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Review: Toxicants in reproductive fluid and in vitro fertilization (IVF) outcome

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Abstract

Some of the physical, chemical, dietary, occupational and environmental factors are having adverse effect on human reproduction. Increasing trend in reproductive disorders in recent years at least in part might be associated with these factors. The data available suggests less success rate of in vitro fertilization (IVF) outcome of parents exposed to some of the reproductive toxic chemicals as compared to parents who were not exposed to such chemicals. However, data are very meager and require more studies as some debatable data also exists. But existing positive findings encourage in advising that sub-fertile subjects, who are planning to go for the IVF, should reduce toxic exposure well in advance by adopting positive life style and work environment. Further, clinician ought to be aware of occupational and environmental exposure history of the participating couple.

Keywords

environmental and occupational factors, reproductive impairment, semen quality, male and female reproduction, IVF

Introduction

The role of environmental, lifestyle and occupational factors might be associated with the increasing trend of certain diseases including reproductive disorders. Approximately 15% of the sexually active populations are affected by clinical infertility and in 50% of cases, a male factor is involved, either as a primary problem or in combination with a problem in the female partner (Pasqualotto et al., 2005). There are reports which indicated that certain environmental chemicals have adverse effects on human reproduction. Kumar (2008) mentioned that evidence of the endocrine disrupting chemicals (EDCs) on the reproductive health disorders and related outcomes, such as deterioration of semen quality, increasing pattern of cryptorchidism, hypospadias, testicular and prostate cancer in the male, and breast cancers and endometriosis, menstrual disorders, etc. in the female and genital defects in both sexes, has accumulated in recent decades steadily. However, more studies are needed with regard to relationship between exposure level and affected parameters.

After the introduction of in vitro fertilization (IVF) clinically, the sub-fertile couple opts for IVF to have children. It is reasonable to believe that success rate of IVF depends upon various factors along with the reproductive health of the parents. Weiss et al. (2006) carried out a study in Tanzania and Germany and reported that the distribution of toxins between agricultural and industrial countries is different and in subjects with high serum concentrations of DDT and DDE, pregnancy rates were impaired. Olivennes et al. (2002) reported that only few studies have addressed the issue of the perinatal outcome of IVF

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Table 1. Chemical in seminal plasma and follicular fluids

SN	Chemical	References
Seminal plasma		
1	Na, Mg, P, K, Ca, Fe, Cu, Zn, Se, Rb, Sr elements were detected in all seminal plasma samples and V, Mn, Co, As, Mo, Cd, Sn, Ba were detected in over 75% of the samples	Haruo (2004)
2	Lead	Benoff et al. (1998)
3	Lead and cadmium	Mankad (2007)
4	Cd	Wu et al. (2008)
5	Mercury	Matthies (1989)
6	Nicotine and/or cotinine	Pacifici et al. (1995); Vine et al.(1993); Zenzes et al. (1999)
7	Trichloroethylene and its metabolites chloral and trichloroethanol were detected in the seminal fluids of mechanics exposed to TCE	Poh-Gek et al. (2003)
Follicular fluids		
1	Cotinine	Weiss and Eckert (1989); Rosevear et al. (1992)
2	B[a]P	Neal et al. (2008)
3	PCBs with 3-7 atoms of chlorine	Drbohlav et al. (2005)
	p,*p*′–DDE, mirex, hexachloroethane, 1,2,4-trichlorobenzene, PCB49, PCB153, PCB180. Cd and cotinine	Younglai (2002).
4	Bisphenol A	Ikezuki et al. (2002)
5	PCBs, most abundant congeners (PCBs 138, 153 and 180), and 1,1-dichloro-2,2-*bis*(4-chlorophenyl)-ethene (*p*,*p*′-DDE) and HCB	Felip De et al. (2004)
6	Cadmium	Zenzes et al. (1995)
7	Polychlorinated dibenzodioxins (PCDDs) and dibenzofurans (PCDFs)	Tsutsumi et al. (1998)

pregnancies and of the children's development and well-being. They mentioned that an analysis of the multiple risk factors involved in these complications is needed. Age and parity may be important factors. Recently, Klonoff-Cohen (2005) mentioned the importance and efficacy of IVF by the fact that >1 $\times 10^6$ babies have been born to infertile couples since clinical introduction of IVF in 1978. They further reported that the parents' contribution has virtually been ignored when considering aspects that influence the success rates. Younglai et al. (2005) reviewed the work on environmental and occupational factors affecting fertility and IVF success and highlighted the paucity of data on the exposure of couples to environmental insults and the association with IVF success, and the problems associated with the interpretation of such data sets. The authors made an attempt to compile the recent data available pertaining to the role of various toxicants and IVF outcome as well as toxicants' levels in reproductive fluid, that is, follicular (FF) and seminal plasma.

Material and methods

The present overview is based on the literature searched through various database such as Pubmed, Goggle, Toxnet, and by consulting various journals and analyzed pertaining to the role of reproductive toxicants and IVF outcome along with data on toxicants' levels in reproductive tract fluid. Some data were also incorporated related to association of certain chemicals and reproductive impairments including relevant experimental studies. The summarized data are presented in Tables 1 and 2.

Toxicants level in follicular fluid

Various exogenous as well as endogenous factors influence reproductive health, pre- and postnatal development. A number of reports are available on association between exposure to certain persistent environmental and industrial chemicals and reproductive health impairments from animal and human studies. There are reports which indicated that various

Table 2. Reproductive toxicants and IVF outcome

SN	Chemical/ factors responsible	Reported effect	References
1	Lead	Increased seminal plasma lead level were negatively correlated with IVF rates.	Benoff et al. (2003)
2	Lead	A negative correlation between seminal plasma Pb and percentage of sperms with normal DNA	Mankad (2007)
3	p,p′-DDE	Associated with failed fertilization in IVF	Younglai et al. (2002)
4	Lead	Lead levels within FF found to be higher in non-pregnant patients compared to pregnant patients	Silberstein et al. (2006)
5	In utero exposure to DES	A significantly impaired implantation rate following IVF, and the outcome of ART remains poor	Pal et al. (1997)
6	B[a]P	Higher level in FF associated with not conceiving pregnancy	Neal et al. (2008)
7	Smoking	Negatives influence on IVF outcome	Klonoff-Cohen (2005)
8	Psychological stress, consumption of caffeine and alcohol, and illicit drug use	Implicated in a poorer IVF outcome, but evidence is inconclusive	Bellver (2008)

toxicants are present in the seminal plasma and/or follicular fluids (FF). These toxicants in reproductive fluid may compromise the reproductive outcome. Drbohlav et al. (2005) explored the possibility of detection of toxic polychlorinated biphenyls in blood and FF of infertile women undergoing IVF + ET (in vitro fertilization and embryo transfer) program and found all Polychlorinated biphenyls (PCBs) with 3–7 atoms of chlorine in these biological fluids. Earlier, Younglai (2002) collected the blood and FF and seminal plasma from the couple who underwent IVF program. The contaminants most frequently found in FF, more than 50% of the sample tested, were *p, p′*–DDE, mirex, hexachloroethane,1,2,4-trichlorobenzene, PCB49, PCB153 and PCB180. Cadmium and cotinine were detected in eight and eighteen of twenty-one FF samples, respectively. Ikezuki et al. (2002) detected bisphenol-A, which is an estrogenic endocrine disrupting chemical, in human serum and FF at approximately 1–2 ng/mL and reported to affect preimplantation embryos or fetuses and alter their postnatal development at doses typically found in environment. Felip De et al. (2004) reported that with respect to PCBs, the sum of the three most abundant congeners (PCBs 138, 153 and 180) was 1230 ng/g. PCDDs, PCDFs, *p,p′*-DDT and 1,1-dichloro-2, 2-*bis* (4-chlorophenyl)-ethane (*p,p′*-DDD) were below their determination limits. 1,1-dichloro-2,2-*bis*(4-chlorophenyl)-ethene (*p,p′*-DDE) and Hexachlorobenzene (HCB) were detected in concentrations, respectively, in the order of 700 and 70 ng/g, lipid basis in FF.

Tobacco smoking is a well-established reproductive hazard that has been linked with decreased fertility in both smokers and passive smokers. Neal et al. (2008) mentioned that the chemical components responsible for the reproductive toxic effects of cigarette smoke are unknown. They measured the levels of benzo[a]pyrene (B[a]P) and other polycyclic aromatic hydrocarbons (PAH) in cigarette smoke, in the serum and in FF. Women exposed to mainstream smoke had significantly higher levels of B[a]P in FF compared to side stream exposed or their non-smoking counterparts. Zenzes and Reed (1989) determined whether there is variation in levels of FF cotinine between the two ovaries of women undergoing IVF-ET who are exposed to cigarette smoke and found that cotinine uptake between the two ovaries of a woman might differs approximately one-fourth of the time. In spite of these differences, the overall correlation between ovaries is high. The clear distinction in levels of FF cotinine among active, passive and nonsmokers demonstrates the reliability of FF cotinine testing. Detection of cotinine in a large proportion of nonsmokers shows how pervasive nicotine is in the environment. Cotinine has been detected in FF of women undergoing assisted conception (Rosevear et al., 1992; Weiss and Eckert, 1989). Further, Zenzes et al. (1995) found that mean level of FF

cadmium was higher in smokers than in nonsmokers, and with a dose effect of smoking.

Toxicants in seminal plasma

Number of toxicants are present in the seminal plasma. Eleven (Na, Mg, P, K, Ca, Fe, Cu, Zn, Se, Rb, Sr) elements were positively detected in all seminal plasma samples and another eight elements (V, Mn, Co, As, Mo, Cd, Sn, Ba) were detected in over 75% of the samples by Haruo (2004). Benoff et al. (1998) observed that greater than 40% of subjects who were even not exposed to lead (Pb) in their workplace and who did not smoke cigarettes exhibited blood and seminal plasma Pb concentrations that were above the permissible limit in men non-occupationally exposed to Pb. Both Pb and cadmium were reported to be found in seminal plasma and had negative relationship with semen quality parameter (Mankad, 2007). Wu et al. (2008) reported that in smokers, each tenfold increase in semen cadmium concentration was associated with a 5.88-fold increase in infertility ratio and low levels of cadmium accumulation in semen may contribute to male infertility by reducing sperm quality. It is known that tobacco smoking affects the reproductive system of both sexes and presence of nicotine and cotinine in reproductive fluid may have significant implications in IVF outcome. Earlier, nicotine and/or cotinine were detected in the seminal plasma by various investigators (Pacifici et al., 1995; Vine et al., 1993; Zenzes et al. 1999). Trichloroethylene (TCE) and its metabolites chloral and trichloroethanol (TCOH) were detected by Poh-Gek et al., (2003) in the seminal fluids of all the eight mechanics exposed to TCE occupationally. Reproductive contaminants found in FF and seminal plasma may compromise the quality of oocytes and or sperm, which may become one of the risk factors for reproductive outcome. The data available suggests certain chemicals or their metabolite reaches the seminal plasma and/or FF, indicating their possible impact in IVF outcome.

Toxicants and IVF outcome

Certain industrial and environmental persistent chemicals and other factors might affect the IVF outcome in addition to other host factors such as paternal age and reproductive health. Younglai et al. (2002) mentioned that more than 50% of the population of women attending a fertility program have had exposure to environmental chemicals sufficient to produce detectable concentrations in their serum and ovarian FF. Of the chemical contaminants detected in the serum and FF of these women, *p,p′*-DDE was the most frequently detected and was associated with failed fertilization. Nevertheless, paternal pesticide exposure has been reported to decrease sperm fertilizing ability in vitro among those seeking IVF treatment (Tielemans et al., 1999).

Tsutsumi et al. (1998) demonstrated the presence of polychlorinated dibenzodioxins (PCDDs) and dibenzofurans (PCDFs) at concentrations of approximately 1 pg/mL in human FF. To study their possible action, two-cell mouse embryos were cultured in the presence of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) and evaluated at 24-hour intervals for their development to the eight-cell and blastocyst stages. The percentage of eight-cell embryos exposed to TCDD at 1, 2 and 5 pM concentrations was significantly lower than that of controls. However, blastocyst formation of the surviving eight-cell embryos was accelerated, with the number of cells in the blastocysts increased in a dose-dependent manner. They suggested that PCDDs and PCDFs may exert some stage-specific effects on early embryonic development. In addition, diethylstilbestrol (DES) was reported to have adverse effect on reproduction. Infertile patients with a history of in utero exposure to DES exhibit a significantly impaired implantation rate following IVF, and the outcome of ART remains poor (Pal et al., 1997). Neal et al. (2008) found significantly higher levels of B [a] P in the FF of women who did not conceive compared to those that conceived.

It is known that Pb can concentrate in bodily compartments where it disrupts cellular processes and can result in detrimental reproductive health consequences. Silberstein et al. (2006) determine Pb levels in blood and FF from nine patients undergoing IVF treatment. Pb levels within FF were found to be significantly higher in non-pregnant patients compared to pregnant patients, suggesting that elevated concentrations of the environmental Pb adversely affect female reproduction. Benoff et al. (2003) in a prospective study of the metal ion levels and sperm function, semen was obtained from partners of 140 consecutive women undergoing their first IVF cycle. Pb levels were negatively correlated with IVF rates. Pb levels were also negatively correlated to two of the three sperm function biomarkers (mannose receptors, mannose-induced acrosome reactions).

Exposure to solvents can occur in different ways. In view of their lipid solubility, it is likely that most organic solvents pass through the placenta into the foetus. In humans, occupational exposure to organic solvents has been related to various disorders of reproductive health. These include menstrual disorders, reduced fertility and adverse pregnancy outcome as reviewed by Kumar (2008). Tielemans et al. (2000) reported a significantly reduced implantation rate among couples with male partners working in occupations with presumably high levels of organic solvent exposure. Conversely, paternal pesticide exposure was significantly associated with an increased implantation rate. Paternal exposures to metal dust or fumes and welding fumes were not related to the probability of implantation. The reasons behind these are not clearly understood. An experimental study reports that in vivo exposure of male rats to TCE results in a decline in the ability of sperm to fertilize oocytes in vitro. The decrease in the percentage of fertilized oocytes occurred without any observed effects on sperm concentration, motility, energy (mitochondrial) status or acrosome integrity (Du Teaux et al., 2004). Hjollund et al. (2004) found no increased risk of spontaneous abortion in IVF-treated women attributable to paternal agricultural application of pesticides and growth retardants. They also mentioned that exposure to potentially harmful pesticides in Denmark is relatively low, and the findings are restricted to countries with similar standards of protection. However, Younglai et al. (2004) reported that DDE was shown to stimulate the aromatase enzyme system of human granulosa cells and to have a synergistic effect with follicle-stimulating hormone (FSH). Later, they suggested that this could be interpreted as having a beneficial effect or as having an adverse effect through premature production of estradiol, which is implicated in oocyte maturation as suggested by Younglai et al., (2005).

Klonoff-Cohen (2005) reviewed the data on the role of lifestyle habits (smoking, alcohol and caffeine use, and psychological stress) on the reproductive endpoints of IVF (i.e. oocyte aspiration, fertilization, embryo transfer, achievement of a pregnancy, live birth delivery and perinatal outcomes, e.g. low birth weight, multiple gestations). There is compelling evidence that smoking has a negative influence on IVF outcomes, whereas for stress, the evidence is suggestive but insufficient due to the heterogeneity of studies. The evidence for the effects of alcohol and caffeine on IVF is inadequate, due to the scarcity of studies. However, Wright et al. (2006) reported that smoking is widely believed to be associated with decreased fecundity in naturally conceiving populations; however, the effect of female smoking on pregnancy outcomes in patients undergoing IVF is unclear. A total of about 21% of IVF patients in a retrospective analysis of 389 consecutive patients undergoing first cycle IVF had past or present exposure to cigarette smoking, with no measurable effect on IVF outcome being reported. Bellver (2008) also reported that psychological stress, consumption of caffeine and alcohol and illicit drug use have been implicated in a poorer IVF outcome, but evidence is inconclusive due to the scarcity and inadequate methodology.

Possible mechanism involved in impairment of IVF outcome

It is known that both mother and father can transmit genetic defects to their offspring, which are detrimental for normal development of offspring. Sperm DNA contributes one half of the genomic material to the offspring. The mechanism behind the adverse IVF outcome due to parental exposure to certain chemicals is not known. But the effect of these chemicals on male and female gametes might have the role in IVF outcome. These chemicals may exert effects at cellular, sub cellular or molecular levels, leading to DNA damages, which in turn affect the IVF outcome. A recent study in rat highlights the effects of toxic metals that disturb membrane integrity of cells via reactive oxygen species (ROS) and thereby classifying mechanism for altered receptor binding, steroidogenesis, and hormone production (Nampoothiri et al., 2007). Earlier, Agarwal and Prabakaran (2005) emphasized to consider ROS as one of the mediators of infertility, causing sperm dysfunction. Although ROS is involved in many physiological functions of human spermatozoa, their excess production results in oxidative stress. ROS causes damage to the spermatozoa DNA, resulting in increased apoptosis of these cells. The production of ROS is greatly enhanced by the influence of various environmental and life style factors such as pollution and smoking. Recently du Plessis et al. (2008) mentioned that gametes and embryos are natural sources of free radicals. When manipulated in vitro during assisted reproductive techniques, these cells run the risk of generating and being exposed to supra-physiological level of ROS. Thus, free radicals and oxidative stress can have a significant impact on IVF outcome. Benoff

et al. (2000) found that higher Pb level in the seminal plasma correlated with low expression of mannose receptors and with inability of sperm to undergo mannose-induced acrosome reaction. Conversely, higher Pb levels were associated with premature (spontaneous) acrosome reaction that occurs before sperm-egg contact, also blocking fertilization. A negative correlation between seminal plasma Pb levels and percentage of sperms with normal DNA was also observed in our laboratory (Mankad, 2007). Lewis and Aitken (2005) mentioned that DNA damage in the male germ cell line has been associated with poor semen quality, low fertilization rates, impaired preimplantation development, increased abortion and an elevated incidence of disease in the offspring. The interest to assess the chromatin quality of human sperm has increased since DNA damage in sperm from infertile men has been associated with infertility. Its importance has become more obvious in the context of increasing use of assisted reproductive techniques in infertility. The data suggest that people should adopt preventive measures by adopting safer work place and life styles in order to reduce or control an unwanted exposure to certain reproductive toxicants. The data available highlights the lack of studies on the exposure of couples to environmental/occupational conditions and the association with IVF outcome. There are needs for (1) to study exposure and effect parameters for example, reproductive outcome and their interpretation with IVF outcome (2) Studies in different ethnic groups on reproduction and differences in response to toxic exposures. These studies could provide information to conclude that particular exposure hazards pose a real risk for adverse IVF outcomes. There may not be conclusive evidence for the various factors discussed and adverse IVF outcome, but some factors might have impact at least in part in IVF outcome.

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