J. Perinat. Med. 41 (2013) 17–22 • Copyright © by Walter de Gruyter • Berlin • Boston. DOI 10.1515/jpm-2012-0097

Review article

Perinatal risks after IVF and ICSI

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Abstract

Pregnancies that occur after infertility treatment, particularly after assisted reproduction, constitute high-risk pregnancies. Occurrences of conditions such as high blood pressure, preeclampsia, growth retardations and bleeding are higher in comparison with the norm of spontaneously entered pregnancies. The rate of premature births and the frequency of intrauterine deaths are much higher than the average for all pregnancies. Furthermore, pregnancies resulting from *in-vitro* fertilisation (IVF) have significantly higher rates of requiring induced labour or caesarean section. However, it is to be assumed that these complications and unfortunate developments are not caused by extracorporeal fertilisation itself, but rather are due to the frequency of multiples and to the risk factors of the women involved. These women are, on average, older and there are often more problems with cycle irregularities, uterine anomalies and obesity than in the total collective of all pregnancies. The methods of modern reproductive medicine often bring a higher rate of multiple pregnancies. The clinical problem of multiple pregnancies is, above all, the raised rate of premature births and intrauterine growth retardations that contribute to the significantly higher rate of morbidity and mortality for these children. The slightly higher rate of congenital defects after IVF and intracytoplasmic sperm injection (ICSI) are also attributed more to the risk profile of the parents and less to the techniques themselves. The most important and easy-to-avoid complication is the multiple pregnancy, and it should be our goal to lower this rate even further.

Keywords: Assisted reproduction; infertility treatment; multiple pregnancy; perinatal risks.

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Introduction

In 1978, the first baby to be conceived through artificial insemination, Louise Joy Brown, was born in England. It was a high-risk birth because the mother had developed preeclampsia. The obstetrician Steptoe did not want to risk a normal birth and decided to deliver the baby through a caesarean section at 38 weeks. The underweight baby girl was born weighing 2700 g at birth [37].

This example highlights the fact that pregnancies made possible through assisted reproduction techniques are associated with additional medical risks. Worldwide, 72.4 million women are currently infertile [7]. In Germany, it is assumed that two million couples are unintentionally without children: every seventh couple hopes in vain for a child. Worldwide, about 40 million couples take advantage of reproductive medicine. About 12,000 children per year are born in Germany after assisted reproduction treatment (ART) [13].

In the last 10 years, the number of the treatment cycles has risen tremendously worldwide, which has in turn resulted in a rising number of multiple pregnancies and births. This trend has been attributed to the increased use of assisted reproduction methods, expressed most strongly in multiple births, and also to the rise in age for giving birth. In the last 30 years, the average age of first-time mothers has risen by 5 years, from 24 to 29.

The progress of pregnancies that occurred after reproductive medicine techniques has meanwhile been documented in numerous surveys. Generally, a pregnancy after *in-vitro* fertilisation (IVF) or related procedures is considered an at-risk pregnancy. Occurrences of conditions such as high blood pressure, preeclampsia, growth retardations and bleeding are higher than in comparison with the norm of spontaneously-entered pregnancies. The rate of premature births and the frequency of intrauterine death is much higher than the average for all pregnancies. Furthermore, pregnancies post-IVF have significantly higher rates of requiring induced labour or caesarean section. Very often labours are induced or caesarean sections are performed as patients or doctors urge induction or labour or caesarean section because they are both anxious about the outcome.

In addition, the incidence of miscarriage and tubal gravidity is above average in pregnancies that occur after reproductive treatment procedures. The increase in risk of complications in pregnancy and to the health of the children could be due to the following factors: influence of the milieu of the follicles and the egg cell quality through ovarian stimulation, influence of sperm function through exposition when confronted with substances in sperm preparation, manipulation of the fertilisation procedure through IVF and ICSI, use of genetically abnormal sperm from subfertile men or imprinting defects. However, it is to be assumed that these complications and unfortunate

developments are not caused by extracorporeal fertilisation itself, but rather are due to the frequency of multiple pregnancies and to the risk factors of the women involved. These women are older on average, and there are often more problems with their menstrual cycles, uterine anomalies and obesity than in the total group of all pregnancies.

Ovarian hyperstimulation syndrome

One of the first complications that can arise during a pregnancy is ovarian hyperstimulation syndrome (OHSS). This syndrome involves a serious complication of gonadotropin treatment, which only occurs under the influence of exogenous or endogenous hCG and is thereby limited to the lutheal phase and the first weeks of the pregnancy. Patho-physiologically, a higher level of capillary permeability is responsible for most of the changes. Main symptoms are multiple lutein cysts in enlarged ovaries, ascites and pleural effusions, abdominal pains and haemoconcentration. The hyperstimulation syndrome is divided into different stages according to its level of seriousness [19]. Stage III occurs in 0.2% of all IVF/ICSI treatments, and it is a complication to be taken very seriously due to the risk of thromboembolic occurrences [13].

Single pregnancies after ART – risks

Further along in pregnancy, the most important risk factor after treatment for sterility is the occurrence of multiple pregnancies, but single pregnancies are also associated with complications – as revealed by the example of Louise Joy Brown. As a meta-analysis of 25 studies published in 2004 reveals, the risk of a premature birth or a birth weight of <1500 g after assisted reproduction is raised by a factor of three in comparison with spontaneously conceived children. The resulting rise in perinatal mortality is by a factor of 1.7 [24].

A large American study with 40,000 IVF children vs. 3 million spontaneously conceived children reveals that the risk for a low birth weight after assisted reproduction is more than twice as high [36]. Interestingly, this effect was not observed in children that were carried by surrogate mothers; in these cases, normal pregnancy progressions were observed. The significantly higher rate of premature births after IVF treatment is clearly for the most part due to multiple pregnancies, but it also twice as high for single pregnancies, at 13%–16%, than it is for regular pregnancies [22, 36].

As mentioned above, it is to be assumed that these complications and unfortunate circumstances are caused by subfertility as a risk factor *per se*. Studies conducted by Williams et al. [41] and Basso and Baird [4] have shown that pregnancies that have taken over a year to initiate have a higher risk for lower birth weights, fetal retardation and caesarean section than pregnancies that were initiated quickly.

This confirms Pandian's study, which proves a higher obstetric risk from patients with idiopathic sterility, independent from the treatment, whether it be hormone stimulation, insemination or IVF, in comparison to a fertile control group [31]. In mothers with idiopathic sterility, 498 single pregnancies were compared with 32,969 control group children. The obstetric databank of Aberdeen from 1989 to 1999 was used as a basis for this comparison. It was shown that complications, such as preeclampsia, abruptio placentae, early contractions, emergency section and inducement of labour occurred at a significantly higher rate than in the control group [31].

In another study, single pregnancies after IVF were compared with the normal obstetric population of England and Wales [15]. A rise in the rate of premature births from 6% to 13% was revealed, as was a rise in the incidence of children with low birth weights from 7% to 11% and a rise in the probability for a lower birth weight, as well as small for gestational age (SGA) from 10% to 17%. It is interesting in this study that the risk in a single pregnancy for fetal retardation and SGA is higher as more embryos are transferred.

In summary, single pregnancies that occur through assisted reproduction methods are at higher risk for obstetric problems in comparison with other single pregnancies, but are still significantly lower than for multiple births.

Multiples after ART – risks

The methods of modern reproductive medicine inherently are associated with higher rates of multiple pregnancies. This is because the use of medicated ovulation induction or controlled ovarian hyperstimulation more often leads to polyovulation than a natural cycle would. For artificial insemination methods such as IVF or ICSI, whereby embryos are transferred to the uterus, the risk of a multiple pregnancy directly depends on the number of transferred embryos. According to the German Embryo Protection law, the maximum number of embryos that can be transferred is limited to three, but the guidelines of the "Bundesärztekammer" in Germany recommend that women under 38 years of age only transfer two embryos. Due to a mostly constant pregnancy rate, the frequency of multiples for the pregnancies registered at the German IVF Registrary (DIR) in the last few years has fortunately decreased, a fact that can be attributed to the restrictive transfer policies for elective transfer of only two embryos for women under 35 years. In the last few years, on average, two embryos were transferred, which led to a multiples rate of over 20% [13].

In many other countries, fertilisation medicine is somewhat more liberal. The decision of how many embryos should be transferred is determined according to the current state of the medical research and experience of how many embryos are necessary for a medically supported fertilisation to have a good chance of a positive outcome.

Not all multiple fetuses reach birth because, depending on the age of the woman, 30%–50% of all multiple pregnancies result in spontaneous regression or miscarriage of one or all fetuses. The probability that for twins one of the fetuses will not develop further is at 30%, but if a heartbeat can be detected in both fetuses, this drops to 10%–15%. Also, in the case of triplets, in about 30%–50% of cases, the retardation of one of the fetuses is likely to occur. Early diagnosis of multiple

gestations can therefore not immediately be assumed to be an intact multiple pregnancy.

Aside from the specific risks connected with multiples-associated syndromes, the clinical problems of multiple pregnancies, particularly the higher rate of premature births and intrauterine growth retardation, are present and significantly influence the higher rate of morbidity and mortality for these pregnancies. In the Bavarian perinatal statistics from 2010, a perinatal mortality rate of 4.2 for single pregnancies and 16.6 for multiple births was recorded [5].

In comparison with single pregnancies, twin and higher grade multiple pregnancies are associated with a higher level of maternal morbidity and a higher rate of perinatal morbidity and mortality. The rate of complications rises with the number of fetuses, as shown in Table 1 [2]. The most significant maternal complications are bleeding before the birth and postpartum, anaemia and pregnancy-induced hypertension and preeclampsia. The significant fetal complications are intrauterine growth retardation, discordant growth, death of a fetus and fetofetal transfusion syndrome. For multiple pregnancies, there is also a higher risk of general obstetric complications with premature contractions or rupture of the membranes, premature birth, underweight newborns and the placenta being released too early.

Not least, the birth itself is more risky than for a single pregnancy. For twin pregnancies, there is a significantly higher incidence of intrapartum deaths.

In light of the overall high rates of morbidity and mortality of high-grade multiple pregnancies, selective reduction presents an alternative. In this case and for multiple pregnancies and the often observed multiple pregnancy-associated pathologies, psychological care becomes of the utmost significance.

Aside from a higher rate of premature births and retardations, multiple pregnancies have a higher rate of miscarriage and, in comparison with single pregnancies, have a higher risk of chromosomal problems in the fetus. If the parents decide to have an invasive prenatal diagnostic test done, the miscarriage risk rate for this procedure is higher than for single pregnancies. The most important maternal complications for multiple pregnancies are bleeding during pregnancy and also postpartum bleeding, anaemia, pregnancy-induced hypertension and preeclampsia.

After assisted reproduction methods have been used, not only is the rate of dizygotic multiple pregnancies higher, but also the rate of monozygotic pregnancies. After spontaneous conception, this happens in 0.4% of all births. Possible explanations for this are delay of early embryo development and damage to the pellucida zone, which could lead to a pathological tying of blastocyst rates. The rate of monozygotic multiple pregnancies has an incidence rate 2–12 times greater in artificially fertilised pregnancies than in the rest of the population [1, 35]. Risk factors include microinjection procedures for a maternal age over 35. The monochorionic multiple pregnancies that result more often from monozygotic multiple pregnancies than for spontaneous conception are a particular danger due to fetofetal transfusion syndrome [1].

Many studies have shown that for multiple pregnancies after assisted reproduction, the rate of birth weights <2500 g and 1500 g are not higher than for spontaneously conceived pregnancies [24, 29, 36]. The rate of premature births and fetal retardations as well as perinatal mortality is not higher for IVF twins when compared to spontaneously conceived twins, and this has been proven by many studies.

This contrasts with single pregnancies for which the risk of low birth weights after assisted reproduction is more than twice as high than for spontaneous pregnancies.

As for twins and triplets conceived through IVF or simple stimulations, there are no significant differences concerning pregnancy length and birth weight when compared to spontaneously conceived triplet pregnancies [18].

Single embryo transfer

As the numbers from the German IVF Register of 2010 reveal, the rate of twin pregnancies from the transfer of two embryos is still between 16% and 22% [13], so that generally a transfer should involve only one embryo. The transfer of a single embryo only makes sense, however, when the embryo with the best development potential can be selected from a cohort of embryos or blastocysts. The advantage of the single embryo transfer (SET) is obvious – for acceptable pregnancy rates, the multiple pregnancy rate can be lowered to the same level as through natural conception. The numbers from Scandinavia have shown that routine SET does not lead to a decrease in the cumulative birth rate, but that the risk of multiple pregnancies is almost completely eschewed [32]. The debate about whether after single embryo transfer the birth rates are the same as after double embryo transfer is not closed because the data are conflicting. The average implantation rate of an embryo after 2 days in culture is about

Table 1 Comparison of perinatal outcomes of twins, triplets and quadruplets [2].

	Twins	Triplets	Quadruplets
Birth weight	2347 g	1687 g	1309 g
Weeks at delivery	35.3	32.2	29.9
Small for gestational age	14%–25%	50%–60%	50%–60%
NICU admissions	25%	75%	100%
Risk of cerebral palsy	4-fold increase	17-fold increase	
Risk of death in 1 year	4-fold increase	20-fold increase	

Data taken from ACOG Practice Bulletin no. 56 [2].

12%. This is dependent mostly on the age of the woman and the embryonic morphology but also on many other factors. The rate of clinical pregnancies, however, rises with the number of embryos transferred (see Figure 1). However, it is necessary to take into account that for patients who have had only one embryo transferred, this was in most cases the only one that was available and for that reason alone has a weaker prognosis. After the transfer of two embryos on day 2 or 3 that have been developed from exactly two zygotes on day 1 in accordance with the law in Germany, a lower rate of pregnancy can be expected than after the elective transfer of two embryos that could be selected from a group of multiple embryos. Because the rate of clinical pregnancies after the elective transfer of two embryos is only lower than after the transfer of three embryos by a few percentage points, it is generally advised that women with good prospects for becoming pregnant limit the number of transferred embryos to a maximum of two. Through the reduction of the multiples rate through ART with the elective transfer of only two embryos, it has been proven that the frequency of premature births and children with low birth weights of under 2500 g as well as the length and costs of neonatal intensive care could be significantly reduced.

A large very current meta-analysis had the goal of comparing pregnancies after elective single embryo transfer (eSET) with pregnancies after double embryo transfer (DET) and with spontaneously conceived pregnancies [21]. It was revealed that pregnancies after eSET in comparison with DET pregnancies had a significantly lower risk in regard to premature birth as well as low birth weights; however, eSET pregnancies compared with spontaneously conceived pregnancies revealed a significantly higher perinatal risk profile.

Fetal malformations after ART

The safety of assisted reproduction methods with regard to the health of the children born is attested in numerous studies. Intracytoplasmic sperm injection (ICSI) is a very invasive procedure, but has become an established procedure worldwide for treating acute male subfertility. The procedure involves injecting one single sperm into the egg cell.

Earlier studies did not find a rise in the malformation rate after ICSI [10, 11, 30, 38, 40]. However, the number of cases was either very low or there was no control group or the investigation technique was not performed in a standard fashion. More recent studies that have been summarised in a good literature overview give evidence of a slightly higher dysplasia risk after ART in comparison to spontaneously conceived pregnancies [6]. In particular, a multi-centre study should be highlighted in which all pregnancies were analysed in their 16th week after ICSI independently from their outcome [27]. In total, 3372 ICSI children and fetuses were examined. As a control group, 8016 children who were spontaneously conceived were used and were recorded within the parameters of the dysplasia monitoring of Saxony Anhalt (Germany). The primary outcome parameter of the study was the rate of major malformation.

The malformation rate, 8.9% for the ICSI singles vs. 6.0 for the control group, is significantly higher, that is, in every 12th pregnancy after ICSI and only in every 15th pregnancy after spontaneous conception does dysplasia take place. Furthermore, here again, differences were observed in the length of carrying time, birth weight and the caesarean section rate (33% vs. 13%). For multiples, no differences were observed in this study, neither in the dysplasia rate nor in other obstetric parameters, between ICSI and spontaneously conceived multiples.

As regards the type of dysplasia, no specific malformation pattern could be observed post-ICSI. The relative risk was primarily for gastrointestinal and urogenital dysplasia. A higher risk of genital dysplasia, e.g., hypospadias, as in other studies was only marginally evidenced here [27].

In this study, it was shown that the following factors influenced the occurrence of malformations: the mother's age, dysplasia in the parents, a stillborn child, or siblings with dysplasia. It should be assumed that through the sperm injection procedure itself, there is no measurably higher risk of

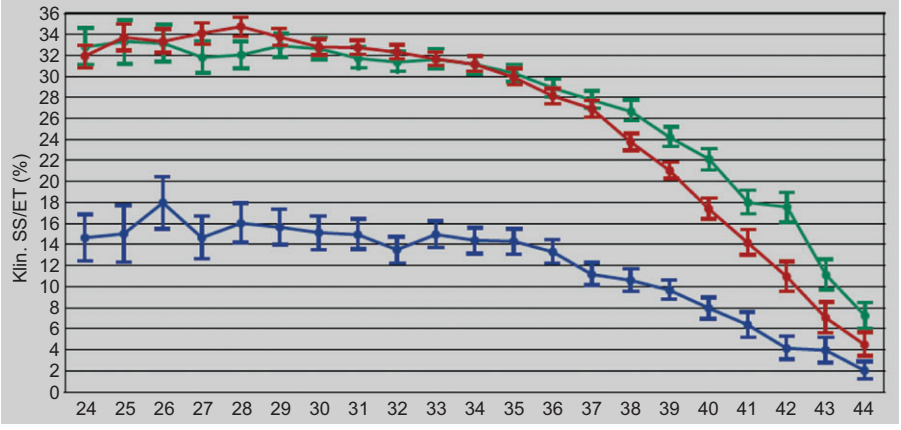

Figure 1 Clinical pregnancies by number of transferred embryos and the age of the woman after IVF and ICSI.
Blue, one transferred embryo; red, two transferred embryos; green, three transferred embryos. Data from the German IVF Registry 2007 [14].

fetal malformations being induced, but rather that the slightly higher risk is due to the parent's risk factors. The seriousness of male subfertility does not play a relevant role. There was no connection observed between the dysplasia rate and the number or origin of sperm.

Many studies have evidenced that the malformation rate even after conventional IVF is slightly higher when compared with spontaneous conception [3, 23]. In studies and meta-analyses in which the malformation rates of IVF and ICSI were compared, there was no difference observed in the dysplasia rate for ICSI compared to the rate for conventional IVF [8, 20, 33].

A very recent Australian study investigating the rate of birth defects after conception through ART and after spontaneous conception (patients with and without a history of infertility) showed an increased risk of birth defects after ICSI [12].

In terms of chromosome anomalies, a study of 1586 ICSI fetuses showed that the rate of chromosomal anomalies after ICSI was higher [9]. In a total of 47, 3%, obvious chromosome sets were detected, which for an average maternal age of 33.5 years represents a significant increase. Particularly the aneuploidy rate of the sex chromosomes seems to be slightly higher after the microinjection (0.2% vs. 0.6%), which cannot be attributed to the technique, but rather to initial problems with the spermiogenesis.

Men with serious oligozoospermia or azoospermia represent a genetic risk group where up to 5% chromosomal aberrations can be observed [39]. It could be shown that paternal age also has an influence on genetic abnormalities in sperm that can lead, at a paternal age over 40, to reduced fertility and an increased rate of adverse outcome in the offspring [34]. However, even when the men themselves show no symptoms, the sperm from men with OAT syndrome more often have chromosomal anomalies. It is to be assumed that through the sperm injection itself, there is no higher genetic risk for a pregnancy, but rather the eventual genetic anomaly from the sperm used for the injection is brought along.

As regards the safety of new methods such as *in-vitro* maturation, it is not yet possible to state conclusively. Because ovarian stimulation therapy, which today is standardly performed with exogenous gonadotropins, carries a high risk of ovarian overstimulation syndrome for some women, *in-vitro* maturation (IVM) has been performed for some time. In the IVM process, small antral follicles that are about 5–10 mm in size are punched out of the ovaries transvaginally, and the immature oocytes inside are brought to maturation *in vitro* through supplementation of gonadotropins to the culture, and subsequently are fertilised through ICSI. The significant advantage of these methods is the complete absence of risk for ovarian hyperstimulation syndrome. Moreover, the method is less costly in terms of time and money (fewer medicines, no costly monitoring). In the meantime, several thousand children have been born; however, at this point it is not possible to attest to the safety of the method.

There is a debate about whether culture media have an influence on birth weight. There are data not showing any association between embryo culture medium and birth weight following IVF [17], whereas others showed differences in birth weight after different culture systems [16]. The possibility that, through the cultivation of egg cells, multiple imprinting defects will arise cannot be excluded [25]. In the last few years, some case studies and control studies have been published that reveal a higher risk for certain imprinting errors. Some studies could demonstrate a multiplication of the risk for Beckwith-Wiedemann Syndrome or Angelman Syndrome after fertilisation therapy [26, 28]. However, despite multiplication of the risk, the absolute risk remains very low because these generally are very rare diseases. It must still be observed whether subfertility or the therapy itself leads to this increase in risk.

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The authors stated that there are no conflicts of interest regarding the publication of this article.

Received May 7, 2012. Revised June 20, 2012. Accepted July 13, 2012. Previously published online August 18, 2012.