Sperm %DFI correlated with AB (r=0.29, P=0.015) and IAF staining (r=0.25, P=0.034), and, sperm %HDS (a marker of chromatin compaction) also correlated with AB (r=0.43, P=0.0006) and IAF staining (r=0.30, P=0.014). Sperm AB staining was highly correlated with IAF staining (r=0.75, P=0.00001). Sperm %DFI, %HDS, AB and IAF staining all correlated inversely with sperm concentration and sperm progressive motility.

CONCLUSION: These data demonstrate that sperm chromatin compaction is abnormal in men with mild male-factor infertility, many of whom are normozoospermic. The strong correlation between AB and IAF nuclear staining suggests that both markers measure the accessibility of the stain to the nuclear target and indirectly assess sperm chromatin compaction.

SPERM BIOLOGY

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THE ARYL-HYDROCARBON RECEPTOR (AHR) PLAYS A ROLE IN MEDIATING SPERMATOCYTE HEXOSE TRAFFICKING. K. Omurtag, B. DeBosch, P. Esakky, E. Schoeller, K. Moley. Obstetrics and Gynecology, Washington University St Louis School of Medicine, St Louis, MO.

OBJECTIVE: Accumulated evidence demonstrates that the toxic effects of dioxins are mediated through AHR with one possible mechanism being the disruption of glucose transport. Glucose transport aberrations are known to disrupt sperm metabolism leading to decreased motility and fertilization in vitro. Considerable epidemiologic research has explored the association between AHR agonists and male infertility. We aim to examine the relationship between AHR and glucose/fructose trafficking in the spermatocyte.

DESIGN: In vitro cell culture model.

MATERIALS AND METHODS: Murine spermatocytes were exposed to 40 mcg/mL of cigarette smoke condensate (CSC) or CSC+AHR antagonist for 6, 18, 24 hrs. Gene and protein expression of GLUT8 and GLUT12 were compared to untreated and DMSO controls using QT-PCR,Western Blot, and IHC. Radio-labeled glucose/fructose uptake assay was performed to show changes in hexose uptake.

RESULTS: In the CSC group, gene expression of Ahr is increased ~2.5 fold at 6 hrs. Glut8 is ~2 fold and ~30 fold increased at 18, 24 hrs post exposure, respectively, and Glut12 increased 6 fold at 24 hrs.

In the CSC+AHR antagonist group, Ahr remains maximally expressed at 6 hrs. Glut8 decreases ~10 fold and ~2 fold at 18, 24 hrs, respectively. Glut12 expression is decreased ~2 fold at 18 hrs but increases 2.4 fold at 24 hrs. AHR protein levels remained unchanged in the CSC group at 18, 24 hrs, while levels of GLUT12 increased 1.5 fold. GLUT8 levels decreased ~2 fold in the CSC group but markedly declined after antagonist exposure. At 18, 24 hrs fructose uptake increased 3 fold among spermatocytes exposed to CSC compared to controls. Glucose uptake was increased, but trend was not significant.

CONCLUSION: Understanding how environmental toxins affect spermatogenesis provides better insight into their effects on male infertility. Our results hint at AHR in mediating the uptake of glucose/fructose within spermatocytes. Additional work using a murine in vivo model to examine these trends is underway.

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AUTOMATED SEMEN ANALYSIS…THE NEW GOLD STANDARD? A COMPREHENSIVE STUDY COMPARING MANUAL AND AUTOMATED SEMEN ANALYSIS. J. Lammers, W. Mansour, S. Lattes, M. Jean, P. Barriere, T. Freour. Médecine et Biologie de la Reproduction, University Hospital of Nantes, Nantes, France.

OBJECTIVE: To compare conventional manual sperm analysis performed according to WHO 5th edition manual (2010) to a fully automated sperm analyzer (SQA-V) in terms of standardization, accuracy and precision.

DESIGN: Prospective study.

MATERIALS AND METHODS: All men who received routine fertility evaluations at the andrology laboratory between February and April 2011 were included. Untreated semen specimens with ejaculate volumes of>2.5 ml were included in the trial. Sperm concentration, total number of sperm, motility, progressive motility, motile sperm concentration (MSC), progressively motile sperm concentration (PMSC) and normal morphology were manually and simultaneously assessed by two independent operators (WHO 2010). In parallel, sample spits were run in duplicate on the SQA-V.

Statistical evaluation comparing mean values and coefficients of variation were performed. Additionally, correlation, mountain plot distribution and ROC curve analysis parameters were included in the evaluation.

RESULTS: 250 patient samples were included in the study. 246 of 250 samples were compared for Sperm Concentration, Total Sperm Number and MSC. Only partial semen analysis was performed in 26 cases of severe oligospermia due to variations between methods in expressing semen parameters. Correlation coefficients were very high for sperm concentration, total sperm number, MSC, and PMSC (r>=0.93 for all parameters). The precision of the SQA-V was excellent for all semen variables (CV<10%) and better than the manual method (CV<=27.4%). Specificity and Negative Predictive Value of the SQA-V versus manual assessment for Morphology were: 98% and 93% correspondingly.

CONCLUSION: The SQA-V demonstrated very good agreement versus strictly controlled manual semen analysis (WHO 2010). This automated system was easy to integrate into the laboratory routine. The SQA-V automated sperm analyzer provides accurate results and can enhance laboratory standardization of sperm analysis when compared to strictly controlled manual sperm analysis.

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SPERM DEOXYRIBONUCLEIC ACID FRAGMENTATION LEVEL, AS MEASURED BY TUNEL TEST, IS NOT RELATED WITH THE IVF OUTCOME IN GOOD PROGNOSIS WOMEN. J. Llacer,[a] B. Lledo,[b] R. Morales,[b] A. Rodriguez,[a] J. Ten,[a] R. Bernabeu.[a] [a]Reproductive Medicine, Instituto Bernabeu, Alicante, Spain; [b]IB Biotech, Instituto Bernabeu, Alicante, Spain.

OBJECTIVE: To establish the impact of sperm DNA fragmentation in IVF outcomes in patients with good prognosis. (normozoospermic men, conventional IVF and egg donation).

DESIGN: Prospective blind-observer case control study.

MATERIALS AND METHODS: Prospective study of 160 egg donation cycles using conventional IVF with>6 oocytes. Endometrial thickness of the recipients was>7mm and normozoospermic men were aged between 26 and 59 years. At the time of insemination an aliquot of capacited seminal sample was dropped onto slides to determine the sperm DNA fragmentation by TUNEL method (terminal deoxyribonucleotidyl transferase-mediated dUTP nick-end labelling). A minimum of 500 spermatozoa were evaluated for each sample.The percentage of TUNEL-positive spermatozoa was referred to as DNA fragmentation index (DFI) and was considered abnormal when it was above 15%. Main reproductive variables were compared with respect to DFI. Statistical analysis was performed using SPSS analysis applying Student's t for independent groups, Pearson correlation to assess confounding factors and Multiple Linear Regression.

RESULTS: After the analytical study, there was no significant difference between DFI with clinical pregnancy, implantation and miscarriage rates, obtaining P>0.05 between the means of DFI of the selected groups.

Results

	DFI>15	DFI<15	
Implantation Rate (%)	36,02	34,24	P=0,304
Clinical Pregnancy Rate (%)	52,00	49,20	P=0,698
Miscarriage Rate	15,40	25,42	P=0,594
Live Birth Rate	46,42	37,25	P=0,105

CONCLUSION: Outcomes in IVF are not influenced by the sperm DNA fragmentation in egg donation at least when conventional IVF is used. This fact can be related with the ability of the egg to repair DNA damage. This repair capacity would be particularly striking in young women without fertility problems such as donor.

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