OPINION

Assisted procreation in cases of hepatitis B, hepatitis C or human immunodeficiency virus infection of the male partner

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Guidelines for assisted procreation impose a special responsibility upon physicians for the health of the expected child because of their active role in inducing pregnancy. Therefore, careful clinical evaluation of both partners has to precede every application of these methods. Risks for the mother’s health or the development of the child count as a relative contraindication for a treatment. To balance these relative contraindications, the existing risk factors have to be recognized through screening examination. If a chronic infection occurs in the male partner, prevention for the female partner is theoretically possible by using a condom. As this inhibits a pregnancy, at least in cases of human immunodeficiency virus and hepatitis C virus infections, realization of a pregnancy requires assisted procreation.

The main question in these cases is whether infectious particles can be eliminated by sperm processing to ensure the safe treatment of the healthy female partner.

Key words: assisted procreation/hepatitis B/hepatitis C/HIV/male infection

Introduction

Guidelines for assisted procreation impose a special responsibility upon physicians for the health of the expected child because of their active role in inducing pregnancy. Therefore, careful clinical diagnostics of both partners have to precede every application of these methods, considering all factors for a successful therapy and the child’s health. Risks for the mother’s health or the development of the child count as relative contraindication for a treatment (Bundesärztekammer, 1998).

To balance these relative contraindications, the existing risk factors have to be recognized through screening examinations. In terms of infectious diseases, upon the recommendation of the German Society for Gynaecology and Obstetrics, besides bacterial colonization and infections (chlamydia, treponema and bacterial vaginose) and a lack of immunity to acute viral infections (varicella and rubella), chronic viral infections [hepatitis B and C and human immunodeficiency virus (HIV)] should also be included in screening methods (Weigel et al., 2002). These chronic infections share the fact of being sexually transmittable, though with extremely different contagiosity, and can lead to a lethal disease like acquired immune deficiency syndrome (AIDS) or hepatic cirrhosis, with different probability and latency after chronic progression.

What consequence does a chronic hepatitis B or C infection or HIV have on reproductive medicine? First of all, it has to be differentiated whether the male or female partner is infected. If the infection occurs in the female partner, the main risk is an infection of the unborn child; the healthy male partner is not endangered by using methods for assisted procreation. In case of an infected male partner, the main risk is an infection of the healthy female partner.

Epidemiologic overview

Hepatitis B

Counting approximately $350 \times 10^6$ chronic infections worldwide, hepatitis B is one of the most common infectious diseases. In northern Europe, the prevalence is less than 1%. In Germany and Austria, about $5 \times 10^6$ patients have been infected with the hepatitis B virus (HBV) in their lifetime, about 700000 being chronic hepatitis B surface antigen (HBsAg) carrier.

Active immunization has been available for nearly 20 years. Still there is a rate of new infections of 2500 cases per year, though with declining numbers since 1997.

Outside high-risk collectives, like i.v. drug addicts and homosexuals, the virus is especially spread sexually or vertically with high contagiosity. The estimated risk of infection by blood transfusion is less than 1 in 250000. Men are affected twice as often as women (Wasley and Alter, 2000; Robert-Koch-Institut, 2004).

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In adults, chronic hepatitis B has been of major importance.
develop a liver cell carcinoma (Tsai et al., 1997; Chiaramonte et al., 1999). Chronic infections can be treated with pegylated α-interferon with a success rate of 30–50%, with seroconversion from HBsAg antigen to anti-HBs antigen carrier and loss of verification for virus replication and conversion into an asymptomatic HBsAg carrier. In 10–15%, even complete cure and seroconversion from HBsAg to anti-HBsAg carrier occur. This immune therapy is protracted and has to last for at least 24 weeks and has significant side effects (Preiser et al., 2000).

The importance of an antiviral therapy with lamivudine and adefovir is yet to be evaluated (Marcellin et al., 2003).

**Hepatitis C**

Worldwide, about $200 \times 10^6$ patients have a chronic infection with hepatitis C virus (HCV). The prevalence shows significant regional differences and exceeds from 0.5% in northern Europe to 15% in some regions of Asia and Africa (Wasley and Alter, 2000). In Germany and Austria, there are approximately 500,000 virus carriers (Robert-Koch-Institut, 2004). Because of the high mutation frequency of the diverse HCV genotypes and subtypes, a vaccine cannot be expected in the near future.

In Germany, there are an estimated 8000 new infections per year, with decreasing numbers. Seventy to eighty percentage of infected patients will become chronic virus carrier (Robert-Koch-Institut, 2004). With a latency of 20–30 years, about 20% of these chronically infected HCV carrier will develop hepatic cirrhosis; of this population, 2–6% per year will develop a hepatocellular carcinoma (Tsai et al., 1997; Chiaramonte et al., 1999). The infection is mainly spread parenterally, about 50% due to i.v. drug abuse. The estimated risk of infection by blood transfusion is less than 1 in 1000,000. Because of improved antiretroviral therapy options, the number of newly diagnosed cases of AIDS has halved within the last 10 years and the HIV/AIDS-related deaths decreased by two-thirds. It seems doubtful though whether this positive trend will last, as there is an increase to about 12% currently in the transmission of resistant HIV varieties (Robert-Koch-Institut, 2004). Results of prospective studies show that without treatment, 50% of the infected patients will contract a severe immune deficiency within 10 years. A reliable vaccine will not be available within the near future because of the high-mutation-frequency HIV.

**Assisted procreation with chronically infected male partners**

If a chronic infection occurs in the male partner, prevention for the female partner is basically possible by using a condom. As this inhibits pregnancy, at least in cases of HIV and HCV infections, the realization of a pregnancy does need the procedure of assisted procreation. The main question in these cases is whether infectious particles can be eliminated by sperm processing, such that a safe treatment of the healthy female partner is possible. Table I outlines the most important factors.

**Hepatitis B**

The healthy female partner of a chronically HBV-infected male can be protected reliably by successful vaccination. HBV DNA can be verified in testicular tissue, just like in other tissues (Mason et al., 1993).

### Table I. Virus proof in ejaculate and sperm processing for hepatitis B, hepatitis C and human immunodeficiency virus (HIV)

<table>
<thead>
<tr>
<th></th>
<th>HBV</th>
<th>HCV</th>
<th>HIV</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Proof of virus</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Seminal plasma</td>
<td>Yes</td>
<td>Yes?</td>
<td>Yes?</td>
</tr>
<tr>
<td>Cell fraction</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Sperm’s integrated genome?</td>
<td>Yes</td>
<td>Yes?</td>
<td>Yes?</td>
</tr>
<tr>
<td>Sperm processing</td>
<td>No (vaccination of partner)</td>
<td>Density gradient + swim-up</td>
<td>Density gradient + swim-up + testing</td>
</tr>
</tbody>
</table>

HBV, hepatitis B virus; HCV, hepatitis C virus.

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**Hepatitis C**

Because the HCV is not a DNA virus, unlike the HBV, and has no reverse transcriptase activity, it cannot integrate DNA within infected cells, sperm or embryos (Steyaert et al., 2000).

The verification of HCV in the ejaculate is difficult and interference prone. Apart from this, the viral load is usually low, with only about 50–200 genome equivalents per ml. Therefore, the stated numbers for detection rate in the ejaculate, either as a free virus in seminal plasma or as an integrated genome in leukocytes or in sperm (Hachouel et al., 1985; Davison et al., 1987). Therefore, the possibility of paternofetal transmission has to be at least discussed.

This seems to be, besides an atypical immune response to a previous infection, the most reasonable explanation for recorded infections in children of HBV-negative mothers (Xu, 1992). Wang and his Chinese group described the intrauterine infection of eight fetuses from HBsAg-negative women, in whom the nucleotide sequence analysis showed a very high homology to the fathers' HBV genotype (Wang et al., 1999).

This leads to the assumption that infected sperm transfer the integrated viral genome to the oocyte and that the child will be infected at the moment of procreation. Until this assumption is proved or disproved by systematic evaluation, couples with an HBsAg-positive male partner should be counselled of this possibility before assisted procreation.

**HIV infection**

It is known that HIV is sexually transmitted and that the virus is excreted with the ejaculate. The risk of transmission correlates with the viral load and can reach 1% per unprotected intercourse (Chakraborty et al., 2001). HIV in the ejaculate has been examined extensively within the last years. HIV, or better its genome, can be verified in white blood cells and cell-free seminal plasma (Baccetti et al., 1991; Anderson, 1992).

Sperm as a conductor are discussed controversially. By using in situ PCR, proviral DNA in spermatogonia, spermatocytes and more rare cases in spermatid were verified histologically in testicular tissue of HIV-infected patients (Nuovo et al., 1994; Muciaccia et al., 1998). Newer research results could not affirm these results (Pudney et al., 1998). Apparently, this clonal infection would not lead to infectious sperm. Few isolated hybridization signals for viral genome have been found in dead or immobile sperm after sperm separation but not in motile sperm, whether using in situ PCR or high-sensitive PCR (Scofield et al., 1994; Quayle et al., 1997).

By analysing ejaculate components by density-gradient centrifugation, HIV RNA can be detected in sperm fractions (Dulioust et al., 1998). But analysing motile sperms by using diverse electro-optical or molecular–biological methods does not verify viral particles or virus genome (Brechard et al., 1997; Quayle et al., 1997; Pudney et al., 1998). With the current knowledge about the possibility of a viral transmission, an association between HIV and mature, vital sperm cannot be constructed. Additionally, Nicopoullos et al. (2004) showed that sperm parameters of HIV-positive men are significantly impaired compared with those of HIV-negative controls, and this correlates with CD4 cell count. They found that the use of antiretroviral medication and low HIV RNA serum virus load measurements are factors that improve intrauterine insemination (IUI) outcome. In contrast to these findings, newer results from Garrido et al. (2005) did not show a correlation between semen parameters, blood CD4 levels and viral load levels.

As the coitus condomatus is an obligate measure to protect the healthy female partner in an HIV-discordant partnership, it is fair to speak of a special form of andrological sterility, concerning the wish for pregnancy, in this situation. Several possibilities are open to these couples. Adoption or insemination with donor sperm has no risk of infection but bears the disadvantage of not using the man’s gametes. Unprotected intercourse on the day of ovulation or recourse offers a decreased risk but is an unsafe method and contradicts totally against

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In situ spermatogonial cells, spermatocytes, spermatids and Sertoli cells show hybridization signals for viral genome sequences (Lang, 1993). Viral DNA can also be found in the ejaculate, either as a free virus in seminal plasma or as an integrated genome in leukocytes or in sperm (Hachouel et al., 1985; Davison et al., 1987). Therefore, the possibility of paternofetal transmission has to be at least discussed.

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The verification of HCV in the ejaculate is difficult and interference prone. Apart from this, the viral load is usually low, with only about 50–200 genome equivalents per ml. Therefore, the stated numbers for detection rate in the ejaculate of viraemic men differ significantly from 0% (Semprini et al., 1998). Today, we have to proceed on the assumption that infected sperm transfer the integrated viral genome to the oocyte and that the child will be infected at the moment of procreation. Until this assumption is proved or disproved by systematic evaluation, couples with an HBsAg-positive male partner should be counselled of this possibility before assisted procreation.

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public health campaigns and cannot be recommended. The last possibility in these cases is medically assisted procreation. These couples can be offered an assisted procreation in co-operation with HIV-specialized physicians, infection control scientists and under defined conditions by using specially processed sperms. According to German–Austrian recommendation, a sequence of density-gradient centrifugation–washing–swim-up should be used in these cases (Weigel et al., 2002; Weigel, 2003).

Every specimen should be tested with highly sensitive gene-amplification techniques to exclude a virus contamination before any medical reproductive procedure is performed. The experiences of Weigel (2003) state that about 3% of the processed specimen show traces of viral RNA. Proviral DNA, however, is obviously being eliminated by the described technique. Other groups report positive reactions of processed specimen in 5–6% by using high-sensitive RT–PCR (Marina et al., 1998; Leruez-Ville et al., 2002).

An earlier processed and tested specimen should not be considered reliable as virus load, and spreading in ejaculate is not constant (Weigel, 2002). Normally, cryopreservation and storage of the specimen are necessary until the test results arrive, which can lead to a loss of sperm motility. Because semen analysis of HIV-infected patients is often restricted, in many cases ICSI is the only therapeutic option left open to couples. This also happens to be the safest method. Bujan et al. (2004) performed IUI on 56 couples, of which all men were HIV positive. Samples were processed by using a combination of density-gradient centrifugation, washing and swim-up, and all HIV tests of the female partners were negative. Garrido et al. (2004) reported pregnancy rates of 40–48% per cycle using sperm washing, nested PCR and ICSI. Without using PCR analysis, Sauer et al. performed ICSI in 34 serodiscordant couples and did not see one HIV infection. Regarding the necessity of a sperm wash and subsequent molecular determination of the absence of the virus previous to its use in fertilizing oocytes, the authors state that it is not possible to guarantee that all viral particles are absent after sperm preparation. Nonetheless, the correct way to resolve the limitations of the available assays will never be avoiding the test in the semen.

Both partners always have to be informed that even if specimens are processed and extensive testing methods are used, a possible infection of the partner or child cannot be completely ruled out. In contrast to the potential risk of unprotected intercourse, this risk is only hypothetical and cannot be quantified. Apart from that, after 10 years of assisted procreation, there is not one known case of horizontal or vertical transmission after adequate preparation. Under the notice of the described procedure as well as a complete documentation, ‘a scientific causality between therapy and infection would not be adequate’ (Eberbach, 1999) in terms of legality and any objections against a treatment arbitrary.

Summary

If a couple is scheduled for assisted procreation and the male partner is infected with a chronic persistent virus, considerations concerning reproductive medicine and infection control have to be made. These include the progress of the infection, the risk of an infection for the female partner and the unborn child and strategies for prevention.

After balancing the medical, ethical and legal aspects, the conclusion depends on the kind of infection and which partner is affected. In cases of HIV-infected male partners, there is an indication for medical assisted procreation. Through adequate processing techniques, motile sperm can be separated and can be used for IUI, IVF or ICSI after testing them with high-sensitive methods.

Even though today’s standards eliminate medical and legal doubts, treatment is limited to a few centres. As HCVs can be verified in the ejaculate, risk reduction for the female partner can be achieved by offering a treatment with specially processed sperm.

Concerning hepatitis B infections, the female partner can be protected successfully by vaccination.

References


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