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Association of urinary bisphenol A concentrations with in vitro fertilisation outcomes: a systematic review and meta-analysis protocol

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ABSTRACT

Introduction Bisphenol A (BPA) is a common environmental endocrine disruptor. BPA has been reported to be associated with female infertility, which may not only affect natural pregnancy and natural fertility but also affect the outcomes of in vitro fertilisation (IVF). BPA exposure may help to partly explain the unsatisfactory IVF outcomes, but the relationship between the concentrations of BPA in urine and IVF outcomes remains controversial. Therefore, we will perform a meta-analysis to identify and review the relationship between urinary BPA concentrations and IVF outcomes.

Methods and analysis A comprehensive literature search will be performed in PubMed, Web of Science and the Cochrane central register of controlled trials for relevant articles using MeSH terms and related entry terms (up to 20 April 2022). The language will be restricted to English. Articles will be screened for inclusion in or exclusion from the study independently by two reviewers after removing the duplicates. The titles and abstracts followed by full-text screening will also be conducted independently by two reviewers. In addition, the references of the included literature will also be traced to supplement our search results and to obtain all relevant literature. The Newcastle-Ottawa Scale will be used to assess the methodological quality of the included studies using a star rating system ranging from 0 to 9 stars. Heterogeneity in estimates from different articles will be quantified, and publication bias will be investigated using funnel plots. Finally, a sensitivity analysis will also be conducted to estimate whether our results could have been markedly affected by a single included study.

Ethics and dissemination Ethical approval is not required for this protocol, as participants are not included. Findings will be disseminated through peer-reviewed publications and conference presentations.

STRENGTHS AND LIMITATIONS OF THIS STUDY

⇒ This is the first meta-analysis to explore the association of urinary bisphenol A (BPA) concentrations with in vitro fertilisation (IVF) outcomes.
⇒ Only articles published in English will be included, which may cause language bias.
⇒ Subgroup analysis based on urine BPA concentrations in individuals of different ethnicities and from different regions may help to determine how BPA affects IVF outcomes.
⇒ Comprehensive assessments may be difficult to establish since most studies are expected to be observational.

INTRODUCTION

The plastic monomer and plasticizer bisphenol A (BPA) is one of the highest volume chemicals produced worldwide and has been used in food packaging since the 1960s.[1] BPA is also used in the production of polycarbonate plastics used in many consumer products, including polycarbonate plastics, medical equipment, polymer-based dental fillings, glasses and personal care products.[2] BPA has been shown to leach from beverage containers under normal conditions of use, which has also been confirmed by the detection of BPA in human serum, urine, amniotic fluid, follicular fluid, placental tissue and umbilical cord blood.[3 4] The US Food and Drug Administration, Health Canada, the European Food Safety Authority and Food Standards Australia New Zealand all assert that current exposure to BPA provides no health hazards or safety concerns to persons of any age group (including unborn children, babies and pregnant women).[5] However, many studies have revealed that even low-level exposure to BPA can potentially have adverse effects on human reproduction and pregnancy by affecting the functions of the ovary, uterus and prostate and can reduce oocyte quality in women undergoing in vitro fertilisation (IVF).[6] Currently, there is limited and controversial evidence on the impact of BPA on adverse reproductive outcomes, and the available evidence-based options are only moderately effective.[6 7]

Studies have revealed that BPA can bind to oestrogen, androgen and thyroid receptors and, therefore, affect oestrogen, androgen and thyroid function in vivo.[8] In some cases,

the levels of total BPA (free and conjugated) in human body fluids are higher than the concentrations that have been reported to stimulate several molecular endpoints in cell culture in vitro.[2] Infertility affects 48.5 million couples worldwide, and significant progress has been made since the introduction of IVF 30 years ago.[9 10] However, the success rate of pregnancy is still relatively low in clinical IVF practice, at only approximately 40%–50% even if embryos with normal morphology are used for transfer.[11 12] Even after taking into account age, infertility type and drugs found to affect the pregnancy success rate of IVF, there are still some patients with unexplained IVF pregnancy failure.[13 14] In view of the universality of BPA in our daily life and the adverse effects of BPA on human reproductive health found in previous studies, studies on the association of BPA with IVF outcomes are also increasing; however, the exact relationship between the two is still unclear.[15 16]

Objectives

A systematic review and meta-analysis are needed to synthesise the evidence, given the potential for public reproduction health implications if BPA is associated with IVF outcomes. Therefore, we describe a protocol for conducting a meta-analysis to investigate whether BPA exposure is associated with IVF outcomes in men and women separately. We aim to provide evidence for further research on BPA exposure and IVF outcomes by reviewing published articles on BPA exposure and IVF outcomes.

METHODS AND ANALYSIS

Study design

This meta-analysis protocol is guided by the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) Protocols[17] (see online supplemental material 1). The search period will range from 1 April 2022 to 20 April 2022.

Search strategy

Studies published before 20 April 2022 will be searched through an electronic search of PubMed, Web of Science and Cochrane Library without language restrictions. MeSH and entry terms will be used to search relevant articles. In addition, manual search and reference searches will also be performed to enlarge the search range, and corresponding authors of the targeted article will be asked for more related original data to avoid potential missing data as much as possible. A detailed search strategy for PubMed is presented in table 1.

Table 1 Search strategy for PubMed

No	Mesh terms and entry terms
#1	((((((((((((Fertilization in Vitro) OR In Vitro Fertilization) OR In Vitro Fertilizations) OR Test-Tube Fertilization) OR Fertilization, Test-Tube) OR Fertilizations, Test-Tube) OR Test Tube Fertilization) OR Test-Tube Fertilizations) OR Fertilizations in Vitro) OR Test-Tube Babies) OR Babies, Test-Tube) OR Baby, Test-Tube) OR Test Tube Babies) OR Test-Tube Baby
#2	(((((bisphenol A) OR 4,4'-dihydroxy-2,2-diphenylpropane) OR diphenylolpropane) OR 2,2-bis(4-hydroxyphenyl)propane) OR bisphenol A, sodium salt) OR bisphenol A, disodium salt
#3	#1 and #2

Inclusion and exclusion criteria

Studies will be considered eligible for this meta-analysis if the patients, intervention, outcomes and study type meet all of the following criteria:

Study types

We will include observational studies or original epidemiological studies (including case–control, nested case–control, cross-sectional and cohort studies). All studies should involve the association of urinary BPA concentrations with IVF outcomes.

Participants

Patients who underwent IVF cycles and assessment of urinary BPA levels will be further assessed in the study. Donor egg, donor sperm or donor embryo cycles will not be included in the analysis.

Interventions

There will be intervention other than the intervention of the normal IVF procedure, and the group analysis will be carried out according to the concentrations of BPA in urine.

Other inclusion criteria

1. The exposure route of BPA for IVF women through daily contact.
2. Studies reported ORs or relative risks for IVF outcomes as well as their 95% CIs.
3. Urine samples were obtained for BPA exposure assessment before IVF.

Exclusion criteria

1. Without reusable data.
2. Intervention studies.
3. The association between urinary BPA concentrations and IVF outcomes was not studied.

Outcomes

The primary outcomes will be the number of oocytes retrieved, high-quality embryo rate, clinical pregnancy rate (CPR) and live birth rate.

1. The number of oocytes retrieved: the number of oocytes obtained in each controlled ovarian stimulation (COS) cycle.
2. High-quality embryos will be defined as grade 1 or grade 2 embryos, and the rate of good-quality embryos will be calculated as the ratio of the number of good-quality embryos to the total number of cleaved embryos.

3. CPR: the ratio of the number of clinical pregnancies to the total number of patients who underwent embryo transfer.

The secondary outcomes will be as follows:

1. Fertilisation rate: normal fertilisation will be identified by the presence of two pronuclei (2PN) at the time of fertilisation assessment. The ICSI fertilisation rate will be identified as 2PN/number of oocytes injected, and the conventional insemination fertilisation rate will be identified as 2PN/number of oocytes inseminated.
2. Blastocyst formation rate: the ratio of the number of blastocysts to the number of zygotes.
3. Embryo implantation rate: the ratio of the number of implanted embryos to the total number of transferred embryos.

Data collection and analysis

Selection of studies

Two authors (X-LC and N-XX) will independently search and screen the titles and abstracts to assess the eligibility of all potential studies according to the inclusion and exclusion criteria. Finally, the screening results of the two people will be summarised. Any disagreements will be resolved through discussion with a third reviewer (C-MX). A fourth and fifth reviewer (X-YZ) will check all procedures before and after data extraction. First, duplicate studies and those not meeting the inclusion criteria based on a review of the titles and abstracts will be excluded. Second, after reading the full text of each study, studies meeting the inclusion criteria but without reusable data will also be removed. Details of the entire selection procedure are shown in the PRISMA flow diagram (figure 1).

Data extraction

We will extract data from the final included studies following a data acquisition. The data will be recorded on worksheets (Excel; Microsoft, Redmond, Washington, USA). The following information will be extracted from each included study: the first author's last name, publication year, main outcomes, study type, study population, sample size, dilution adjustment method, exposure time, concentrations of urine BPA, covariate adjustment in the model, Newcastle-Ottawa Scale (NOS) and Grading of Recommendations, Assessment, Development and Evaluations (GRADE) scores and exposure contrast. The results will be recorded as the mean±SD for continuous variables and the proportion of participants with the percentage for dichotomous data. If necessary, original data will be obtained by contacting the corresponding authors of the literature by email. A detailed list of the information and data to be extracted is presented in online supplemental material 2).

Quality assessment

The quality of the methods for the included studies will be independently assessed by the two reviewers (X-LC

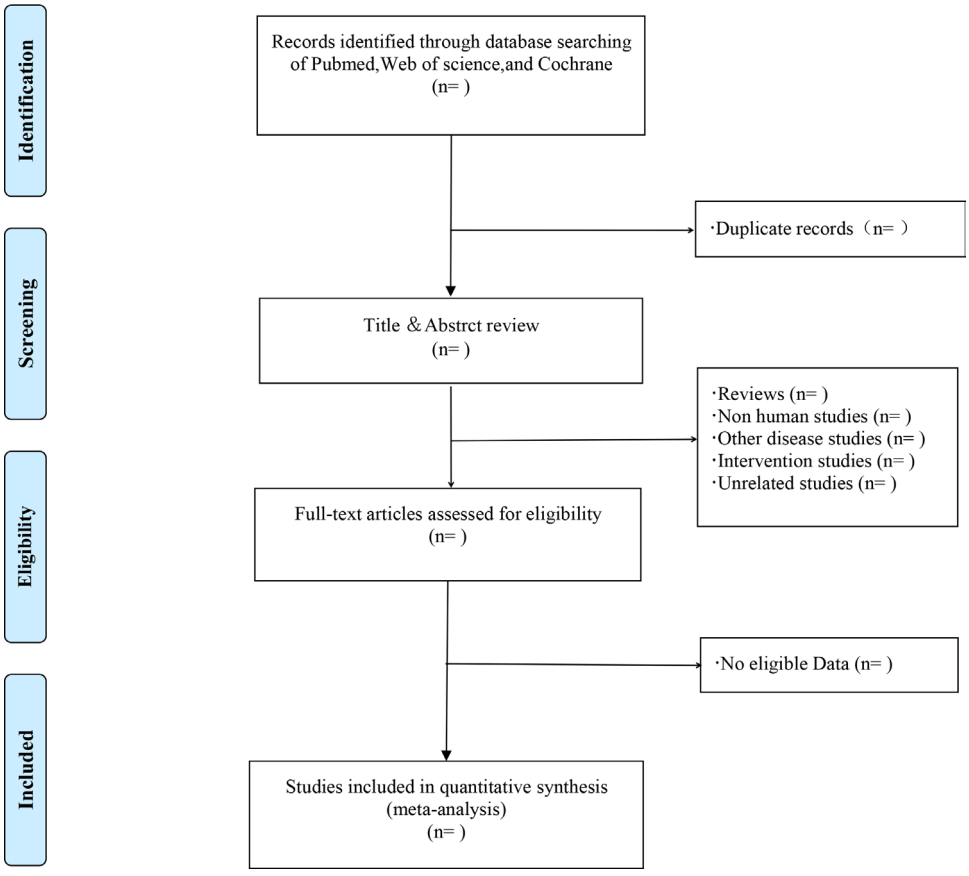

Figure 1 The PRISMA flow diagram. PRISMA, Preferred Reporting Items for Systematic Reviews and Meta-Analyses.

and N-XX) using the NOS.[18] In addition, the GRADE tool will also be used to grade the evidence quality of the major outcome indicators.[19] The NOS uses a star rating system to assess the study quality. The quality of the study will be assessed as low (<5 stars), moderate (5–7 stars) or high (≥8 stars).The risk of bias in the included studies will also be assessed according to the NOS. Discrepancies will be settled in the same manner as described above.

Assessment of heterogeneity and bias

Statistical heterogeneity will be assessed according to the Cochrane Q test by the standard χ^2 test (α=0.1) and I^2 test. If p≥0.05 and if I^2≤50%, a fixed-effects model will be used. If p<0.05 or I^2>50%, random-effects models will be used. If the heterogeneity is statistically significant, we will conduct a subgroup analysis to investigate the possible sources of heterogeneity.

Egger's publication bias plot will be used to visualise the publication bias graphically. The trim-and-fill method will be used if publication bias occurs. If the publication bias is still significant (p<0.05), we will carefully discuss it in the Discussion section, and we will also state it as one of the limitations of our meta-analysis.

Subgroup analysis

First, the data from men and women will be analysed separately. We will also perform a subgroup analysis based on the level of BPA exposure. The estimates for the various categories and the midpoints of the BPA in each category will be extracted if studies provide the association of BPA with the outcome as categorical data. In cases where the paper does not specify the highest or lowest bounds, known as 'open categories', we will use a number that is 15% higher or lower than the nearest available cut point.[20 21] The estimates and medians or geometric mean of BPA concentrations throughout the entire population will be collected from studies that exclusively assess the relationship between BPA exposure as a continuous variable. Then, using the BPA concentrations collected from the included studies, we will classify them into tertiles and perform a subgroup analysis to determine whether there is a dose-response trend.

Subgroup analyses based on the following pre-specified variables will be conducted to further investigate the cause of heterogeneity: adjustment techniques for urinary dilution concentration (adjusted by creatinine, urine specific gravity and no correction), study area (America, Europe and Asia), and research type (cohort, nested case–control and case–control).

Sensitivity analysis

We will perform a sensitivity analysis to assess the susceptibility of the included studies of this meta-analysis. We will aim to change the inclusion criteria, exclude low-quality studies or adopt different statistical methods to re-estimate the combined effect and compare it with the results of the meta-analysis before exclusion to explore the impact of a study on the combined effect and the robustness of our results.

Patient and public involvement

No patients will be involved.

Statistical analyses

All statistical analyses will be performed using Stata V.16 for Windows. The association between the exposure to BPA and IVF outcomes will be assessed by calculating the pooled OR and 95% CI. When quantitative analysis cannot be conducted, we will only describe the results. If the included literature only reports the OR value and 95% CI of IVF outcomes and BPA exposure or provides the OR value and accurate p value to calculate 95% CI, the effect size will be determined as lnOR, and the SE of effect size (SE) will be calculated as (upper limit of CI-lower limit of CI)/3.92.

DISCUSSION

Our protocol describes a method for the synthesis of current evidence on the association of urinary BPA concentrations with IVF outcomes. According to numerous toxicokinetic investigations on BPA in humans, BPA is rapidly eliminated from the body in the form of glucuronide conjugated BPA (BPA-G) in the urine within 24 hours of exposure.[22] Therefore, the most common biomarker of BPA exposure status is total (free/unconjugated plus conjugated) BPA in urine, which can be measured through biomonitoring studies using enzymatic (such as glucuronidase and sulfatase) treatments.[23] It is worth noting that studies measuring urinary BPA mainly use on-site urine samples rather than 24-hour urine samples or multipoint measurements. Point-sample measurement is affected by greater intraindividual and interindividual variability. However, BPA exposure seems to occur every day, and if the included sample size is relatively large, the potential deviation is irrelevant.

Although BPA threshold levels that signify a possible health concern have not been consistently identified, human biomonitoring-derived BPA cut-offs or biomonitoring equivalents (BEs) in urine have been proposed as practical reference values for people to contextualise the exposure level of diverse populations. The German Human Biomonitoring Commission defined the reference HBM-I value as 200 µg/L, and the US EPA defined the BE value as 2000 µg/L.[24] A previous meta-analysis demonstrated that higher BPA exposure was related to an increased risk of preterm birth and decreased length of gestational age and found that the third trimester of pregnancy may be a critical susceptible period.[25] Studies also indicated that both prenatal and concurrent exposure to BPA can be classified as endocrine-disrupting chemicals that can hamper fetal development and may have long-term negative health outcomes in humans.[26] Consistently, Minatoya *et al* and Liang *et al* found that BPA exposure was related to child neurodevelopment disorders(eg,

attention-deficit/hyperactivity disorder symptoms), oxidative stress and immune disturbance among women with unexplained recurrent spontaneous abortion.[27 28] Overall, this suggests that more concern should be given to BPA or other BPA analogues due to their potential reproductive and developmental toxicity.

This proposed meta-analysis will probably be the first to explore the true relationship between BPA exposure and IVF outcomes among infertile adults who underwent IVF cycles. Many studies have investigated the correlation between BPA exposure and IVF outcomes in women or men, including embryo quality, fertilisation rate and clinical pregnancy, but no consistent conclusion has been reached.[29–35] We anticipate that we will have sufficient studies to pool together to conduct a meta-analysis for some of the primary and secondary outcomes. Although these studies were conducted in infertile adults who underwent IVF, BPA concentrations among these participants may be modest and may not have reached a biologically significant level. We will be unable to perform a dose-response analysis of the relationship between BPA exposure and IVF outcomes due to the relatively low BPA exposure levels and limited published studies, so we will only evaluate the subgroup analysis based on the tertile distribution of BPA median concentrations reported in these studies. The GRADE tool will be used as a reference for the construction of the summary table of the included studies.[19] In addition, we may be able to generate evidence on the risk of BPA exposure on IVF outcomes, which may help to provide some directions and hypotheses for further research. In view of the potential risk of BPA exposure and the increase in the number of IVF cycles, clarifying the impact of BPA exposure on IVF will help to improve IVF outcomes and provide better guidance for patients who undergo IVF procedure.

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