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Exposure to mercury and human reproductive health: a systematic review

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Highlights

- Increased mercury levels were associated with infertility or subfertility status;
- Infertile males had higher levels of mercury in hair than fertile males;
- Mercury reduced semen quality parameters;
- Increased mercury levels were associated with incidence of menstrual and hormonal disorders.

Abstract

Background: Evidences from human and animal studies suggest that reproductive function may be affected by mercury. The aim of this review was to explore the mercury influence on human fertility.

Methods: A systematic search was made in PubMED for papers published between 1975-2017, according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses guidelines.

Results: Increased mercury levels were associated with infertility or subfertility status. Further, infertile subjects with unexplained infertility showed higher levels of mercury in hair, blood and urine than fertile ones. Mercury exposure induced sperm DNA damage and abnormal sperm morphology and motility. Additionally, mercury levels were related with higher incidence of menstrual and hormonal disorders and increased rates of adverse reproductive outcomes.

Conclusions: Our review showed that mercury negatively impacts human reproduction, affecting the reproductive and endocrine systems in both male and female. However, the molecular mechanisms underlying the mercury-associated decline on fertility remains unknown.

Abbreviations:

BMI – body mass index; BTB – blood-testis-barrier; DFI – DNA fragmentation index; DNA – deoxyribonucleic acid; ELSPAC – European Longitudinal Study of Pregnancy and Childhood; FSH – follicle-stimulating hormone; GnRH – gonadotropin-releasing hormone; GSTM – glutathione S-transferase M; HDS – high DNA stainable; Hg – mercury; IVF – *in vitro* fertilization; LH – luteinizing hormone; MeHg – methylmercury; PCO – polycystic ovary; PRISMA - Preferred Reporting Items for Systematic Reviews and Meta-Analyses; SHBG – sex hormone-binding globulin; ROS – reactive oxygen species; TSH – thyroid-stimulating hormone; TTP – time-to-pregnancy; USA – United

States of America; USEPA – United States Environmental Protection Agency; WHO – World Health Organization.

Keywords:

Epidemiology; Human fertility; Mercury exposure; Reproduction; Systematic Review.

1. Introduction

Infertility is defined, by the World Health Organization (WHO), as a disease of the reproductive system that consists in the failure to achieve a clinical pregnancy after 12 months or more of regular unprotected sexual intercourse [1]. It is estimated that nearly 15% of couples are infertile and in approximately 50% the causes are unknown. Lifestyle factors related with smoking, alcohol or caffeine intake and environmental risk factors, namely exposure to heavy metals such as mercury (Hg), lead, cadmium and/or mixture of metals may impact the reproductive system [2–5].

Hg is a naturally-occurring chemical element found on Earth, existing in three different chemical forms: elemental, inorganic and organic (e.g. methylmercury, MeHg) [6]. In humans, Hg exposure occurs predominantly through the consumption of seafood or sashimi, but also dental amalgams, button cell batteries, broken thermometers and compact fluorescent light bulbs and skin-lightening creams. Hg exposure is also dependent on the living region, education level and type of job [7–9].

Hg causes adverse effects on male reproductive functions in rats, namely impairment of spermatogenesis, decrease in sperm motility and increase in the number of sperm head abnormalities [10–12]. In females, data indicate that Hg exposure can result in an accumulation in ovaries of mice that may cause alterations in reproductive behavior and contribute to infertility or ovarian failure [13]. Moreover, there is transplacental passage of Hg in animals [14]. However, despite numerous animal studies, whether Hg affects

human reproductive health remains unclear.

To our knowledge, no systematic review and/or meta-analysis reported the effect of Hg on human fertility and generally on human reproductive health. Given the Hg exposure in the general population, understanding the impact of Hg on human fertility is relevant for public health policies. Therefore, we systematically reviewed the available literature to evaluate the influence of Hg exposure on human fertility and/or adverse reproductive outcomes.

2. Methodology

2.1. Literature search methodology

For the purpose of this systematic bibliographic review, a search was made in the scientific PubMed database from 1 January 1975 to 30 November 2017, according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines [15]. In addition, other papers were located by manual search targeting journals, particularly those less likely to be indexed and references cited by papers retrieved.

The search strategy was designed to identify relevant studies addressing Hg exposure and the effects on human fertility, including effects on sperm parameters, testis and ovarian functions, reproductive hormones and reproductive outcomes. The following search terms were used in PubMed: (mercury OR methylmercury) AND (fertility OR infertility OR reproduction OR pregnancy OR time-to-pregnancy OR testis OR testicular OR sperm OR spermatozoa OR semen OR ovary OR ovaries OR oocyte OR follicle OR ovarian OR embryo quality).

To be considered eligible for this review, studies were required to: (1) be published in English; (2) state that their main aim was to evaluate the effect of Hg exposure on human

reproductive health, including semen parameters, time-to-pregnancy (TTP), gonadal endocrine function, fertility status and adverse reproductive outcomes; (3) use quantitative methodology; (4) focus on human studies.

Studies were excluded if they were case-reports, review articles and preclinical or *in vitro* studies. Studies involving patients with infertility accompanied by other disorders of the reproductive system (e.g. varicocele) were also excluded.

Two reviewers screened out non-relevant titles and examined the abstracts to determine if studies fell within the inclusion criteria for the review. Whenever a title or abstract could not be rejected with certainty, the full text was obtained for further screening. The full text was then obtained for the remaining studies and determined if those manuscripts met the inclusion criteria.

2.2. Data extraction

To summarize the methodological and the sample characteristics of the selected studies, **Table 1** was drawn up showing the following parameters: author, year of publication, country, number of subjects, study design, setting, outcome measured and age of patients included (range or mean ± standard deviation).

Table 1 - Main characteristics of studies on Hg exposure and human fertility and reproductive outcomes.

First author (year), country	Groups	Design	Setting	Outcome measure	Range or mean age
De Rosis et al. (1985), Italy [16]	♀ Exposed – 153 ♀ Non-exposed - 293	Cross-sectional*	Two factories located in southern and northern Italy	Questionnaire concerning reproductive history, demographic data and history of exposure. Relation between Hg exposure and prevalence of reproductive disorders.	N/D
Lauwerys et al. (1985), Belgium [17]	♂ Exposed – 103 ♂ Non-exposed - 101	Questionnaire study	Six factories in Belgium	Questionnaire concerning demographic data, reproductive history and working conditions. Relation between Hg exposure and reproductive outcomes.	Exposed – 21-60 Non-exposed – 22-58
Sikorski et al. (1987), Poland [18]	♀ Exposed – 81 ♀ Non-exposed - 34	Cross-sectional*	Dental service surgeries in the Lublin	Questionnaire concerning demographic data, reproductive history and working conditions. Relation between total Hg levels in the scalp and pubic hair and reproductive process of women working in dental surgeries.	Exposed - 21-56 Non-exposed - 20-46
Alcser et al. (1989), USA [19]	♂ Exposed – 241 ♂ Non-exposed - 254	Retrospective cohort	Department of Energy plant	Questionnaire concerning reproductive history, demographic data, relevant occupational exposure and lifestyle factors. Relationship of male occupational elemental Hg exposure and several reproductive outcomes.	20-50
Cordier et al. (1991) [20]	♂ Exposed – 152 ♂ Non-exposed - 374	Epidemiological	Chloralkali plant	Questionnaire concerning occupational history and reproductive history. Relationship of male occupational elemental Hg exposure and reproductive outcomes: spontaneous abortions.	Exposed - 37.0 ± 0.5 Non-exposed - 37.3 ± 0.3
McGregor et al. (1991) [21]	> 5 years exposed – 11 < 5 years exposed – 18 Control – 63	Cross-sectional	Industries that involve vapor Hg exposure	Effect of Hg exposure on testicular endocrine function.	> 5 years exposed – 50 ± 11.4 < 5 years exposed – 35.2 ± 13.7 Control – 41.3 ± 12.6

Chia et al. (1992), Singapore [22]	♂ - 35	Cross-sectional*	Andrology Clinic at Singapore General Hospital	Relationship of semen parameters (volume, total sperm count, sperm viability, progressively motile sperm and sperm morphology) and blood concentrations of Hg.	37.7 ± 5.5
Barregârd et al. (1994) [23]	♂ Exposed – 41 ♂ Non-exposed – 41	Cross-sectional*	Chloralkali plant	Effect of Hg exposure on testicular endocrine function.	Exposed – 19-65 Non-exposed 18-61
Rowland et al. (1994), USA [24]	♀ Exposed – 296 ♀ Non-exposed - 111	Retrospective cohort	California Department of Consumer Affairs	Effect of occupational Hg exposure on female fertility.	N/D
Hanf et al. (1996), Germany [25]	Cases – 80 Cohort control – 7	Case-control*	Cases - Women's Hospital, University of Tiibingen Cohort controls – workers at a thermometer manufacturing plant	Questionnaire concerning the number and size of amalgam fillings. Relationship of Hg concentrations in urine and ejaculate and semen parameters.	Cases – 35 Control – 45
Dickman et al. (1998), China [26]	♀ - 35 ♂ - 166	Cross-sectional*	IVF center	Questionnaire about demographic data, lifestyle factors and clinical data. Relationship of Hg concentrations found in the hair of subjects and fertility status.	25-72
Gerhard et al. (1998), Germany [27]	♀ Infertile - 501	Observational	Department of Gynecological Endocrinology and Reproduction, University Hospital of Obstetrics and Gynaecology, Heidelberg, Germany	Relation between urinary Hg excretion and female infertility.	30 ± 7
Dahl et al. (1999), Norway [28]	♀ Exposed – 558 ♀ Non-exposed - 450	Retrospective observational	Exposed group - Norwegian Dental Association Non-exposed group - Norwegian Educational Association	Questionnaire concerning reproductive history, occupational history and demographic data. Effect of occupational Hg exposure on female fertility.	N/D
Leung et al. (2001), China [29]	♂ - 59 (divided into two groups according Hg concentration)	Cross-sectional*	Assisted Reproduction Unit of the Chinese University of Hong Kong	Questionnaire concerning the duration of infertility, presence of female factors, demographic data and social habits. Association between whole blood Hg concentrations and semen quality parameters.	36-41
Choy et al. (2002) China	♂ - 111	Prospective observational	IVF Unit of the Prince of Wales Hospital in Hong Kong	Relationship between semen parameters and Hg concentrations in blood and seminal fluid of males.	38

[30]					
Choy et al. (2002), China [31]	♂ Cases – 150 Control – 26 ♀ Cases – 155 Control – 26	Case-control	IVF Unit of the Prince of Wales Hospital in Hong Kong	Relationship between blood Hg concentration and male/female fertility outcomes-	♂ Cases – 38.0 ± 4.2 Control – 34.3 ± 5.8 ♀ Cases – 34.8 ± 3.3 Control – 31.0 ± 5.1
Arakawa et al., (2006), Japan [32]	♀ - 180 ♂ - 180	Prospective cohort	Two hospitals in Sendai	Questionnaire about lifestyle parameters, TTP and biological attributes. Associations between hair Hg concentrations and TTP.	♀– 30.6 ♂– 31.9
Cole et al. (2006), Canada [33]	♀ - 41 ♂ - 41	Cross-sectional*	Department of Obstetrics and Gynecology of St. Joseph's Hospital in Hamilton	Questionnaire regarding demographic data, work history, environmental exposures, personal consumption, medical and reproductive history. Effects of maternal and paternal Hg exposure on TTP.	♀– 20-34 ♂– 20-45
Rignell-Hydbom et al. (2007), Sweden [34]	♂ - 195	Sub-cohort	Cohort of fishermen from the Swedish	Questionnaire about lifestyle, medical and reproductive data. Association between MeHg exposure and human sperm motility, sperm concentration, total sperm count, sperm chromatin integrity and the proportion of Y-chromosome bearing sperms.	24-67
Xue et al. (2007), USA [35]	♀ -1024	Prospective study	Prenatal clinics of Michigan communities	Personal interviews regarding demographic data, dietary habits and lifestyle factors. Associations between maternal fish consumption, Hg levels and risk of preterm delivery.	< 25
Al-Saleh et al. (2008), Saudi Arabia [36]	**Pregnancy outcome** Cases – 321 Controls – 203 **Fertilization outcome** Cases – 63 Controls – 556	Case-control*	IVF embryo transfer unit, King Faisal Specialist Hospital and Research	Questionnaire about demographic data, menstrual history, fertility problems, lifestyle factors and environmental and occupational history. Influence of Hg exposure on *in vitro* fertilization and pregnancy outcome.	19-50

Meeker et al. (2008), USA [37]	Study – 146 Comparison – 73	Cross-sectional*	Two Michigan infertility clinics	Relationship between Hg exposure at environmental levels and human semen-quality parameters.	18-55
Laks (2009), USA [38]	♀ - 485	Sub-Cohort*	National Health and Nutritional Survey	Assess chronic Hg exposure within the US population. Associations between Hg concentrations in blood and hormone levels.	35-40
Meeker et al. (2010), USA [39]	♂ - 219	Cross-sectional	Two Michigan infertility clinics	Associations between Hg exposure and male reproductive hormone levels.	18-55
Bloom et al. (2010), USA [40]	♀ - 58 ♂ - 36	Preliminary study	University of California at San Francisco Center for Reproductive Health	Association between Hg exposure and *in vitro* fertilization endpoints.	♀- 35.8 ± 3.9 ♂– 38.4 ± 4.3
Bloom et al. (2011), USA [41]	♀ - 54 ♂ - 36	Preliminary prospective cohort	University of California at San Francisco Center for Reproductive Health	Association between Hg levels and embryo quality indicators during *in vitro* fertilization.	♀- 35.63 ± 3.81 ♂– 38.50 ± 4.28
Dickerson et al. (2011) [42]	♀ - 30	Prospective pilot	N/D	Assess the impact of long and short-term Hg exposure status on the gonadal response to gonadotrophin stimulation during a standard long-protocol agonist *in vitro* fertilization cycle.	32.7 ± 4.4
Jackson et al. (2011), USA [43]	♀ - 259	Cohort	University at Buffalo	Relationship between Hg blood levels and female reproductive hormones.	18-44
Mendiola et al. (2011), Spain [44]	Case – 30 Control - 31	Pilot study	Three infertility centers of the Instituto Bernabeu in Murcia and Alicante	Interview face-to-face and lifestyle questionnaire. Relationship between Hg in three different body fluids and seminal and hormonal parameters.	33.5 ± 3.8
Pollack et al. (2011), USA [45]	♀ - 252	Cohort	University at Buffalo	Association between Hg and female reproductive hormones.	18-44
Hanna et al. (2012), USA [46]	♀ - 58	Pilot study	University of California at San Francisco (UCSF) Center for Reproductive Health	Association between Hg exposure and DNA methylation changes.	28-44
Louis et al. (2012), USA [47]	♀ - 501 ♂ - 501	Cohort	Four Michigan counties.	Person interviews about health and reproductive history and demographic data.	♀ – 19-40 ♂ – 19-51

Chevrier et al. (2013), France [48]	♀ - 394	Cohort	Routine prenatal care visits	Associations between couples Hg exposure and adverse reproductive outcomes (incident pregnancy loss). Questionnaire about family, social and demographic data and diet and lifestyle habits. Associations between Hg serum levels, seafood consumption and female fertility.	25-35
Mocevic et al. (2013), Greenland, Poland and Ukraine [49]	♂ - 529	Cross-sectional	Clinic of the Gynaecological and Obstetric Hospital of the Warsaw School of Medicine, Poland Three maternity hospitals and eight antenatal clinics, Ukraine 4 settlements and 15 municipalities, Greenland	Relationship between blood concentrations of Hg and semen characteristics and serum levels of reproductive hormones.	Greenland – 31.0 Poland – 30.4 Ukraine – 26.8
Zeng et al. (2013), China [50]	♂ - 118	Cross-sectional	Reproductive Center of Tongjing Hospital in Wuhan	Questionnaire regarding demographic data, lifestyle habits, occupational exposure and medical characteristics. Association between urinary Hg concentrations and circulating testosterone in men.	30.8 ± 5.6
Kim et al. (2014), USA [51]	♂ - 30	Cross-sectional*	University of California at San Francisco	Questionnaire about dietary and health data. Association between Hg levels in seminal plasma and semen quality and *in vitro* fertilization outcomes.	32-45
Tanrikut et al. (2014), Turkey [52]	♀ Infertile (cases) - 33 ♀ Fertile (controls) - 32	Case-control*	Department of Obstetrics and Gynaecology, Inonu University, School of Medicine	Questionnaire about demographic and socio-economic data. Relationship between endometrial concentrations of Hg and unexplained infertility.	Infertile ♀ - 29.00 ± 5.24 Fertile ♀ - 30.84 ± 5.06
Lei et al. (2015), China [53]	♀ Infertile - 310 ♀ Fertile - 57	Preliminary study	Department of Obstetrics and Gynecology in the Taiwan Adventist Hospital	Interview face-to-face about sociodemographic data, lifestyle information and menstruation history. Relationship between blood-MeHg levels and women reproductive hormones.	Infertile ♀- 35.2 ± 3.9 Fertile ♀ - 34.8 ± 4.1
Lenters et al. (2015), Greenland, Poland and Ukraine [54]	♂ - 602	Cross-sectional	Central Hospital in Warsaw, Poland Three hospitals and eight antenatal clinics in Kharkiv, Ukraine Local hospitals in 19 municipalities and settlements across Greenland.	Identification of exposure profiles associated with biomarkers of male reproductive function.	Greenland – 31.0 Poland – 30.4 Ukraine – 26.8

Wright et al. (2015), USA [55]	♀ - 205	Prospective cohort	Massachusetts General Hospital Fertility Center	Questionnaires about demographic data, medical history, lifestyle factors and occupation. Associations between hair Hg levels and gamete, embryo and clinical outcomes.	27-43
Zeng et al. (2015), China [56]	Study – 225 Comparison - 169	Cross-sectional	Reproductive Center of Tongjing Hospital in Wuhan	Association between urinary Hg concentrations and semen quality parameters.	22-50
Hsi et al. (2016), China [57]	♀ Pregnant – 62 ♀ Infertile - 162	Cross-sectional*	Northern and Central Taiwan	Questionnaire regarding demographic, lifestyle and reproductive data. Correlation between hair MeHg concentrations and daily MeHg exposure among pregnant and infertile women.	Pregnant women – 31.6 ± 4.9 Infertile women – 34.4 ± 3.7
Zhou et al. (2016), China [58]	♂ - 207	Cross-sectional	Reproductive Center of Tongjing Hospital in Wuhan	Questionnaire about demographic data, lifestyle habits, occupational exposure and medical characteristics. Association between urinary Hg concentrations and sperm DNA damage.	31.6 ± 5.3
Buck Louis et al. (2017), USA [59]	♀ - 344 ♂ - 344	Cohort	16 counties in Michigan and Texas	Person interview about anthropometric data and lifestyle. Relationship between environmentally Hg concentrations and incident pregnancy loss.	♀ - 29.8 ± 3.9 ♂– 31.6 ± 4.6
Mínguez-Alarcón et al. (2017), USA [60]	♂ - 129	Prospective cohort	Massachusetts General Hospital Fertility Center	Questionnaire about lifestyle factors, reproductive health and medical history. Association between hair Hg levels and semen parameters and fish intake.	31.8-42.5

N/D – not defined; ♀ - women; ♂ - men; ELSPAC - European Longitudinal Study of Pregnancy and Childhood; Hg – mercury; MeHg – methylmercury; DNA – deoxyribonucleic acid; USA – United States of America; IVF – *in vitro* fertilization; TTP – time to pregnancy.

* The design of the study was not defined by authors.

3. Results

3.1. Selection of papers

A total of 2486 unduplicated abstracts were found in PubMed. After titles and abstract examination, 54 papers were selected for perusal of the complete text and 2 more were added after manual search [38,39]. Finally, a total of 45 papers [16–60] were included in the review (

Figure *1*).

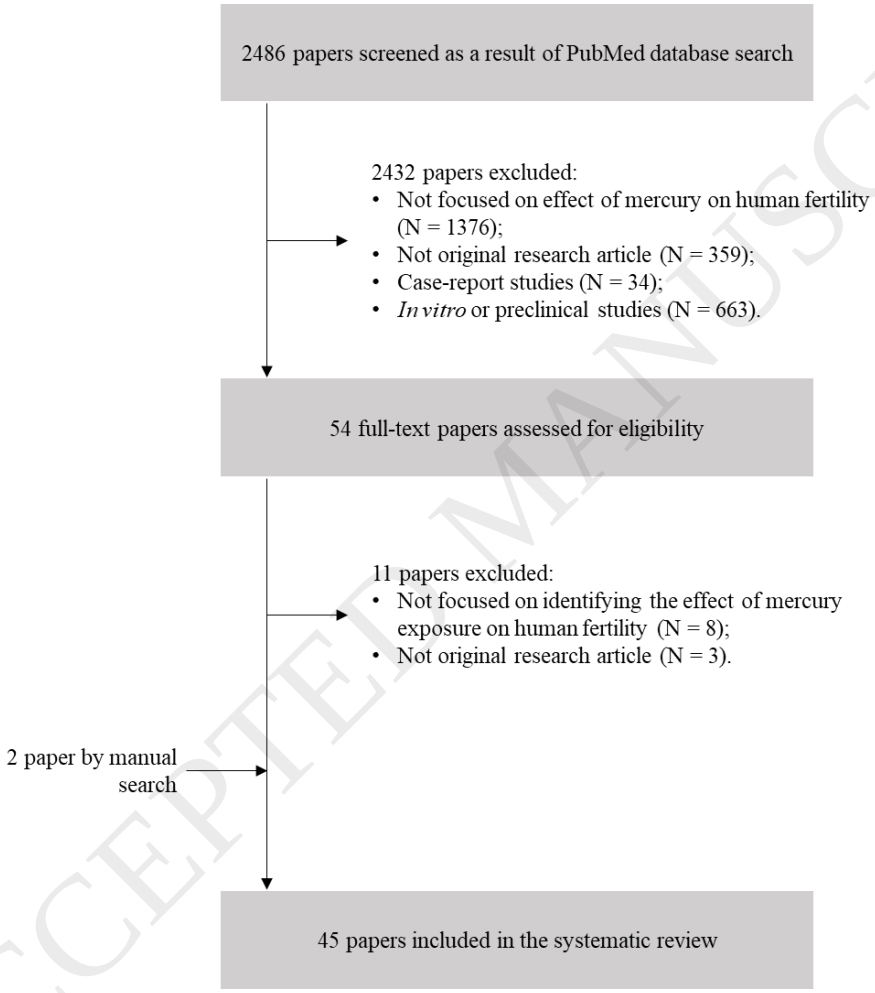

Figure 1 - Flow diagram showing study extraction and selection. Visualization of the literature search and the study selection process according to the PRISMA guidelines [15]**.**

3.2. Characteristics of selected papers

The general characteristics of the selected studies are summarized in **Table 1**. The majority of the studies were drawn from different continents including Europe (n=11)

[16–18,25,27,28,34,44,48,49,54], Asia (n=13) [22,26,29–32,36,50,52,53,56–58] and North America (n=17) [19,24,33,35,37–41,43,45–47,51,55,59,60]. The other four studies did not have information concerning the country [20,21,23,42]. The country with most studies was United States of America (USA) [19,24,35,37–40,43,45–47,51,55,59–61]**.**

The majority of the studies included in this review were undertaken at different hospital departments [18,22,25,27,28,32,33,35,48,49,52–54,60], *in vitro* fertilization centers [26,29–31,36,40,41,46,50,55,56,58] or infertility clinics [37,39,44]. Some studies were undertaken at factories associated with Hg exposure [16,17,19–21,23,24]. In terms of age, the patients included in the studies were very heterogeneous, ranging from 18-72 years old.

3.3. Male fertility

Men are unavoidable exposed to Hg due to its ubiquity in nature and persistence in the environment. It has been suggested that Hg exposure can interfere with the male reproductive health. Higher blood and hair Hg concentrations were associated with male subfertility or infertility status [26,31]. Several studies assessed the relationship between male occupational and environmental Hg exposure and reproductive outcomes [17,19,20,47]. The risk of spontaneous abortion raised significantly with the increasing Hg concentrations (OR = 2.26; 95% CI = 0.99-5.23) [19,20]. No significant associations were demonstrated between occupational and environmental Hg exposure and a decreased fertility or increased adverse reproductive outcomes, such as abnormalities or illnesses in the children of the exposed workers [17,19,20,47].

Regarding the associations between male Hg exposure and *in vitro* fertilization outcomes, an inverse association between blood Hg in the male partner of infertile couples and embryo fragmentation was found [41]. Kim et al. [51] described positive associations

between seminal plasma Hg levels of infertile males and good embryo quality, implantation, pregnancy and live birth. These observations do not support the hypothesis that Hg exposure has an adverse impact on male fertility and can be explained by the fact that developing sperm are protected by the blood-testis-barrier (BTB). Indeed, Choy et al. [30] showed that blood Hg concentrations were significantly higher than in seminal plasma (41.4 ± 1.7 nmol/L versus 22.1 ± 2.0 nmol/L, respectively; $p<0.05$).

3.3.1. Impact of Hg on semen parameters

Semen quality, a predictor of male fertility, is declining worldwide [62]. Many factors have been proposed as causes for this decline, including lifestyle habits and pollutants exposure [4,5,63,64]. Thus, the relation between semen parameters and lifestyle habits or environmental pollutants exposure has become a topic of interest. Animal studies have shown that Hg exposure induces abnormalities in sperm morphology and motility [65,66]. However, human studies are few and contradictory. Eight of 11 studies that evaluated the effects of Hg exposure presented the results for semen quality parameters [22,29,34,37,44,49,56,60] **(Table 2)**, while the others only presented the results of the statistical analysis between Hg concentrations in seminal fluid and semen parameters [25,30,51]. Most of participants were male partners of infertile couples attending fertility clinics [22,25,29,30,37,44,51,56,60]. The age range of the subjects was 18-67 years old. Most of the studies did not find significant correlations between Hg concentration in biological samples and semen parameters (sperm concentration, motility and morphology) [22,25,29,34,37,44,49,51,56]. Hanf et al. [25] recruited 80 men from a fertility clinic to correlate the Hg concentration in urine and ejaculate semen with the semen quality parameters. The author analyzed the semen samples according to WHO guidelines of 1999 [67] and calculated the numerical "fertility index" according to Matthies et al. [68]. Results showed that urinary Hg levels were within the normal values

for a non-exposed population. Moreover, the Hg concentration measured in semen was below the detection limit (5 µg/L) in several samples. No positive correlations could be established between subject urine or ejaculate Hg concentrations and the quality of their semen.

In contrast, there were evidences that all semen quality parameters were lower in subjects with high Hg levels and that infertile males with abnormal semen parameters had significantly higher blood Hg concentrations than fertile males [29–31]. Choy et al. [30] showed that seminal Hg concentrations were positively correlated with abnormal sperm morphology ($r = 0.26$, $p = 0.02$), as well as negatively correlated with normal sperm motility ($r = -0.21$; $p = 0.03$). However, no correlations were found between the percentage of motile sperm or sperm concentrations and Hg concentrations.

Recently, contrary to previous studies and for the first time, Mínguez-Alarcón et al. [60] reported that hair Hg levels, among men with higher fish intake, were positively related with sperm concentration, total sperm count and progressive motility.

Table 2 - Semen parameters of subjects included in all studies and *p*-value or correlation coefficient (R) of semen parameters and concentration of Hg.

First author (year), country	Data are expressed as	Semen volume (mL)	Concentration (10^6/mL)	Sperm count (millions)	Motile sperm (%)	Normal morphology (%)
Chia et al. (1992), Singapore [22]	Mean (± SD) and range	2.8 (± 1.0) 0.8 – 4.8	25.7 (± 31.1) 1.5 – 113.0	N/A	30.5 (± 18.2) 0.0 – 69.0	22.0 (± 17.2) 0.0 – 99.0
	Correlation coefficient (r) of semen parameters and concentration of Hg	r = -0.20 NS	r = 0.30 NS		r = 0.02 NS	r = -0.13 NS
Leung et al. (2001), China [29]	Median (interquartile range)	N/A	**Control** 65.7 (44.6 – 112.4) **Cases** 46.1 (36.4 – 92.5)	N/A	**Control** 49.0 (31.8 – 60.0) **Cases** 45.4 (31.9 – 58.2)	**Control** 50.0 (35.0 – 60.0) **Cases** 40.0 (35.0 – 55.0)
	p-value for differences between the 2 groups		*p* = 0.25 NS		*p* = 0.60 NS	*p* = 0.33 NS
Rignell-Hydbom et al. (2007), Sweden [34]	Median (5-95% range) *p*-value for differences between the exposure groups	**Hg < 1.08 µg/L** 3.5 (0.9 – 5.9) **Hg 1.08 – 1.86 µg/L** 3.0 (0.8 – 5.5) **Hg 1.86 – 2.79 µg/L** 3.4 (1.2 – 6.2) **Hg 2.79 – 4.40 µg/L** 3.1 (0.7 – 5.8) **Hg > 4.40 µg/L** 3.3 (1.2 – 7.7) *p* = 0.6 NS	**Hg < 1.08 µg/L** 42 (6 – 119) **Hg 1.08 – 1.86 µg/L** 54 (4 – 200) **Hg 1.86 – 2.79 µg/L** 48 (9 – 176) **Hg 2.79 – 4.40 µg/L** 53 (17 – 207) **Hg > 4.40 µg/L** 48 (16 – 110) *p* = 0.9 NS	**Hg < 1.08 µg/L** 135 (14 - 590) **Hg 1.08 – 1.86 µg/L** 170 (13 - 477) **Hg 1.86 – 2.79 µg/L** 127 (15 - 732) **Hg 2.79 – 4.40 µg/L** 159 (26 - 386) **Hg > 4.40 µg/L** 134 (45 - 342) *p* = 1.0 NS	**Hg < 1.08 µg/L** 62 (17 - 90) **Hg 1.08 – 1.86 µg/L** 63 (21 - 86) **Hg 1.86 – 2.79 µg/L** 54 (15 - 83) **Hg 2.79 – 4.40 µg/L** 64 (14 - 87) **Hg > 4.40 µg/L** 60 (12 - 84) *p* = 0.9 NS	N/A
Meeker et al. (2008), USA [37]	Adjusted OR (95% CI)	N/A	**Hg < 25th** 1	N/A	**Hg < 25th** 1	**Hg < 25th** 1

	p-value for trend		**Hg 25th-50th** 0.52 (0.18 – 1.49) **Hg 50th-75th** 0.47 (0.16 – 1.36) **Hg > 75th** 0.36 (0.12 – 1.03) *p* = 0.06 NS		**Hg 25th-50th** 0.44 (0.16 – 1.16) **Hg 50th-75th** 0.74 (0.30 – 1.83) **Hg > 75th** 0.35 (0.14 – 0.90) *p* = 0.07 NS	**Hg 25th-50th** 0.48 (0.19 – 1.17) **Hg 50th-75th** 0.69 (0.29 – 1.64) **Hg > 75th** 0.43 (0.18 – 1.02) *p* = 0.1 NS
Mendiola et al. (2011), Spain [44]	Mean (± SD) *p*-value for differences between the 2 groups Mean	**Control** 3.5 (± 1.4) **Cases** 3.8 (± 1.2) *p* > 0.05 NS	**Control** 39.5 (± 14.6) **Cases** 3.3 (± 4.1) *p* > 0.05 NS	**Control** 151 (± 71.9) **Cases** 12.6 (± 9.1) *p* > 0.05 NS	**Control** 52.2 (± 12.3) **Cases** 27.4 (± 18.6) *p* > 0.05 NS	**Control** 22.3 (± 4.5) **Cases** 3.7 (± 1.5) *p* > 0.05 NS
Mocevic et al. (2013), Greenland, Poland and Ukraine [49]	*p*-value for differences between the exposure groups β (95% CI): association between Hg exposure and outcomes	**Hg 0.2 – 0.9 ng/mL** 3.7 **Hg 0.9 – 2.7 ng/mL** 3.9 **Hg 2.7 – 385.8 ng/mL** 3.5 -0.018 (-0.052, 0.015)	**Hg 0.2 – 0.9 ng/mL** 84.6 **Hg 0.9 – 2.7 ng/mL** 75.6 **Hg 2.7 – 385.8 ng/mL** 70.8 -0.022 (-0.081,0.038)	**Hg 0.2 – 0.9 ng/mL** 306 **Hg 0.9 – 2.7 ng/mL** 302 **Hg 2.7 – 385.8 ng/mL** 241 -0.062 (-0.133, 0.009)	**Hg 0.2 – 0.9 ng/mL** 42.3 **Hg 0.9 – 2.7 ng/mL** 43.6 **Hg 2.7 – 385.8 ng/mL** 45.0 0.027 (-0.021, 0.076)	**Hg 0.2 – 0.9 ng/mL** 7.5 **Hg 0.9 – 2.7 ng/mL** 6.8 **Hg 2.7 – 385.8 ng/mL** 6.7 -0.013 (-0.052, 0.027)
Zeng et al. (2015), China [56]	Geometric mean (range)	N/A	51.67 (4.38 – 264.39)	146.29 (8.20 – 1196.78)	0.00 (0.00 – 83.41)	22.36 (1.00 – 51.00)
Mínguez-Alarcón et al. (2017), USA [60]	Mean (95% CI) β (95% CI)	**Hg 0.03 – 0.37 ppm** 2.92 (2.37, 3.48) **Hg 0.38 – 0.67 ppm** 2.41 (1.99, 2.84)	**Hg 0.03 – 0.37 ppm** 37.9 (27.6, 51.9) **Hg 0.38 – 0.67 ppm** 48.0 (32.6, 70.9)	**Hg 0.03 – 0.37 ppm** 96.2 (72.5, 128) **Hg 0.38 – 0.67 ppm** 99.3 (69.4, 142)	**Hg 0.03 – 0.37 ppm** 20.5 (17.0, 24.1) **Hg 0.38 – 0.67 ppm** 26.9 (21.2, 32.6)	**Hg 0.03 – 0.37 ppm** 6.0 (4.8, 7.2) **Hg 0.38 – 0.67 ppm** 6.5 (5.4, 7.6)

* p-value < 0.05 when compared that quartile with the lowest quartile of exposure	**Hg 0.70 – 1.25 ppm** 2.89 (2.48, 3.30)	**Hg 0.70 – 1.25 ppm** 43.6 (33.2,57.4)	**Hg 0.70 – 1.25 ppm** 114 (85.8, 151)	**Hg 0.70 – 1.25 ppm** 22.6 (17.9, 27.4)	**Hg 0.70 – 1.25 ppm** 5.7 (4.6, 6.8)
	Hg 1.26 – 8.01 ppm 2.94 (2.61, 3.27)	**Hg 1.26 – 8.01 ppm** 60.9 (48.1, 77.2)*	**Hg 1.26 – 8.01 ppm** 167 (129, 217)*	**Hg 1.26 – 8.01 ppm** 29.2 (24.8, 33.7)*	**Hg 1.26 – 8.01 ppm** 6.3 (5.0, 7.5)
	0.01 (-0.19, 0.21) $p = 0.92$	19.2 (9.2, 29.2) $p = 0.0002$	22.8 (9.3, 36.3) $p = 0.001$	3.4 (1.9, 4.9) $p < 0.0001$	0.59 (-0.28, 1.47) $p = 0.18$

N/A – not applicable; NS – no significant; CI – confidence interval; USA – United States of America

3.3.2. Effect of Hg exposure on sperm DNA

Sperm deoxyribonucleic acid (DNA) damage can be considered a marker of sperm function that, together with the conventional semen analysis predicts male fertility [69]. Few human studies examined the association between Hg exposure and sperm DNA damage [34,58]. Rignell-Hydbom et al. [34] assessed if Hg exposure affected male reproductive function through sperm DNA integrity. For that, authors measured the sperm chromatin integrity and expressed the extend of DNA denaturation in terms of DNA fragmentation index (DFI). Moreover, authors evaluated the fraction of high DNA stainable (HDS) cells, which represents immature spermatozoa with incomplete chromatin condensation. No significant associations between Hg and DFI or HDS were found. However, authors found that DFI tends to increase with increasing Hg concentrations, suggesting a prejudicial effect of Hg on sperm chromatin integrity.

Additionally, Zhou et al. [58] evaluated the effect of Hg exposure in sperm DNA damage among men recruited from a fertility clinic. Authors showed that urine Hg levels were associated with increasing trends for DNA tail length, suggesting that environmental Hg exposure may result in increased sperm DNA damage. Moreover, authors suggested that Hg-induced sperm DNA damage may be triggered by generation of reactive oxygen species (ROS) and alteration of the antioxidant defense system. However, this study has several limitations such as the fact that men were only recruited from a fertility clinic, not representing the general population and authors only collected a single urine sample from each subject to assess Hg levels which may not reflect the subject's exposure.

3.3.3. Effect of Hg exposure on male endocrine system

The normal function of the negative feedback mechanism between the hypothalamus, the pituitary and the testis is required for normal testosterone synthesis and sperm production

[70]. The secretion of gonadotrophin-releasing hormone (GnRH) by the hypothalamus stimulates the synthesis of luteinizing hormone (LH) and follicle-stimulating hormone (FSH) in the pituitary that, in turn, induces the testosterone production and spermatogenesis in testis. This increase, in turn, increases androgen-binding protein and sex hormone-binding globulin (SHBG) protein synthesis. When the levels of testosterone are adequate, a negative feedback mechanism is responsible for decreasing both hypothalamic GnRH secretion and LH and FSH release into the bloodstream. Animal and human evidence suggests that heavy metals exposure (e.g. Hg, lead, cadmium, etc.) may have adverse impact on male endocrine system and thus may play a role in the increasing fertility problems [31]. Thus, this section summarizes data concerning the relationship between Hg exposure and male endocrine hormones. Eight studies investigated the effect of Hg on male endocrine system [21,23,29,39,44,49,50,54].

Approximately 60% of serum testosterone is associated with SHBG [71]. Subjects occupationally exposed to Hg showed a significant negative correlation between SHBG and years of Hg exposure ($r = -0.440$, $p = 0.008$) [21]. This suggests higher levels of bioavailable testosterone. In accordance with this, a study performed by Barregârd et al. [23] showed that serum total testosterone was positively correlated with cumulative Hg exposure ($r = 0.49$, $p = 0.002$).

Authors found that inhibin B serum levels increased with increasing blood Hg concentrations and with Hg exposure [49,54]. Increasing levels of inhibin B was related with high Sertoli cells activity and was correlated with suppression of FSH secretion and higher sperm concentration and counts [70].

In occupationally exposed subjects, thyroid-stimulating hormone (TSH) showed a positive correlation ($r = 0.397$, $p = 0.017$) with blood Hg [21]. Contrary results were

obtained by Barregârd et al. [23] that described a negative correlation between serum TSH and cumulative exposure index (r = -0.36, p = 0.03).

Prolactin is a hormone produced in the pituitary gland that helps the regulation of testosterone production. Elevated levels of prolactin have been associated with male infertility, once the hormone suppresses testosterone synthesis [74]. Subjects occupationally exposed to Hg showed a trend to a negative association between prolactin and years of Hg exposure [20]. By contrast, Barregârd et al. [22] showed slightly lower concentration of prolactin in exposed workers.

There are few evidences that Hg exposure causes alterations in the male endocrine system. However, how Hg exert its effects is still unknown. In a number of studies the levels of hormones LH, FSH, SHBG, testosterone, free androgen index, inhibin B, estradiol and prolactin were not significantly related to Hg exposure [21,29,39,44,49,50,54]. These results may be explained by the small number of participants in those studies. Larger studies involving other populations would be desirable.

3.4. Female fertility

Some epidemiological and experimental studies examined the associations and the effects of Hg exposure on female reproductive outcomes [16,18,24,27,28,31–33,35,36,38,40–43,45,47,48,52,53,55,57]. Positive correlations between Hg concentrations in blood and hair and unexplained infertility (r = 0.48; p < 0.01) were found [31,57]. On contrary, Hg concentrations was not detected in the endometrial samples of infertile or fertile women [52]. Several studies suggested that women environmentally and occupationally exposed to Hg had reduced fecundability, higher prevalence and incidence rates of menstrual disorders and longer TTP [16,18,24,27,33,48]. Moreover, the prevalence of menstrual

disorders in Hg-exposed women was significantly associated with the number of years working in the dental professional area [18].

Sikorski et al. [18] showed that adverse reproductive outcomes (e.g. spontaneous abortion, stillbirth and congenital malformation) in Hg exposed women were significantly associated with Hg levels determined in their scalp and pubic hair. Moreover, there are evidences that Hg levels were related with the higher risk of preterm birth among women with low to moderate exposure [35]. In contrast, some studies reported no associations between Hg concentrations and reduced fecundability, infertility and longer TTP [28,32,47,52,55].

3.4.1. Impact of Hg concentrations on ovarian follicle numbers and IVF outcomes

To assess the effect of Hg exposure on pregnancy and fertilization rate, Al-Saleh et al. [36] measured the levels of Hg in both blood and follicular fluid samples collected from women undergoing IVF treatment. Results showed a positive correlation between Hg levels in blood and in follicular fluid ($r = 0.25$, $p = 0$) and a negative correlation between blood Hg levels and the number of eggs ($r = - 0.09$, $p = 0.03$). Hg concentration in hair of women exposed to Hg was negatively correlated with the ovarian response to gonadotrophin stimulation after an IVF cycle, namely with the follicle count ($\beta=0.19$; $p=0.19$) and the number of oocytes ($\beta=0.38$, $p<0.05$) retrieved after ovarian stimulation [42].

However, some studies reported no correlations between Hg exposure in female subjects and embryo fragmentation, ovarian stimulation outcomes or fertilization rate, oocyte maturity, pregnancy loss or live birth rate [40,41,55].

3.4.2. Endocrine system

As previously described, there are several reports of the adverse effect of Hg on female fertility. However, how Hg adversely impacts female fertility remains unknown. Female reproduction is a process regulated by hormones and is susceptible to the effects of exposure to endocrine disrupting chemicals. Some studies reported that Hg may influence physiologic levels of female reproductive hormones [27,38,43,45,53]. Gerhard et al. [27] reported that women with thyroid dysfunction and polycystic ovary (PCO) syndrome showed a higher urine Hg excretion compared to controls without hormonal disorders. Moreover, elevated Hg excretion was associated with secondary infertility and luteal insufficiency or hyperandrogenemia [27]. Increasing Hg exposure was also associated with a decrease in LH, estradiol and progesterone levels [38,45]. Lei et al. [53] showed that prolactin levels were significantly lower in infertile women with higher Hg levels. These observations suggest that Hg may have a significant impact on female reproduction. However, no significant associations were found between Hg blood concentrations and FSH levels [43,53].

3.4.3. DNA methylation changes

Animal studies suggest that the toxic reproductive effects of Hg may be associated with epigenetic changes, namely, alterations in DNA methylation. To confirm this assumption, Hanna et al. [46] investigated the association of DNA methylation and Hg exposure, in women undergoing IVF treatment. In brief, authors evaluated the methylation at the CpG sites by the GSTM1_P266 Illumina probe that hybridized to the promoter regions of both the *GMST1* and *GMST5*. A trend of increased methylation in the gene promoters of *GMST1* and *GMST5* in women with the higher Hg exposure level was found. Then, authors investigated the nature of this methylation change by exploiting a deletion polymorphism, which encompasses the *GMST1* gene and found that the trend of increased

methylation with increased Hg exposure was limited to those women with a null *GMST1* genotype. The GSTM1 null allele has been associated with recurrent pregnancy loss in Japanese women and has been suggested to play a role on human reproduction [72]. Moreover, *GSTM1* is a member of the GSTM gene family, which encodes for enzymes that are involved in the mediation of oxidative stress [73]. To our knowledge the influence of *GSTM5* on human reproduction has not been studied, but possibly might play a role in reproductive outcomes due to the importance of ROS for human reproduction.

4. Discussion

Occupational and environmental pollutants exposure (e.g. Hg) may be one of the possible causes for the increase of adverse reproductive outcomes in modern societies [74,75]. According to this assumption, some authors explored the relationship between Hg concentrations and decreased human fertility. We performed a systematic review of studies focused on the evaluation of the effects of Hg exposure on human fertility and adverse reproductive outcomes (Figure 2). Papers showed that seafood consumption, alcohol and coffee intake, smoking habit and use of dental amalgam fillings are the main sources of Hg exposure and are positively associated with Hg levels in human samples in infertile subjects. Moreover, infertile subjects show higher levels of Hg than fertile ones, namely in the hair and blood [26,31,57].

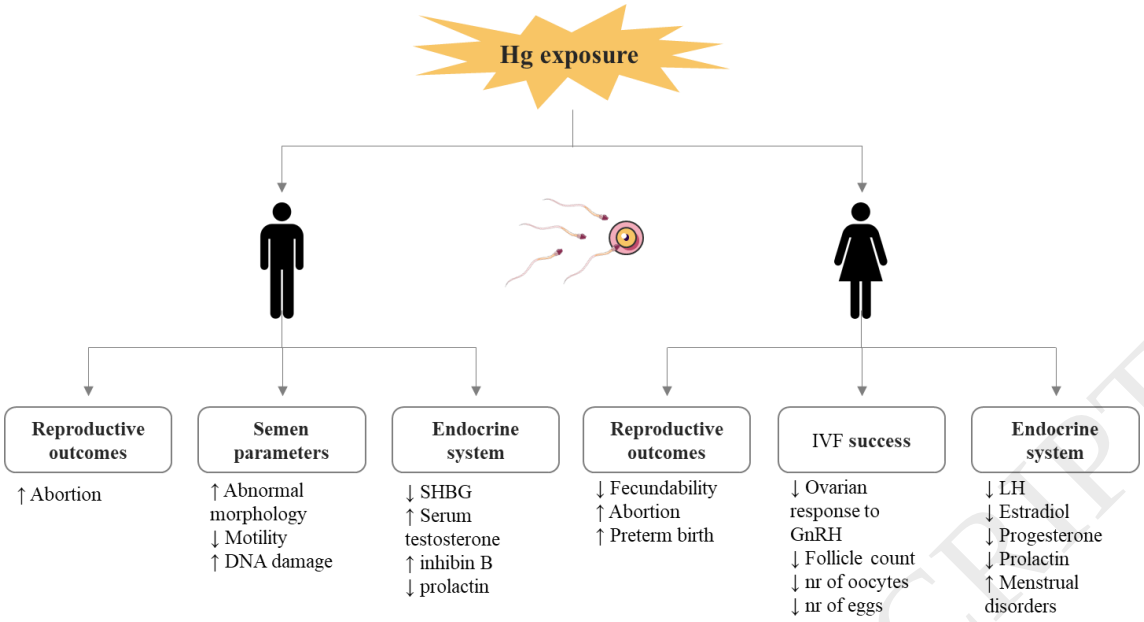

Figure 2 – Reproductive outcomes associated to environmental and occupational Hg exposure.

Eight papers studied the correlation between Hg levels in several male samples including semen [22,29,30,37,44,56,58,60]. Leung et al. [29] and Choy et al. [30] showed that semen quality parameters were significantly lower in subjects with higher levels of Hg in the body. Dickman et al. [26] revealed that infertile men had higher levels of Hg in the hair than fertile men. Additionally, Zhou et al. [58] detected that Hg exposure induced sperm DNA damage, a marker of sperm function and some authors showed that higher Hg concentrations were associated with longer TTP. A positive association between urinary Hg levels and sperm DNA damage was found. In general, these authors found that Hg exposure may decrease the quality of semen. However, contrary to previous studies, Mínguez-Alarcón et al. [60] observed that higher Hg levels were positively correlated with better semen quality parameters and do not support the hypothesis that Hg is harmful to men's reproductive health. Those contrary results were only among men with high fish intake. Better semen quality was linked to higher intake of fish possibly due to the beneficial effects of ω-3 polyunsaturated fatty acids [69–72].

Negative associations between Hg levels in female biological samples and quality of embryos, ovarian response to gonadotrophin, follicle count and the number of oocytes were also found, suggesting that Hg may influence negatively the IVF treatments. Furthermore, menstrual disorders and adverse reproductive outcomes, such as spontaneous abortions, malformations and stillbirth, were associated with higher levels of maternal Hg exposure. This allows to suggest that Hg is harmful to IVF treatment outcomes.

Our review also showed that Hg influences the female and male endocrine system, which could lead to a decrease in ovarian and testicular function and impair human fertility. A decrease in LH, estradiol, progesterone and prolactin levels were found in females with higher Hg exposure. In men exposed to Hg, authors found a decrease in the levels of SHBG and prolactin and an increase in the levels of serum testosterone and inhibin B. These hormonal alterations may explain the increase in menstrual disorders and adverse reproductive outcomes and the decrease in fecundability and IVF success observed in women exposed to Hg. Moreover, the deregulation of male endocrine system may explain the decrease in the semen quality parameters.

Although the potential adverse effect of Hg exposure showed by our review, several limitations may be pointed out. The large heterogeneity in the articles retrieved made it impossible to perform a meta-analysis across all studies. The second limitation is that many of the studies included relatively small sample sizes. Further, most publications included in this systematic review evaluated multiple parameters and biological samples, but not all studies evaluated the same parameters, making it difficult to draw strong conclusions. The last limitation was the lack of molecular studies to justify the adverse effects of Hg on human reproductive health. Thus, studies evaluating the effects of Hg at

the molecular level are needed as they may provide an insight on the molecular mechanism of Hg infertility-related.

5. Conclusions

In conclusion, the results yielded by our search showed that increased Hg levels were associated with infertility or subfertility status. In males, Hg exhibited adverse effects on semen quality parameters and induced sperm DNA damage. In females, Hg levels were associated with longer TTP and adverse reproductive outcomes such as spontaneous abortions and malformations and increased rates of menstrual disorders. A possible reason for this adverse effect of Hg on human reproductive health is the fact that Hg may be an endocrine disruptor leading to hormonal disorders which could lead to a decrease in ovarian and testicular function and impair human fertility. However, the molecular mechanisms underlying Hg effects on human fertility remain unknown. It is imperative further investigate the molecular effects of Hg exposure on the male and the female reproductive system.

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References

1. Zegers-Hochschild F, Adamson GD, de Mouzon J, Ishihara O, Mansour R, Nygren K, Sullivan E, van der Poel S, International Committee for Monitoring Assisted Reproductive Technology, World Health Organization. The International Committee for Monitoring Assisted Reproductive Technology (ICMART) and the World Health Organization (WHO) Revised Glossary on ART Terminology, 2009. Hum Reprod [Internet]. 2009 Nov 1 [cited 2018 Jan 23];24(11):2683–7. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19801627
2. Jenardhanan P, Panneerselvam M, Mathur PP. Effect of environmental contaminants on spermatogenesis. Semin Cell Dev Biol [Internet]. 2016 Nov [cited 2018 Feb 6];59:126–40. Available from: http://www.ncbi.nlm.nih.gov/pubmed/27060550
3. Rattan S, Zhou C, Chiang C, Mahalingam S, Brehm E, Flaws JA. Exposure to endocrine disruptors during adulthood: consequences for female fertility. J Endocrinol [Internet]. 2017 Jun [cited 2018 Feb 6];233(3):R109–29. Available from: http://www.ncbi.nlm.nih.gov/pubmed/28356401
4. Silva JV, Cruz D, Gomes M, Correia BR, Freitas MJ, Sousa L, Silva V, Fardilha M. Study on the short-term effects of increased alcohol and cigarette consumption in healthy young

men's seminal quality. Sci Rep [Internet]. 2017 Apr 3 [cited 2018 Jan 26];7:45457. Available from: http://www.ncbi.nlm.nih.gov/pubmed/28367956

5. Ricci E, Viganò P, Cipriani S, Somigliana E, Chiaffarino F, Bulfoni A, Parazzini F. Coffee and caffeine intake and male infertility: a systematic review. Nutr J [Internet]. 2017 Dec 24 [cited 2018 Jan 30];16(1):37. Available from: http://www.ncbi.nlm.nih.gov/pubmed/28646871
6. US EPA O. Basic Information about Mercury [Internet]. [cited 2018 Jan 30]. Available from: https://www.epa.gov/mercury/basic-information-about-mercury
7. Bank MS. Mercury in the environment: pattern and process. Berkeley: Univ of California Press; 2012.
8. Ask K, Akesson A, Berglund M, Vahter M. Inorganic mercury and methylmercury in placentas of Swedish women. Environ Health Perspect. 2002;110(5):523–6.
9. Al-Saleh I, Shinwari N, Mashhour A, Mohamed GED, Rabah A. Heavy metals (lead, cadmium and mercury) in maternal, cord blood and placenta of healthy women. Int J Hyg Environ Health [Internet]. 2011;214(2):79–101. Available from: http://dx.doi.org/10.1016/j.ijheh.2010.10.001
10. Fossato da Silva DA, Teixeira CT, Scarano WR, Favareto APA, Fernandez CDB, Grotto D, Barbosa F, Kempinas WDG. Effects of methylmercury on male reproductive functions in Wistar rats. Reprod Toxicol [Internet]. 2011 May [cited 2018 Feb 6];31(4):431–9. Available from: http://www.ncbi.nlm.nih.gov/pubmed/21262343
11. Boujbiha MAM, Hamden K, Guermazi F, Bouslama A, Omezzine A, El Feki A. Impairment of Spermatogenesis in Rats by Mercuric Chloride: Involvement of Low 17β-Estradiol Level in Induction of Acute Oxidative Stress. Biol Trace Elem Res [Internet]. 2011 Sep 7 [cited 2018 Feb 6];142(3):598–610. Available from: http://www.ncbi.nlm.nih.gov/pubmed/20820944
12. Martinez CS, Escobar AG, Torres JGD, Brum DS, Santos FW, Alonso MJ, Salaices M, Vassallo D V., Peçanha FM, Leivas FG, Wiggers GA. Chronic Exposure to Low Doses of Mercury Impairs Sperm Quality and Induces Oxidative Stress in Rats. J Toxicol Environ

Heal Part A [Internet]. 2014 Feb 20 [cited 2018 Feb 6];77(1–3):143–54. Available from: http://www.ncbi.nlm.nih.gov/pubmed/24555655

13. Al-Saleh I, Shinwari N, Al-Amodi M. Accumulation of Mercury in Ovaries of Mice After the Application of Skin-lightening Creams. Biol Trace Elem Res [Internet]. 2009 Oct 18 [cited 2018 Nov 30];131(1):43–54. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19224137

14. Takahashi Y, Tsuruta S, Arimoto M, Tanaka H, Yoshida M. Placental transfer of mercury in pregnant rats which received dental amalgam restorations. Toxicology [Internet]. 2003 Mar 14 [cited 2018 Feb 6];185(1–2):23–33. Available from: http://www.ncbi.nlm.nih.gov/pubmed/12505442

15. Moher D, Liberati A, Tetzlaff J, Altman DG, PRISMA Group. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. BMJ [Internet]. 2009 Jul 21 [cited 2018 Mar 26];339:b2535. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19622551

16. De Rosis F, Anastasio SP, Selvaggi L, Beltrame A, Moriani G. Female reproductive health in two lamp factories: effects of exposure to inorganic mercury vapour and stress factors. Br J Ind Med [Internet]. 1985 Jul [cited 2018 Oct 31];42(7):488–94. Available from: http://www.ncbi.nlm.nih.gov/pubmed/4015997

17. Lauwerys R, Roels H, Genet P, Toussaint G, Bouckaert A, De Cooman S. Fertility of male workers exposed to mercury vapor or to manganese dust: a questionnaire study. Am J Ind Med [Internet]. 1985 [cited 2018 Oct 31];7(2):171–6. Available from: http://www.ncbi.nlm.nih.gov/pubmed/3976664

18. Sikorski R, Juszkiewicz T, Paszkowski T, Szprengier-Juszkiewicz T. Women in dental surgeries: reproductive hazards in occupational exposure to metallic mercury. Int Arch Occup Environ Health. 1987;59:551–7.

19. Alcser KH, Brix KA, Fine LJ, Kallenbach LR, Wolfe RA. Occupational Mercury Exposure and Male Reproductive Health. Am J Ind Med. 1989;15:517–29.

20. Cordier S, Deplan F, Mandereau L, Hemon D. Paternal exposure to mercury and

spontaneous abortions. Br J Ind Med. 1991;48:375–81.

21. McGregor AJ, Mason HJ. Occupational mercury vapour exposure and testicular, pituitary and thyroid endocrine function. Hum Exp Toxicol [Internet]. 1991 May 2 [cited 2018 Oct 31];10(3):199–203. Available from: http://journals.sagepub.com/doi/10.1177/096032719101000309

22. Chia SE, Ong CN, Lee ST, Tsakok FH. Blood concentrations of lead, cadmium, mercury, zinc, and copper and human semen parameters. Arch Androl. 1992;29(2):177–83.

23. Barregård L, Lindstedt G, Schütz A, Sällsten G. Endocrine function in mercury exposed chloralkali workers. Occup Environ Med [Internet]. 1994 Aug [cited 2018 Oct 31];51(8):536–40. Available from: http://www.ncbi.nlm.nih.gov/pubmed/7951778

24. Rowland AS, Baird DD, Weinberg CR, Shore DL, Shy CM, Wilcox AJ. The effect of occupational exposure to mercury vapour on the fertility of female dental assistants. Occup Environ Med [Internet]. 1994 Jan [cited 2018 Oct 31];51(1):28–34. Available from: http://www.ncbi.nlm.nih.gov/pubmed/8124459

25. Hanf V, Forstmann A, Costea JE, Schieferstein G, Fischer I, Schweinsberg F. Mercury in urine and ejaculate in husbands of barren couples. Toxicol Lett. 1996;88:227–31.

26. Dickman MD, Leung CKM, Leong MKH. Hong Kong male subfertility links to mercury in human hair and fish. Sci Total Environ. 1998;214:165–74.

27. Gerhard I, Monga B, Waldbrenner A, Runnebaum B. Heavy metals and fertility. J Toxicol Environ Heal - Part A [Internet]. 1998 Aug 15 [cited 2018 Oct 31];54(8):593–611. Available from: http://www.tandfonline.com/doi/abs/10.1080/009841098158638

28. Dahl JE, Sundby J, Hensten-Pettersen A, Jacobsen N. Dental workplace exposure and effect on fertility. Scand J Work Environ Heal [Internet]. 1999 Jun [cited 2018 Oct 31];25(3):285–90. Available from: http://www.ncbi.nlm.nih.gov/pubmed/10450781

29. Leung TY, Choy CMY, Yim SF, Lam CWK, Haines CJ. Whole blood mercury concentrations in sub-fertile men in Hong Kong. Aust New Zeal J Obstet Gynaecol. 2001;41(1):75–7.

30. Choy CMY, Yeung QSY, Briton-Jones CM, Cheung CK, Lam CWK, Haines CJ.

Relationship between semen parameters and mercury concentrations in blood and in seminal fluid from subfertile males in Hong Kong [2]. Fertil Steril. 2002;78(2):426–8.

31. Choy CMY, Lam CWK, Cheung LTFP, Briton-Jones CM, Cheung LTFP, Haines CJ. Infertility, blood mercury concentrations and dietary seafood consumption: A case-control study. BJOG An Int J Obstet Gynaecol [Internet]. 2002 Oct [cited 2018 Jan 30];109(10):1121–5. Available from: http://www.ncbi.nlm.nih.gov/pubmed/12387464

32. Arakawa C, Yoshinaga J, Okamura K, Nakai K, Satoh H. Fish consumption and time to pregnancy in Japanese women. Int J Hyg Environ Health. 2006;209:337–44.

33. Cole DC, Wainman B, Sanin LH, Weber JP, Muggah H, Ibrahim S. Environmental contaminant levels and fecundability among non-smoking couples. Reprod Toxicol. 2006;22:13–9.

34. Rignell-Hydbom A, Axmon A, Lundh T, Jönsson BA, Tiido T, Spano M. Dietary exposure to methyl mercury and PCB and the associations with semen parameters among Swedish fishermen. Environ Heal [Internet]. 2007 Dec 8 [cited 2018 Nov 5];6(1):14. Available from: http://ehjournal.biomedcentral.com/articles/10.1186/1476-069X-6-14

35. Xue F, Holzman C, Rahbar MH, Trosko K, Fischer L. Maternal fish consumption, mercury levels, and risk of preterm delivery. Environ Health Perspect. 2007;115(1):42–7.

36. Al-Saleh I, Coskun S, Mashhour A, Shinwari N, El-Doush I, Billedo G, Jaroudi K, Al-Shahrani A, Al-Kabra M, El Din Mohamed G. Exposure to heavy metals (lead, cadmium and mercury) and its effect on the outcome of in-vitro fertilization treatment. Int J Hyg Environ Health. 2008;211(5–6):560–79.

37. Meeker JD, Rossano MG, Protas B, Diamond MP, Puscheck E, Daly D, Paneth N, Wirth JJ. Cadmium, lead, and other metals in relation to semen quality: Human evidence for molybdenum as a male reproductive toxicant. Environ Health Perspect. 2008;116(11):1473–9.

38. Laks DR. Assessment of chronic mercury exposure within the U.S. population, national health and nutrition examination survey, 1999-2006. BioMetals [Internet]. 2009 Dec 21 [cited 2018 Nov 13];22(6):1103–14. Available from:

http://www.ncbi.nlm.nih.gov/pubmed/19697139

39. Meeker JD, Rossano MG, Protas B, Padmanahban V, Diamond MP, Puscheck E, Daly D, Paneth N, Wirth JJ. Environmental exposure to metals and male reproductive hormones: circulating testosterone is inversely associated with blood molybdenum. Fertil Steril [Internet]. 2010 Jan [cited 2018 Nov 12];93(1):130–40. Available from: http://www.ncbi.nlm.nih.gov/pubmed/18990371

40. Bloom MS, Parsons PJ, Steuerwald AJ, Schisterman EF, Browne RW, Kim K, Coccaro GA, Conti GC, Narayan N, Fujimoto VY. Toxic trace metals and human oocytes during in vitro fertilization (IVF). Reprod Toxicol [Internet]. 2010 Jun [cited 2018 Nov 5];29(3):298–305. Available from: http://www.ncbi.nlm.nih.gov/pubmed/20096775

41. Bloom MS, Parsons PJ, Kim D, Steuerwald AJ, Vaccari S, Cheng G, Fujimoto VY. Toxic trace metals and embryo quality indicators during in vitro fertilization (IVF). Reprod Toxicol [Internet]. 2011;31(2):164–70. Available from: http://dx.doi.org/10.1016/j.reprotox.2010.11.011

42. Dickerson EH, Sathyapalan T, Knight R, Maguiness SM, Killick SR, Robinson J, Atkin SL. Endocrine disruptor & nutritional effects of heavy metals in ovarian hyperstimulation. J Assist Reprod Genet. 2011;28(12):1223–8.

43. Jackson LW, Howards PP, Wactawski-Wende J, Schisterman EF. The association between cadmium, lead and mercury blood levels and reproductive hormones among healthy, premenopausal women. Hum Reprod [Internet]. 2011 Oct 1 [cited 2018 Nov 5];26(10):2887–95. Available from: http://www.ncbi.nlm.nih.gov/pubmed/21778284

44. Mendiola J, Moreno JM, Roca M, Vergara-Juárez N, Martínez-García MJ, García-Sánchez A, Elvira-Rendueles B, Moreno-Grau S, López-Espín JJ, Ten J, Bernabeu R, Torres-Cantero AM. Relationships between heavy metal concentrations in three different body fluids and male reproductive parameters: a pilot study. Environ Heal [Internet]. 2011;10(6):1–7. Available from: http://ehjournal.biomedcentral.com/articles/10.1186/1476-069X-10-6

45. Pollack AZ, Schisterman EF, Goldman LR, Mumford SL, Albert PS, Jones RL,

Wactawski-Wende J. Cadmium, lead, and mercury in relation to reproductive hormones and anovulation in premenopausal women. Environ Health Perspect [Internet]. 2011 Aug [cited 2018 Nov 5];119(8):1156–61. Available from: http://www.ncbi.nlm.nih.gov/pubmed/21543284

46. Hanna CW, Bloom MS, Robinson WP, Kim D, Parsons PJ, Vom Saal FS, Taylor JA, Steuerwald AJ, Fujimoto VY. DNA methylation changes in whole blood is associated with exposure to the environmental contaminants, mercury, lead, cadmium and bisphenol A, in women undergoing ovarian stimulation for IVF. Hum Reprod [Internet]. 2012 May [cited 2018 Nov 6];27(5):1401–10. Available from: http://www.ncbi.nlm.nih.gov/pubmed/22381621

47. Louis GMB, Sundaram R, Schisterman EF, Sweeney AM, Lynch CD, Gore-Langton RE, Chen Z, Kim S, Caldwell K, Barr DB. Heavy Metals and Couple Fecundity, the LIFE Study. Chemosphere. 2012;87(11):1201–7.

48. Chevrier C, Warembourg C, Gaudreau E, Monfort C, Le Blanc A, Guldner L, Cordier S. Organochlorine pesticides, polychlorinated biphenyls, seafood consumption, and time-to-pregnancy. Epidemiology. 2013;24(2):251–60.

49. Mocevic E, Specht IO, Marott JL, Giwercman A, Jönsson BAG, Toft G, Lundh T, Peter Bonde J. Environmental mercury exposure, semen quality and reproductive hormones in Greenlandic Inuit and European men: A cross-sectional study. Asian J Androl [Internet]. 2013 Jan 10 [cited 2018 Nov 6];15(1):97–104. Available from: http://www.asiaandro.com/Abstract.asp?doi=10.1038/aja.2012.121

50. Zeng Q, Zhou B, Feng W, Wang YX, Liu AL, Yue J, Li YF, Lu WQ. Associations of urinary metal concentrations and circulating testosterone in Chinese men. Reprod Toxicol [Internet]. 2013;41:109–14. Available from: http://dx.doi.org/10.1016/j.reprotox.2013.06.062

51. Kim K, Bloom MS, Kruger PC, Parsons PJ, Arnason JG, Byun Y, Goins S, Fujimoto VY. Toxic metals in seminal plasma and in vitro fertilization (IVF) outcomes. Environ Res [Internet]. 2014;133:334–7. Available from:

http://dx.doi.org/10.1016/j.envres.2014.06.014

52. Tanrikut E, Karaer A, Celik O, Celik E, Otlu B, Yilmaz E, Ozgul O. Role of endometrial concentrations of heavy metals (cadmium, lead, mercury and arsenic) in the aetiology of unexplained infertility. Eur J Obstet Gynecol Reprod Biol. 2014;179:187–90.

53. Lei H-L, Wei H-J, Chen P-H, Hsi H-C, Chien L-C. Preliminary study of blood methylmercury effects on reproductive hormones and relevant factors among infertile and pregnant women in Taiwan. Chemosphere [Internet]. 2015;135:411–7. Available from: http://linkinghub.elsevier.com/retrieve/pii/S0045653515004634

54. Lenters V, Portengen L, Smit LAM, Jönsson BAG, Giwercman A, Rylander L, Lindh CH, Spanò M, Pedersen HS, Ludwicki JK, Chumak L, Piersma AH, Toft G, Bonde JP, Heederik D, Vermeulen R. Phthalates, perfluoroalkyl acids, metals and organochlorines and reproductive function: A multipollutant assessment in Greenlandic, Polish and Ukrainian men. Occup Environ Med [Internet]. 2015 Jun [cited 2018 Nov 6];72(6):385–93. Available from: http://oem.bmj.com/lookup/doi/10.1136/oemed-2014-102264

55. Wright DL, Afeiche MC, Ehrlich S, Smith K, Williams PL, Chavarro JE, Batsis M, Toth TL, Hauser R. Hair mercury concentrations and in vitro fertilization (IVF) outcomes among women from a fertility clinic. Reprod Toxicol. 2015;51:125–32.

56. Zeng Q, Feng W, Zhou B, Wang YX, He XS, Yang P, You L, Yue J, Li YF, Lu WQ. Urinary metal concentrations in relation to semen quality: A cross-sectional study in China. Environ Sci Technol. 2015;49(8):5052–9.

57. Hsi HC, Hsu YW, Chang TC, Chien LC. Methylmercury Concentration in Fish and Risk-Benefit Assessment of Fish Intake among Pregnant versus Infertile Women in Taiwan. Hu Y, editor. PLoS One [Internet]. 2016 May 17 [cited 2018 Nov 6];11(5):e0155704. Available from: http://dx.plos.org/10.1371/journal.pone.0155704

58. Zhou Y, Fu XM, He DL, Zou XM, Wu CQ, Guo WZ, Feng W. Evaluation of urinary metal concentrations and sperm DNA damage in infertile men from an infertility clinic. Environ Toxicol Pharmacol [Internet]. 2016;45:68–73. Available from: http://dx.doi.org/10.1016/j.etap.2016.05.020

59. Buck Louis GM, Smarr MM, Sundaram R, Steuerwald AJ, Sapra KJ, Lu Z, Parsons PJ. Low-level environmental metals and metalloids and incident pregnancy loss. Reprod Toxicol [Internet]. 2017;69:68–74. Available from: http://dx.doi.org/10.1016/j.reprotox.2017.01.011

60. Mínguez-Alarcón L, Afeiche MC, Williams PL, Arvizu M, Tanrikut C, Amarasiriwardena CJ, Ford JB, Hauser R, Chavarro JE. Hair mercury (Hg) levels, fish consumption and semen parameters among men attending a fertility center. Int J Hyg Environ Health [Internet]. 2017;30315–2(17):1438–4639. Available from: http://dx.doi.org/10.1016/j.ijheh.2017.10.014

61. Bloom MS, Buck Louis GM, Sundaram R, Kostyniak PJ, Jain J. Associations between blood metals and fecundity among women residing in New York State. Reprod Toxicol [Internet]. 2011 Feb [cited 2018 Nov 13];31(2):158–63. Available from: http://www.ncbi.nlm.nih.gov/pubmed/20933593

62. Skakkebaek NE, Rajpert-De Meyts E, Buck Louis GM, Toppari J, Andersson A-M, Eisenberg ML, Jensen TK, Jørgensen N, Swan SH, Sapra KJ, Ziebe S, Priskorn L, Juul A. Male Reproductive Disorders and Fertility Trends: Influences of Environment and Genetic Susceptibility. Physiol Rev [Internet]. 2016 Jan [cited 2018 Jan 29];96(1):55–97. Available from: http://www.ncbi.nlm.nih.gov/pubmed/26582516

63. Lafuente R, García-Blàquez N, Jacquemin B, Checa MA. Outdoor air pollution and sperm quality. Fertil Steril [Internet]. 2016 Sep 15 [cited 2018 Jan 30];106(4):880–96. Available from: http://www.ncbi.nlm.nih.gov/pubmed/27565259

64. Ricci E, Al-Beitawi S, Cipriani S, Alteri A, Chiaffarino F, Candiani M, Gerli S, Viganó P, Parazzini F. Dietary habits and semen parameters: a systematic narrative review. Andrology [Internet]. 2018 Jan [cited 2018 Jan 30];6(1):104–16. Available from: http://www.ncbi.nlm.nih.gov/pubmed/29266782

65. Boujbiha MA, Hamden K, Guermazi F, Bouslama A, Omezzine A, Kammoun A, Feki A El. Testicular toxicity in mercuric chloride treated rats: Association with oxidative stress. Reprod Toxicol [Internet]. 2009 Jul 1 [cited 2018 Nov 14];28(1):81–9. Available from:

https://www.sciencedirect.com/science/article/pii/S0890623809000690

66. Homma-Takeda S, Kugenuma Y, Iwamuro T, Kumagai Y, Shimojo N. Impairment of spermatogenesis in rats by methylmercury: involvement of stage- and cell- specific germ cell apoptosis. Toxicology [Internet]. 2001 Nov 1 [cited 2018 Nov 14];169(1):25–35. Available from: https://www.sciencedirect.com/science/article/pii/S0300483X01004875

67. World Health Organisation. Collection and examination of human semen. In: WHO laboratory manual for the examination of human semen and sperm-cervical mucus interaction. Cambridge: Cambridge University Press; 1999. p. 4–30.

68. Matthies J, Schwarz I, Donat H. [Effect of heavy metal ions on male fertility]. Zentralbl Gynakol [Internet]. 1989 [cited 2018 Jan 23];111(3):155–66. Available from: http://www.ncbi.nlm.nih.gov/pubmed/2718652

69. Agarwal A, Said TM. Role of sperm chromatin abnormalities and DNA damage in male infertility. Hum Reprod Update [Internet]. 2003 [cited 2018 Jan 23];9(4):331–45. Available from: http://www.ncbi.nlm.nih.gov/pubmed/12926527

70. Corradi PF, Corradi RB, Greene LW. Physiology of the Hypothalamic Pituitary Gonadal Axis in the Male. Urol Clin North Am [Internet]. 2016 May [cited 2018 Nov 19];43(2):151–62. Available from: http://www.ncbi.nlm.nih.gov/pubmed/27132572

71. Rabijewski M, Zgliczyński W. [Pathogenesis, evaluation and treatment of hypogonadism in men]. Endokrynol Pol [Internet]. 2009 [cited 2018 Nov 15];60(3):222–33. Available from: http://www.ncbi.nlm.nih.gov/pubmed/19569024

72. Sata F, Yamada H, Kondo T, Gong Y, Tozaki S, Kobashi G, Kato EH, Fujimoto S, Kishi R. Glutathione S-transferase M1 and T1 polymorphisms and the risk of recurrent pregnancy loss. Mol Hum Reprod [Internet]. 2003 Mar [cited 2018 Nov 9];9(3):165–9. Available from: http://www.ncbi.nlm.nih.gov/pubmed/12606593

73. Valko M, Leibfritz D, Moncol J, Cronin MTD, Mazur M, Telser J. Free radicals and antioxidants in normal physiological functions and human disease. Int J Biochem Cell Biol [Internet]. 2007 Jan [cited 2018 Nov 9];39(1):44–84. Available from: http://www.ncbi.nlm.nih.gov/pubmed/16978905

74. Kumar S. Occupational, Environmental and Lifestyle Factors Associated With Spontaneous Abortion. Reprod Sci [Internet]. 2011 Oct 29 [cited 2018 Feb 6];18(10):915–30. Available from: http://www.ncbi.nlm.nih.gov/pubmed/21960507

75. Kumar S, Sharma S, Thaker R. Occupational, environmental, and lifestyle factors and their contribution to preterm birth – An overview. Indian J Occup Environ Med [Internet]. 2017 [cited 2018 Feb 6];21(1):9. Available from: http://www.ncbi.nlm.nih.gov/pubmed/29391742