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Sperm Cryopreservation in Cancer Patients

J. Skrzypek, W. Krause

Cancer patients are treated multiprofessionally by medical specialists. We predominantly see testicular tumors and haematological malignancies. From 2002 to 2006, 82 cancer patients presented for cryopreservation of semen in our department. Age distribution and various oncological diseases are consistent with epidemiological data. In all malignancies, semen quality was reduced, with a pronounced reduction in testicular tumors.

In cancer treatment, a variety of therapeutic strategies such as surgery, radiotherapy, and chemotherapy are well established. Especially radiotherapy and chemotherapy are potentially hazardous for spermatogenesis, the individual risk, however, cannot be predicted. Therefore, cryopreservation of spermatozoa should be offered to all patients prior to cancer treatment. *J Reproduktionsmed Endokrinol* 2007; 4 (2): 106–8.

Key words: cryopreservation, malignant neoplasms, semen, spermatozoa

Experimental Research with Sperm Cryopreservation

According to the literature, among patients asking for sperm cryopreservation prior to cancer treatment, men with testicular tumors in general show poorer sperm quality than those with other types of cancer. Sperm quality is defined by sperm count, sperm motility, and sperm morphology as well as freezability, i.e. the rate of post-thaw viability of sperm.

Agarwal et al [1] reported that semen quality did not significantly differ among patients with stage I, II, or III of testicular cancer. Semen quality was higher among patients with pure seminomas than those with pure embryonal tumors; quality was lowest among patients with mixed germ cell tumors. Although sperm motility decreased after cryopreservation, the decline did not differ significantly between patients and controls. Two years later, a report from the same group demonstrated that patients with leukemia or advanced organ cancer had a higher pre-freeze and post-thaw motility than patients with testicular cancer [2].

Lass et al [3] found significantly lower sperm counts in 79 men with testicular tumors as compared to 121 men with hematological malignancies. No differences within the groups, i.e. between seminoma and non-seminoma, as well as between lymphomas and leukemias, were observed. Crha et al [4], as well as Gandini et al [5], again confirmed that men with testicular cancer had significantly higher frequency of severe sperm pathology. Only Chung et al [6] found that sperm counts, motility, and morphology did not vary among the different cancer groups.

Surgery, radiation, and chemotherapy of cancer may result in the impairment of spermatogenesis. The degree of toxicity for gonadal function depends on the chemotherapeutic agents as well as radiation dosages [7]. Cryopreservation of semen should be offered to adolescents as well, since an impairment of fertility is already observed in children and young patients treated for malignancies [8]. Postovsky et al [9] reported on a series of 27 patients,

age 14 to 19, of which 24 succeeded in providing semen for analysis and cryopreservation. Prior to cancer treatment, patients should be informed about the possibilities of cryopreservation and assisted reproduction techniques (ART), enabling them to achieve a decision on the storage of spermatozoa. In adolescents, sensitive handling is required. Parents should be part of the counselling process to gain support and to achieve an informed consent [10].

The Impact of Sperm Cryopreservation

Most studies show sperm count in patients with testicular cancer similar to those with haematological or other malignancies. Skakkebaek [11] introduced the term “testicular dysgenesis syndrome”, summarizing poor semen quality, testicular cancer, undescended testes (cryptorchidism), and hypospadias as symptoms of one underlying entity. According to Agarwal and Allamaneni [12], sperm quality reduction is aggravated by para- and endocrine as well as systemic effects of the testicular tumor. The disruption of the blood/testis barrier by the tumor leads to the development of antisperm antibodies. Padron et al [2] describe that “poor semen quality” will also enclose poorer freezability of spermatozoa.

This raises the question whether poorer pre-freeze and post-thaw sperm quality in tumor patients also indicates poorer fertility. Magelssen et al [13] compared fertility in men treated for testicular tumor prior to and after treatment. In this study, 279 of 422 men (66 %) had fathered a child prior to the disease. Twenty years after tumor therapy, 513 of 1359 men (37.7 %) had fathered a child spontaneously. While these figures do indicate lower fertility following tumor treatment, the study does not disclose the number of men remaining deliberately childless.

The same group (Brydoy et al [14]) supplied answers to this question: of 1433 men, 554 attempted to father a child after treatment for testicular cancer. 393 (71 %) of them succeeded: 48 % after high-dose chemotherapy and 92 % after orchidectomy. Only patients with a recovered spermatogenesis are included in these figures. However, semen cryopreservation is performed in order to obtain a fertility reserve for patients whose spermatogenesis does not recover after tumor therapy. Findings on the fertility rate following the use of cryopreserved semen are rare, since stored sperm is generally used in only 10 % of these cases. Of course, for fertilization the application of cryopreserved semen always requires assisted reproduc-

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tion techniques. The number of attempts quoted in the literature is too low to reach valid statements on fertility variations associated with different tumor types.

Agarwal et al [15] reported on a total of 87 ART cycles, in 42 of which intrauterine insemination, in 26 IVF, and 19 ICSI was performed. 18.3 % of the treatments resulted in a pregnancy, 75 % of the pregnancies resulted in a live-birth. There was no significant difference in the outcomes when the results were stratified by type of ART and malignancy.

Meseguer et al [16] reported on 30 ICSI cycles using cryopreserved semen of cancer patients. The number of pregnancies was identical to those of the control group, but the study did not differentiate the outcome according to the type of cancer.

In a survey of oncologists of two cancer centres in the USA, Schover et al [17] observed that 91 % of the responders (response rate 24 %) agreed that sperm banking should be offered to all men at risk of infertility as a result of cancer treatment. In contrast, 48 % of the oncologists either never discuss the topic or mention it to less than a quarter of the eligible patients. The questionnaire revealed a lack of knowledge about cryopreservation and ART. A questionnaire targeted at consultant oncologists, haematologists, and urologists listed in the Irish Medical Directory (response rate 63 %) showed that only 73 % of the responders routinely discuss semen freezing with their male tumor patients. Deficits in the knowledge about ART and existing facilities were detected as well [18]. Taking into account that in both surveys the responders had an increased awareness about the fertility risks of tumor treatment, reality is likely to be worse. On the basis of our practical experience, we can assume the situation in Germany to be comparable, but no data are available.

In conclusion, patients asking for cryopreservation of semen undoubtedly have poorer sperm parameters, if they suffer from testicular tumors rather than from hematological or other malignancies. Since the hazardous effects of different therapies for malignancies cannot be predicted, cryopreservation of semen should be offered to all eligible patients. To improve the awareness of risks to fertility through cancer treatment in children, adoles-

cents, and young males, particular efforts by all medical specialities are needed.

Our Results

Below, let us quote from the observations in patients seen in our group. From January 2002 to December 2005, semen samples of 86 men were prepared for cryopreservation. In all cases, cryopreservation was performed prior to cytotoxic treatment, regardless of whether chemotherapy or radiotherapy was applied. Patients were classified by the stage of their disease. Not included in the following data were 4 men, 3 who asked for cryopreservation to establish a fertility reserve prior to vasectomy and 1 prior to immunosuppressive therapy.

Tumor diagnoses are summarized in Figure 1. The seminal parameters prior to cryopreservation are listed in Table 1.

Cryopreservation of human spermatozoa is performed in our laboratory using the following method:

- Preparation: Mixing of the semen with the same volume of SteriTec L (containing 20 % glycerol, i.e. end concentration of 10 % glycerol; SteriPharm, Berlin, Germany) as a cryoprotectant and filling of straws as indicated below.

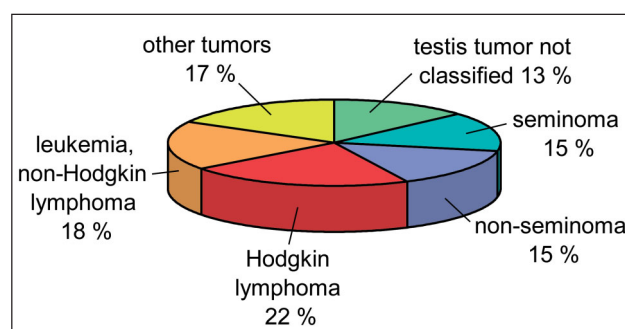


Figure 1: Distribution of tumor type in 82 patients requiring cryopreservation. "Other tumors" comprise the following diagnoses: colon adenocarcinoma, anaplastic carcinoma (organ not known), cardiacar carcinoma, Ewing sarcoma, glioblastoma, nephrosarcoma, medulloblastoma, neuroectodermal tumor (without further classification), pancreatic carcinoma, pheochromocytoma, prostate carcinoma.

Table 1: Age and seminal values according to tumor types

Tumor type	N	Age (years)	Semen volume (ml)	Sperm count (10 ⁶ /ml)	Sperm motility grades a and b prior to cryopreservation (%)	Sperm motility grades a and b after cryopreservation (%)	Non-vital sperm after cryopreservation (%)
Seminoma	12	28.9 ± 5.4	3.9 ± 1.8	32.6 ± 34.1	31.3 ± 15.2	5.0 ± 5.3	69.3 ± 14.6
Non-seminoma	12	26.7 ± 7.6	3.9 ± 1.6	54.9 ± 57.7	32.3 ± 17.1	7.9 ± 5.9	64.3 ± 20.6
Testicular tumor, unclassified	11	28.8 ± 6.6	3.3 ± 1.7	37.1 ± 40.0	23.8 ± 21.4	6.7 ± 8.2	69.8 ± 15.1
Testicular tumor in general	35	28.1 ± 9.2	3.7 ± 1.7	41.6 ± 43.4	29.3 ± 17.0	6.5 ± 5.9	67.7 ± 23.3
t test with "hematological malignancies"		0.95	0.35	0.005	0.20	0.94	0.52
t test with "other tumors"		0.12	0.12	0.02	0.82	0.38	0.22
Hodgkin lymphoma	18	27.2 ± 6.6	2.8 ± 1.1	88.5 ± 91.9	33.3 ± 22.9	4.8 ± 4.6	71.6 ± 14.5
Leukemia and non-Hodgkin lymphoma	15	29.5 ± 4.8	3.9 ± 2.2	83.6 ± 60.9	37.7 ± 18.2	8.1 ± 6.3	68.6 ± 10.9
Hematological malignancies in general	33	28.2 ± 5.9	3.3 ± 1.8	86.3 ± 78.1	35.3 ± 20.7	6.4 ± 5.6	70.2 ± 12.8
t test with "other tumors"		0.12	0.45	0.64	0.44	0.36	0.41
Other tumors	16	31.5 ± 8.5	2.9 ± 1.4	75.9 ± 56.7	30.5 ± 18.6	4.9 ± 3.5	73.3 ± 9.9

The group "testicular tumor" comprises the "seminoma", "non-seminoma", and "not classified testicular tumor" groups. The group "hematological malignancies" summarizes "Hodgkin lymphoma" and "non-Hodgkin lymphoma and leukemias".

- Cooling and freezing: After cooling for 8 minutes at 4 °C in the refrigerator, the samples are portioned into Minitüb straws of 0.25 ml each (MTG Vertriebs GmbH, Altdorf, Germany). The straws are numbered and 12 straws are stored bundled in a cassette with the same number. This number is recorded in the patient's file as well as in the database Winsperm®. The samples are frozen using nitrogen vapor cooling in a computer-assisted device (Cryoserv). The frozen samples are transferred to liquid nitrogen.
- Quality control: after cooling to 4 °C and after 24 h storage in liquid nitrogen, sperm count and motility are determined prior to the addition of the cryoprotectant.
- Long-term storage: For storage beyond 4 weeks, the cassettes are transferred to the Cryo-Bank Krefeld (Air Liquide, Krefeld, Germany). This service is provided at the patient's order and expense.

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