

Cancer could return unless stored ovarian tissue undergoes adequate testing before reimplantation

April 22 2008

Cancer patients who have been successfully treated for their disease face the prospect of its return if stored ovarian (or testicular) tissue is transplanted back into their bodies without adequate checks, according to researchers at two university hospitals in Israel.

Writing in Europe's leading reproductive medicine journal, *Human Reproduction*, today, the researchers say that hundreds of cancer patients worldwide have ovarian tissue and, in some cases, testicular tissue frozen in the hope of being able to have children after their cancer treatment has finished; but they warn that few fertility centres have the skills and use the technology needed to check the tissue for residual cancer cells, making it possible for the original cancer to re-infect the body when the tissue is re-implanted to restore the patients' fertility.

"The interest in ovarian tissue storage as a real option for preserving fertility in cancer patients has increased. However, genuine concerns regarding the possible recrudescence [re-appearance] of the primary disease following re-implantation of stored ovarian tissue with malignant cells exist," write the authors.

The first author of the report, Dr Dror Meirow, said: "We think it's vitally important to raise awareness amongst cancer patients, fertility specialists, oncologists and haematologists. There are few fertility centres in the world with the expertise and the technology to run the



types of tests on tissue that are needed to detect residual cancer.

"However, not every reproductive service that has surgical skills and freezing facilities can be safely responsible for ovarian tissue cryopreservation. We suggest that these centres should store tissue for future investigation, and samples can be shipped to specialist centres for analysis."

Dr Meirow, who leads the fertility preservation programme in the IVF Unit at Chaim Sheba Medical Center, Tel Hashomer (headed by Professor Jehoshua Dor), carried out the research with Professor Dina Ben Yehuda, director of the Hematology Division at Hadassah University Hospital, Jerusalem. Dr Meirow said that fertility centres with close connections to cancer and haematological centres should be able to work together in order to adopt the correct methods for checking stored tissue.

Before collecting tissue from the 58 young women in this study, Dr Meirow and his colleagues used various imaging methods (sonography, CT and PET scans) to look for cancer in the pelvis and ovaries of the patients; the women were about to receive chemotherapy for haematological cancers such as Hodgkin's lymphoma, non-Hodgkin's lymphoma and leukaemia, between 1997 and 2007. They found cancer in the pelvic area of two patients, and therefore ovarian tissue was not harvested. They collected tissue from the other 56 patients and, in addition to freezing strips for future transplantation; they also froze a smaller piece of ovarian tissue separately for each patient. They planned to use these extra strips for future checks for the presence of cancer cells, using the most modern methods that would be available at the time the tissue was thawed and prepared for transplantation.

When the ovarian tissue was thawed, they used several different methods to detect minimal residual disease:



-- Histological evaluation (close examination of thin strips of tissue)

-- Immunohistochemical staining (using antibodies to detect abnormal cells)

-- PCR (polymerase chain reaction, which amplifies sections of DNA) and real-time PCR to detect molecular markers that would indicate the presence of cancer cells.

The results were compared with the same tests carried out on the patients' diseased tissue to check that the tests were capable of detecting cancer when it was known to be present.

None of the tests detected minimal residual disease in ovarian tissue for any of the patients, with one exception: modern highly sensitive realtime PCR detected cells in one CML patient where previous tests had not detected any sign of cancer, and so ovarian tissue transplantation was not carried out.

Dr Meirow said: "For this research we concentrated on haematological malignancies, which are common in young patients, but we are working also on solid tumours. Tumour cells of haematological cancers do not often form clumps of cells, which are easier to detect. Therefore, it is highly important to identify single cancer cells among hundreds of thousands of normal cells.

"It is important to use the method and the specific probe that is suitable for each patient, and with this research we had a positive control test from the tumour itself."

The authors suggest that, as methods for detecting cancer cells are improving all the time and ovarian tissue can be stored for more than ten years, tests to detect residual disease should be carried out just before transplantation rather than at the time of collection. To do this, it is necessary to freeze smaller piece of ovarian tissue separately for



minimal residual disease investigation.

Dr Meirow said: "Following our pioneer report in 2005 on pregnancy and delivery post transplantation of ovarian tissue, the next step is to inform patients about the increasing success of ovarian tissue transplantation, to continue to improve the success of ovarian tissue transplantation, but also to call on fertility centres that store ovarian tissue to look for minimal residual disease and to start freezing tissue now for future investigation. All of this holds true for testicular tissue too, although we are not so advanced in successfully removing, storing and transplanting testicular tissue as we are with ovarian tissue."

Citation: Searching for evidence of disease and malignant cell contamination in ovarian tissue stored from hematologic cancer patients. *Human Reproduction* Vol.23, Issue 5. doi:10.1093/humrep/den055.

Source: European Society for Human Reproduction and Embryology

Citation: Cancer could return unless stored ovarian tissue undergoes adequate testing before reimplantation (2008, April 22) retrieved 28 April 2024 from <u>https://medicalxpress.com/news/2008-04-cancer-ovarian-tissue-adequate-re-implantation.html</u>

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