though we perform this simple inexpensive test routinely irrespective of male age, it behooves the treating infertility specialist to evaluate HOS especially in males of advanced reproductive age.

It is not clear what the specific etiologic factor is for developing the hypothesized toxic factor that leads to a subnormal HOS test. Whatever this toxic factor is, it seems to increase in frequency with age.

Table 1. — Frequency of low HOS tests according to age.

	Age range					
	≤ 29.9	30-34.9	35-39.9	40-44.9	45-49.9	≥ 50
No. evaluated	481	1173	1288	835	357	175
No. with low HOS score % with low	26	77	103	81	46	44
HOS score	5.41	6.56	8.00	9.7	12.9	25.1

Revised manuscript accepted for publication February 10, 2011

References

- [1] Check J.H.: "The infertile male -Diagnosis". Clin. Exp. Obstet. Gynecol., 2006, 33, 133.
- [2] Check J.H., Epstein R., Nowroozi K., Shanis B.S., Wu C.H., Bollendorf A.: "The hypo-osmotic swelling test as a useful adjunct to the semen analysis to predict fertility potential". *Fertil. Steril.*, 1989, 52, 159.
- [3] Barratt C.L.R., Osborn J.C., Harrison P.E., Monless N., Dumphy B.C., Lenton E.A., Codie I.D.: "The hypoosmotic swelling test and the sperm mucus penetration test in determining fertilization of the human oocytes". *Hum. Reprod.*, 1989, 4, 430.
- [4] Sjoblum P., Coccia E.: "On the diagnostic value of the hypoosmotic sperm swelling test in an in vitro fertilization program". J. In Vitro Fertil. Embryo Transfer., 1989, 6, 41.
- [5] Avery S., Bolton U.M., Mason B.A.: "An evaluation of the hypoosmotic sperm swelling test as a predictor of fertilizing capacity in vitro". *Int. J. Androl.*, 1990, 13, 93.
- [6] Chan S.Y.W., Wang C., Chan S.T.H., Ho P.C.: "Differential evaluation of human sperm hypoosmotic swelling test and its relationship with the outcome of in vitro fertilization of human oocytes". *Hum. Reprod.*, 1990, 5, 84.
- [7] Check J.H., Stumpo L., Lurie D., Benfer K., Callan C.: "A comparative prospective study using matched samples to determine the influence of subnormal hypo-osmotic test scores of spermatozoa on subsequent fertilization and pregnancy rates following in-vitro fertilization". *Hum. Reprod.*, 1995, 10, 1197.
- [8] Katsoff D., Check M.L., Check J.H.: "Evidence that sperm with low hypoosmotic swelling scores cause embryo implantation defects". Arch. Androl., 2000, 44, 227.

- [9] Kiefer D., Check J.H., Katsoff D.: "The value of motile density, strict morphology, and the hypoosmotic swelling test in in vitro fertilization-embryo transfer". Arch. Androl., 1996, 37, 57.
- [10] Check J.H., Katsoff D., Check M.L.: "Some semen abnormalities may cause infertility by impairing implantation rather than fertilization". *Med. Hypoth.*, 2001, 56, 653.
- [11] Jeyendran R.S., Van der Ven H.H., Perez-Pelaez M., Crabo B.G., Zaneveld L.J.D.: "Development of an assay to assess the functional integrity of the human sperm membrane and its relationship to other semen characteristics". *J. Reprod. Fertil.*, 1984, 70, 219.
- [12] Check J.H., Katsoff D., Check M.L., Choe J.K., Swenson K.: "In vitro fertilization with intracytoplasmic sperm injection is an effective therapy for male factor infertility related to subnormal hypo-osmotic swelling test scores". J. Androl., 2001, 22, 261.
- [13] Katsoff D., Check J.H.: "Two methods of achieving pregnancies despite subnormal hypo-osmotic swelling test scores". Fertil. Steril., 1997, 68, 549.
- [14] Check M.L., Katsoff D., Check J.H., Summers-Chase D.: "Effect of treating sperm with low hypo-osmotic swelling test scores with chymotrypsin on pregnancy rates after conventional in vitro fertilization-embryo transfer". Fertil. Steril., 2004, 82, 741.

Address reprint requests to: J.H. CHECK, M.D., Ph.D. 7447 Old York Road Melrose Park, PA 19027 (USA) e-mail: laurie@ccivf.com