10.45-11.00

menting longitudinal quality control schemes for diagnostic purposes as well as for multicentric studies.

\*Blandin T., Bordeaux; Daudin M., Lamartre A., Toulouse, France; Diaz I., Bogota, Colombia; El Matribi S., Paris; Gony B., Caen; Keskes L., Sfax, Tunisia; Kolbelsen M., Virant-Klun I., Ljubljana, Slovenia; Lornage J., Pitaval G., Lyon, France; Nomal N., Pointe à Pître; Simon O., Rennes, France.

## 10.30–10.45 O-118. New definition for Sertoli cell-only syndrome

Paz G., Bar-Shira Maymon B., Yogev L., Hauser R., Bochan A., Kleiman S. and Yavetz H.

## Institute for the Study of Fertility, Lis Maternity Hospital, Tel Aviv Sourasky Medical Center, Tel Aviv, Israel

**Introduction:** Sertoli cell-only (SCO) is defined and based on the histological appearance of paraffin sections of testicular biopsies. Thus, if the tubules contain only Sertoli cells without any germ cells, the biopsy will be diagnosed as belonging to an infertile man with SCO syndrome. We are challenging this definition by employing immunohistochemical methods to prove the absence of germ cells and to verify the maturity of the Sertoli cells present in the tubules under investigation.

Materials and methods: Testicular biopsies were taken from 40 azoospermic men who underwent testicular sperm extraction (TESE). One portion of the biopsy was taken for histological work-up. The sample was fixed in 4% buffered formalin and embedded in paraffin. Sections of 3  $\mu$ m were mounted on glass slides and stained by haematoxylin–eosin. Additional slides were prepared for immunohistochemical staining of RNA binding motif protein (RBM) and cytokeratin 18 (CK-18) The stained cells were assessed by light microscopy.

**Results:** In all subjects with normal spermatogenesis (six obstructive azoospermic men), RBM expression was detected in germ cells, e.g. spermatogonia, spermatocytes and round spermatids. Elongated spermatids and mature sperm cells were never stained. Sertoli cells in these cases were poorly stained for CK-18 (proving their maturity). In contrast, in 12 men defined as SCO none of the cells were positively stained for RBM. Part of the Sertoli cells in this group were stained for CK-18. In 22 men mixed atrophy was found in the histology of the biopsies; tubules with spermatogenesis were positively stained for RBM in contrast to the negative stained tubules with absence of germ cells. The latter were often stained for CK-18.

**Conclusion:** In the present study we were able to show that seminiferous tubules in azoospermic men may contain premeiotic germ cells located between immature Sertoli cells. Diagnosis of SCO can be made only after probing the biopsy with antibody generated against germ cells. Thus, the use of the immunohistochemical tools is essential for definition of the spermatogenic status of a testicular biopsy and may have a prognostic value in the near future.

## O-119. High sperm hyperhaploidy rates for chromosomes 1, 17, X and Y in men with severe male factor infertility

Bernardini L.<sup>1</sup>, Gianaroli L.<sup>2</sup>, Fortini D.<sup>2</sup>, Selman H.<sup>2</sup>, Conte N.<sup>1</sup> and Venturini P.L.<sup>1</sup>

<sup>1</sup>Dept of Ob/Gyn, S.Martino's Hospital, University of Geneva and <sup>2</sup>S.I.S.M.E.R., Reproductive Medicine Unit, Bologna, Italy

**Introduction:** Whether infertile patients undergoing intracytoplasmic sperm injection (ICSI) are at increased risk for higher frequencies of sperm aneuploidy is the subject of current debate. In previous studies we reported that as compared to normal healthy controls, infertile men with low counts of morphologically normal and motile spermatozoa carry a 2fold increase of sex chromosome aneuploidy. In this study we attempted to assess whether this is particularly true whenever the quality of semen analyzed becomes poorer.

Materials and methods: Sperm aliquots of ejaculated semen from 10 patients, scheduled for ICSI and presenting with severe oligoasthenoteratozoospermia (OAT) and normal peripheral karyotype, were processed for double target in-situ hybridization. For each individual patient, centromeric DNA probes for chromosomes 1, 17, X and Y were employed in duplicate. Slides were scored blindly by two observers and 2000 cells, on average, analysed per patient. Mean  $\pm$  SD patient age was 38.5  $\pm$  3.1. Comparison of the sample means was performed by non-parametric rank sum test for independent samples.

Results: Results are presented in Table I.

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	Controls	OAT patients	P values		
No. of cases	10	10			
Total normal motile counts	$30 \times 10^{6}$	$0.86 \times 10^{6}$	< 0.0001		
No. of cells scored/patient	2000	2000			
In-situ hybridization					
Disomy 1 + 17	0.66	2.95	< 0.001		
DNA ploidy XX	0.15	0.24	NS		
YY	0.25	1.5	< 0.001		
XY	0.45	3.1	< 0.001		
Diploidy	0.4	0.5	NS		
Total	1.9	8.3	< 0.0008		

NS = not significant.

**Conclusion:** Our data show that OAT patients carry markedly high rates of sperm aneuploidy for different chromosomes. Diploidy appears to undergo no significant changes as a function of semen quality. This makes hyperhaploidy a source of concern during preconception counselling of men with severe male factor infertility.