January 2017

New Insights into Signal Detection of the Effects of Exposures during Pregnancy

Fatma Etwel
The University of Western Ontario

Supervisor
Dr. Michael J Rieder
The University of Western Ontario

Joint Supervisor
Dr. Gideon Koren
The University of Western Ontario

Graduate Program in Physiology and Pharmacology

A thesis submitted in partial fulfillment of the requirements for the degree in Doctor of Philosophy

Abstract

There is inadequate information on the fetal safety of drugs during pregnancy for the majority of marketed drugs. It is challenging to examine the safety and efficacy of drugs during pregnancy due to the ethical issues of exposing unborn babies to these chemicals. It often takes many years before associations between a drug and its safety, efficacy, and toxicity in pregnancy can be established. This thesis will examine strategies in signal detection of the effects of drug exposures during pregnancy.

Meta-analyses have become useful in the area of clinical teratology. Observational studies provide the main source of information in these meta-analyses. Although the quality of meta-analysis of small observational studies is challenging, it is an effective strategy, as shown in the present study, in predicting correct signals to estimate teratogenicity years before large cohort studies become available.

Results of retrospective pregnancy registries are commonly reported in regulatory documentations. However, little data are available on the precision of the estimates from such registries. The present study confirms a consistent bias against the null hypothesis in a retrospective registry which needs to be considered when interpreting such data as a strategy in generating safety/risk signals of new drugs.

H1 antihistamines are used for the treatment of nausea and vomiting during pregnancy as well as the symptomatic relief of allergy. Although they are felt to be safe, several studies have challenged this assumption. By using

meta-analysis, the safety of antihistamines has been confirmed in this thesis with over 1.3 million exposed and control subjects.

Typically, after experimental animal studies, novel therapeutic modalities are tested by randomized controlled trials. Cumulative meta-analysis is an effective strategy to detect a possible time- dependent effect and potential bias against the null hypothesis, whether antioxidant treatment decreases the rates of preeclampsia. I have shown that the initial favorable effect seen in the first studies is nullified as the sample sizes and number of studies is increased.

There is a need to continue using and developing the above strategies to study the safety and efficacy of drugs to improve maternal fetal health.

Keywords

Meta-analysis, Pregnancy outcomes, Retrospective pregnancy registries, H1 antihistamine, Cumulative meta-analysis.

Co-Authorship Statement

Chapter 3: The role of meta-analyses in identifying human teratogenicity.

Dr. Koren provided guidance during the study design and critically revised the manuscript.

Chapter 4: Bias against the null hypothesis in retrospective registries of gestational drug exposure.

Dr. Koren provided guidance during the study design, analysis of data and preparation of the manuscript.

Chapter 5: The fetal safety of cetirizine: An observational cohort study and meta-analysis.

Dr. Djokanovic N, Dr. Moretti ME, Dr. Boskovic R and Dr. Martinovic identified and recruited patients and performed telephone interviews. Dr.Koren provided guidance during study design, analysis and interpretation of data and preparation of the manuscript.

Chapter 6: The risk of adverse pregnancy outcome after first trimester exposure to H1 antihistamines: a systematic review and meta-analysis.

Dr. Lauren H Faught performed study selection and preparation of the manuscript and Dr. Michael J Rieder and Dr. Gideon Koren provided guidance during the systematic review design and critically revised the manuscript.

Chapter 7: When original positive studies of novel therapies are subsequently nullified: Cumulative meta-analyses in preeclampsia.

Dr. Koren provided guidance during the study design, analysis and critically revised the manuscript.

Dedication

This thesis is dedicated to my parents, Professor Abdurahman Tawil and Hamida Elbanani, for their unconditional love and endless support.

Acknowledgment

I wish to express my thankfulness to my supervisors Dr. Gideon Koren and Dr. Michael J Rieder for their help, guidance and generosity of time. I also wish to thank my advisory committee members Dr. Doreen Matsui, Dr. Bard Urquhart and Dr. Rommel Tirona. I would like to thank Dr. Lauren H Faught for her help and support.

I am grateful to the Libyan Ministry of Higher Education for its financial sponsorship for my postgraduate education in Canada. Moreover, I am deeply grateful to the department of Physiology and Pharmacology at Western University for giving me the opportunity to enhance my education.

My deepest gratitude goes to my husband, Salaheddin Dred for his encouragement and understanding and my children, Jumana, Fathi and Faiz for accepting the little time and attention I gave them during my study. Without their support this study would not have been completed.

Table of Contents

List of Tables

List of Figures

List of Appendices

Appendix

List of Abbreviations

NVP	Nausea and Vomiting during Pregnancy
AMSTAR	A Measurement Tool for Assessment of Multiple Systematic Reviews
MINORS	Methodological Index for Non-Randomized Studies
CI	Confidence Interval
MA	Meta-Analysis
MM	Major Malformations
OR	Odds Ratio
RR	Relative Risk
ACEI	Angiotensin Converting Enzyme Ihibitors
df	degree of freedom
AHs	Antihistamines
H1AHs	H1 Antihistamines
H2AHs	H2 Antihistamines
RCTs	Randomized Controlled Tials
IUGR	Intrauterine Growth Restriction
No.	Number
CVS	Congenital Varicella Syndrome

Chapter 1: General introduction.

Presently, it often takes many years before any association is established between a drug and its toxicity, safety, and/or efficacy in pregnancy are established. With 50% of all pregnancies being unplanned [1], large numbers of women are exposed inadvertently to medications in early pregnancy. In addition, many pregnant women suffer from conditions that require continued treatment during pregnancy [2]; therefore, there is a need to be able to identify fetal risks as soon as possible. This 'integrated articles' thesis is based on five articles I have published over the last four years, all focusing on signal detection of the effects of exposures during pregnancy.

1.1. Understanding the role of meta-analysis in maternal-fetal health research.

Usually, randomized controlled drug studies cannot include pregnant women due to ethical issues surrounding the fear of potential teratogenic risk given the unknown safety profile of the new drugs. Therefore, prenatal adverse drug effect data are rarely known from such sources. Some pregnant women may be exposed to a new drug in the premarketing phase unintentionally; hence data are often collected in retrospective or prospective observations. Thus, post marketing epidemiological studies based on observational data are the main methods by which to study potential teratogenic effects when randomization is impossible [3].

However, over the past two decades, systematic reviews and meta-analyses of observational studies have been increasingly utilized in the field of clinical teratology as part of the general trend of a 500-fold increase in publications [4]. A systematic review is defined as "the application of scientific strategies that limit bias by a systematic assembly, critical appraisal, and synthesis of all relevant studies on a specific topic" [5]. In clinical teratology, the specific topic is typically the risks of major congenital malformations, miscarriage, or prematurity in the

exposed versus non-exposed groups, or the long term neurodevelopmental effects of in-utero exposure [6]. Meta-analysis is defined as the statistical synthesis of results from several independent, but 'combinable' studies, leading to a quantitative summary of the pooled results [5].

In the hierarchy of studies, meta-analyses and systematic reviews of randomized studies are ranked at the highest level of evidence. In contrast, systematic reviews and meta-analyses of observational studies possess more weaknesses and potential biases. Reporting of observational research is often not detailed and clear enough to assess the strengths and weaknesses of the investigations [7]. Furthermore, multiple deficiencies, such as heterogeneity [8], methodological quality [9], deficiencies in statistical methods [10], and controlling for all potential sources of bias [11], have been widely reported. As a result, the quality of published meta-analyses of observational studies ranges widely and the conclusions reached from the same set of studies may be conflicting.

Typically, after a new drug is introduced into the market, case reports, case series, and small, underpowered controlled cohort studies begin to emerge. Synthesizing these small studies in meta-analyses allows for an increase in sample size and, hence in the power to distinguish relatively small teratological signals. Only much later, when the drug has been used on a large scale, are large cohort studies published, and they are sufficiently powered to draw more solid conclusions on fetal safety/ risks.

There is a need to study and examine the validity of the conclusions reached by meta-analyses of small observational teratology studies as compared with the newer, very large studies. As meta-analyses of small observational studies are available years before large, appropriately powered cohort studies become

available, they may be important to consider in clinical counseling. It is therefore logical to try to estimate how early a valid signal (positive or negative) can be generated from meta-analyses before the large cohort studies are published (Chapter 3).

1.2. Understanding the role of retrospective pregnancy exposure registries in maternal-fetal health research.

Another important strategy to assess drug safety during pregnancy, employed mostly by drug companies, is using pregnancy exposure registries. Prospective pregnancy exposure registries, where pregnant women are enrolled before the outcomes of their pregnancies are known, are recognized as an important method for ascertaining risks associated with a drug exposure during pregnancy. In contrast, retrospective pregnancy exposure registries are based on women/physicians contacting the registry after pregnancy outcomes are known. Retrospective registries have been regarded as the weakest type of teratogenic evidence and were never viewed by clinicians and scientists as data that can lead to quantitative estimates of risk/safety. However, is this viewpoint justified?

There is a reporting bias that makes interpretation of retrospective pregnancy exposure registries challenging [12]. Until now, this type of bias has led clinicians, scientists and regulatory bodies to discard such data. But if the bias leads to a stable increase in signal, it may also mean that, if the retrospective study does not show malformation rates above the expected baseline of 3-5%, the drug is probably not associated with an increased teratogenic risk. Since most medicinal drugs are not teratogenic [13], the ability to use such data may empower women and their health professionals to use much needed drugs years before large prospective studies are conducted.

There is a need to compare the rates of reported malformations in retrospective pregnancy exposure registries vs. prospective pregnancy exposure of the same kind of drug at the same period of time via the same drug company in an effort to make the retrospective registries more useable and more a part of the overall analysis of teratogenicity (Chapter 4).

1.3. Applying meta-analysis to address unresolved research question in teratology with H1 antihistamine as an example.

H1 antihistamine is classified as either the first or old generation H1 antihistamine such as hydroxyzine or as the second or new generation H1 antihistamine such as cetirizine. The first generation class of H1 antihistamine has the ability to cross the blood brain barrier which can be considered a disadvantage when we want to treat allergy because it will cause a sedative side effect. However crossing the blood brain barrier can be an advantage when we want to treat nausea and vomiting during pregnancy (NVP). The second generation class of H1 antihistamine has less ability to cross the blood brain barrier which gives this class a big advantage when we want to treat allergy or asthma without the sedative side effect [14].

The best approach to quantify the safety/risk of H1 antihistamine during pregnancy is to conduct a meta-analysis of all available observational control studies and this is what Seto and his colleagues did twenty years ago and they concluded that H1 antihistamine can be safely used during pregnancy [15]. Another research group re-analyzed the Seto meta-analysis without adding new research and they found contradictory results [16]. Therefore, there is a need to conduct more observational studies on H1 antihistamines and also conduct a new meta-analysis on all available studies on H1 antihistamines to assess the risk of malformation and other pregnancy outcomes. We focused on assessing the safety

of using cetirizine and its prodrug hydroxyzine during the first trimester by conducting meta-analyses of the current and all available cohort studies that studied adverse pregnancy outcomes.

The current meta-analyses focus primarily on first trimester exposure. This was accomplished by including studies where the first trimester exposure to H1 antihistamines was necessary. Studies where mothers were also exposed in subsequent trimesters were included but this was not a requirement. The reasons for focusing on the first trimester exposure are twofold. First of all, the first trimester is a critical time for fetal development and thus an extremely important period for assessing the potential adverse effects of drugs. Gross structural abnormalities, which can be readily apparent in newborns, occur following exposure to teratogenic agents during this stage of organogenesis, when tissues and organs are developing [17]. As a result, most studies assessing the effects of drug exposure during pregnancy typically look at the first trimester exposure or view this timing of exposure as important.

Presently there is sufficient research to power a safety analysis given the large number of studies focusing on first trimester exposure. This can be partly caused by the commonplace use of H1 antihistamines during the first trimester. A woman may be given H1 antihistamines intentionally to treat NVP or another condition that presents during the first trimester. Alternatively, with 50% of pregnancies being unplanned [1], a woman may take prescribed or over the counter H1 antihistamines for treatment of a medical condition while being unaware of her pregnancy. Therefore, it is necessary to adequately and accurately assess the risks associated with H1 antihistamine use through meta-analysis (Chapter 5 and Chapter 6).

1.4. Cumulative meta-analysis as a tool in detecting the emergence of evidence in teratology, treatment of preeclampsia as an example.

Meta-analysis in general is not a statistical method that simply combines results from different studies and aggregates them as a large study. In meta-analysis, studies that include larger sample sizes and a lesser degree of variabilities are weighted more than other smaller studies. In meta-analysis there are two models, fixed effect and random effect. Fixed effect model is usually used when the studies come from the same research group and are highly homogenous. On the other hand, the random effect model is used when the studies come from different research groups. The big difference between the two models is the weighting of the studies. Studies with a smaller sample size have more weight (credit) in the random effect model estimate than on the fixed effect model estimate [18].

The forest plot is the results figure of a typical meta-analysis. This figure shows the effect measure of each study (like risk ratio or odds ratio) as well as its confidence interval and the weighted level. Also, the forest plot shows the summary of the overall effect measure and its confidence interval.

In case of cumulative meta-analysis, the result of the studies are accumulated from the earliest to the latest, in a way that each new study includes a synthesis of all prior studies. This sequential combining of the studies' results has an advantage of viewing whether there is consistency in the results of consecutive studies or if there is change in the direction of the overall estimate when more recent studies are added [18].

Taking the advantage of cumulative meta-analysis, the researcher can use it as a tool in detecting the emergence of evidence especially after a new remedy for pregnancy complications enters the market. Conducting cumulative meta-analysis

to examine changes over time in the pooled effect size of randomized control trials published on the protective effects of antioxidants and low dose aspirin against preeclampsia is used in this thesis to predict the potential bias against the null hypothesis (Chapter 7).

1.5. References.

1. Finer LB, Zolna MR. Unintended pregnancy in the United States: incidence and disparities, 2006. Contraception. 2011 Nov;84(5):478-85.

2. Bakker MK, Jentink J, Vroom F, Van Den Berg PB, De Walle HE, De Jong-Van Den Berg LT. Drug prescription patterns before, during and after pregnancy for chronic, occasional and pregnancy-related drugs in the Netherlands. BJOG. 2006 May;113(5):559-68.

3. Schaefer C, Ornoy A, Clementi M, Meister R, Weber-Schoendorfer C. Using observational cohort data for studying drug effects on pregnancy outcome--methodological considerations. Reprod Toxicol. 2008 Sep; 26(1):36-41.

4. Cook DJ, Mulrow CD, Haynes RB. Systematic reviews: synthesis of best evidence for clinical decisions. Ann Intern Med. 1997 Mar 1; 126(5):376-80.

5. Crowther MA, Cook DJ. Trials and tribulations of systematic reviews and meta-analyses. Hematology Am Soc Hematol Educ Program. 2007:493-7.

6. Ornoy A, Arnon J. Clinical teratology. West J Med. 1993 Sep;159(3):382-90.

7. Dixon E, Hameed M, Sutherland F, Cook DJ, Doig C. Evaluating meta-analyses in the general surgical literature: A critical appraisal. Ann Surg 2005; 241:450-459.

8. Maguire MJ, Hemming K, Hutton JL, Marson AG. Overwhelming heterogeneity in systematic reviews of observational anti-epileptic studies. Epilepsy Res 2008; 80:201-212.

9. Simunovic N, Sprague S, Bhandari M. Methodological issues in systematic reviews and meta-analyses of observational studies in orthopaedic research. J Bone Joint Surg Am 2009; 91:87-94.

10. Morshed S, Tornetta P 3rd, Bhandari M. Analysis of observational studies: A guide to understanding statistical methods. J Bone Joint Surg Am 2009; 91:50-60.

11. Wang PS, Schoenbaum M. Invited Commentary: Assessing treatment effects by using observational analyses - opportunities and limitations. Am J Epide- miol 2009; 170:286-287.

12. Bar-Oz B, Moretti ME, Mareels G, Van Tittelboom T, Koren G. Reporting bias in retrospective ascertainment of drug-induced embryopathy. Lancet. 1999; 354:1700-1.

13. Koren G, Pastuszak A, Ito S. Drugs in pregnancy.N Engl J Med 1998; 338:1128-37.

14. Briggs GG, Freeman RK, Yaffe SJ. Drugs in pregnancy and lactation. 7th ed. Philadelphia: Lippincott Williams & Wilkins; 2002.

15. Seto A, Einarson T, Koren G. Pregnancy outcome following first trimester exposure to antihistamines: meta-analysis. Am J Perinatol. 1997;14(3):119-24.

16. Chin JW, Gregor S, Persaud N. Re-analysis of safety data supporting doxylamine use for nausea and vomiting of pregnancy. Am J Perinatol. 2014 Sep;31(8):701-10.

17. Ruedy J. Teratogenic Risk of Drugs Used in Early Pregnancy. Canadian Family Physician. 1984; 30:2133-2136.

18. Borenstein M, Hedges LV, Higgins JPT, Rothstein HR. Introduction to Meta-Analysis. John Wiley & Sons, Ltd, Chichester, UK 2009.

Chapter 2: Objectives and hypotheses.

2.1. Objectives.

1) To systematically review the literature to examine the validity of the conclusions reached by meta-analyses of small observational studies by comparing the results reached in these meta-analyses with more recent, large, methodologically superior studies published on the same topic at a later date.

2) To compare the rates of major congenital malformations reported in retrospective versus prospective registries of the same drug to quantify the potential bias of retrospective reports.

3) To systematically review the literature to conduct meta-analyses to measure the rate of adverse pregnancy outcomes of cetirizine and all H1-antihistamines after the first trimester exposure.

4) To conduct cumulative meta-analyses to examine changes over time in the pooled effect size of randomized control trials published on the protective effects of antioxidants and low dose aspirin against preeclampsia, and to identify determinants that may affect such changes.

2.2. Hypotheses.

1) The direction of the conclusions reached by meta-analyses of small studies on several teratology topics are similar to the results reached by more recent, large, methodologically superior studies published on the same topic at a later date.

2) There is a systematic bias of retrospective registries when the rates of major congenital malformations reported in retrospective registries are much higher than prospective registries of the same drug, which also suggests that if the

retrospective registries did not show increase in major congenital malformations above the baseline risk, the drug is probably not a major teratogen.

3) The use of cetirizine and all H1-antihistamines during the first trimester is not associated with an increased risk of adverse pregnancy outcomes.

4) Cumulative meta-analysis is an effective tool in predicting potential bias against the null hypothesis of randomized control trials published on the protective effects of antioxidants and low dose aspirin against preeclampsia.

Chapter 3: The role of meta-analyses in identifying human teratogenicity.

This chapter has been published previously as part of a book chapter.

Etwel F, Hutson JR, Madadi P, Gareri J, Koren G. Fetal and perinatal exposure to drugs and chemicals: novel biomarkers of risk. Annu Rev Pharmacol Toxicol. 2014; 54:295-315.

3.1. Introduction.

Typically, randomized controlled drug studies exclude pregnant women owing to fear of teratogenic risk. As a result, prenatal adverse drug effects data are rarely available from randomized trials, and epidemiological studies based on observational data constitute the main data source [1].

Reporting of observational research is often not sufficiently detailed and clear to assess the potential strengths and weaknesses of these investigations. Multiple deficiencies such as heterogeneity, methodological quality, insufficient statistical methods, and control of potential sources of bias have been widely recognized [2, 3].

Typically, after many years during which small cohort studies report on fetal safety/risk of a particular drug, administrative databases may report on large numbers of patients exposed to that agent. Although the strength of these studies lies in their large size, these large cohort studies take many years to conduct and are published long after the small cohort studies are published. The synthesis of these small cohort studies into a systematic review and meta-analysis may yield an important early signal for the safety/risk of drug use in pregnancy.

Over the past two decades, such systematic reviews and meta-analyses of observational studies have been increasingly published in the field of clinical teratology [4]. To date, the conclusions reached by meta-analyses of small observational teratological studies have not been validated through comparison with those reached by more recent, very large cohort studies.

3.2. Methods.

As the first step in validating such conclusions, we identified all meta-analyses of small observational teratological studies published in peer review

journals. Meta-analyses were eligible for consideration if the outcome measures were the risk of congenital malformation and/or the risk of long-term neurodevelopment of children after in utero exposure to therapeutic drugs in the first trimester of pregnancy.

As the second step, we identified subsequently published large teratological cohort studies on the same drug addressing the same endpoint. Large cohort studies were judged suitable for inclusion in this review if the number of women exposed to the drug in question exceeded either 1,000 or the combined number of total cases in the corresponding meta-analysis.

3.3. Results.

Of more than 60 meta-analyses on medicinal drugs published by December 31, 2012, 9 meta-analyses could be matched to large, later cohort studies on the same drug (Table 3). There were 7 "negative" meta-analyses (i.e., showing no teratological effects) and 2 "positive" ones (showing either morphological or developmental adverse effects). In all 9 instances, the meta-analyses accurately predicted the results of the later, large cohort studies. The AMSTAR scores were in the medium range in all 9 meta-analyses, and the MINORS quality score of the large cohort studies was 17.9 ± 1.4 (mean ± standard deviation), which is considered to be on the border of "good" quality (Appendix 2: Detailed characteristics of the included studies).

Table 3. Characteristics of the nine meta-analyses included in the comparison with large cohort studies

Type of exposure and outcome	MA reference	Association measure (95% CI)	Large study reference	Association measure (95% CI)
Benzodiazepines and MM	[7]	OR = 0.90 (0.61-1.35)	[8]	OR = 1.12 (0.91-1.36)
Untreated epilepsy and MM	[9]	OR = 1.92 (0.92-4.00)	[10]	OR = 1.00 (0.8-1.4)
Proton-pump inhibitors and MM	[11]	RR = 1.18 (0.72-1.94)	[12]	OR = 1.10 (0.91-1.34)
H2 blockers and MM	[13]	OR = 0.99 (0.60-1.65)	[14]	OR = 1.14 (0.89-1.45)
ACE inhibitors and MM	[15]	OR = 1.41 (0.66-3.04)	[16]	OR = 1.12 (0.83-1.49)
Valproic acid and MM	[17]	RR = 3.77 (2.18 - 6.52)	[18]	(Spina bifida) RR = 12.7 (7.7-20.7)
Valproic acid and reduction in IQ	[19]	P = 0.001	[20]	P = 0.009
Carbamazepine and reduction in IQ	[19]	P = 0.39	[20]	P = 0.20
Varicella infection and MM	[21]	Risk = 2.2 % (0-4.6%)	[22]	Risk = 0.4% (0.05-1.5%)

CI: Confidence Interval, MA: Meta-Analysis, MM: Major Malformations, OR: Odds Ratio, RR: Relative Risk.

3.4. Discussion.

It is encouraging that meta-analyses of earlier, albeit smaller, cohort studies tend to generate an accurate overall teratogenic signal in estimating human teratogenicity years before large and methodologically superior cohort studies are published. The meta-analyses offer clinicians, scientists, and regulators an earlier signal for the presence or lack of teratogenic risk and hence can have an important impact on clinical practice.

3.5. Conclusion.

Meta-analyses of small cohort studies of pregnancy outcome appropriately predict results of large cohort studies.

3.6. References.

1. Schaefer C, Ornoy A, Clementi M, Meister R, Weber-Schoendorfer C. Using observational cohort data for studying drug effects on pregnancy outcome—methodological considerations. Reprod. Toxicol. 2008; 26:36–41.

2. Maguire MJ, Hemming K, Hutton JL, Marson AG. Overwhelming heterogeneity in systematic reviews of observational anti-epileptic studies. Epilepsy Res. 2008; 80:201–12.

3. Simunovic N, Sprague S, Bhandari M. Methodological issues in systematic reviews and meta-analyses of observational studies in orthopaedic research. J. Bone Jt. Surg. Am. 2009; 91(Suppl. 3):87–94.

4. Koren G. Medication safety in pregnancy and breastfeeding. In Motherisk Archives of Systematic and Evaluative Reviews, ed. G Koren. 2007; pp. 313–603.

5. Sequeira-Byron P, Fedorowicz Z, Jagannath VA, Sharif MO. An AMSTAR assessment of the methodological quality of systematic reviews of oral healthcare interventions published in the Journal of Applied Oral Science (JAOS). J. Appl. Oral Sci. 2011; 19:440–47.

6. Slim K, Nini E, Forestier D, Kwiatkowski F, Panis Y, Chipponi J. Methodological index for non-randomized studies (minors): development and validation of a new instrument. ANZ J. Surg. 2003; 73:712–16.

7. Dolovich LR, Addis A, Vaillancourt JM, Power JD, Koren G, Einarson TR. Benzodiazepine use in pregnancy and major malformations or oral cleft: meta-analysis of cohort and case-control studies. BMJ. 1998; 317:839–43.

8. Wikner BN, Stiller CO, Bergman U, Asker C, Källén B. Use of benzodiazepines and benzodiazepine receptor agonists during pregnancy: neonatal outcome and congenital malformations. Pharmacoepidemiol. Drug Saf. 2007; 16:1203–10.

9. Fried S, Kozer E, Nulman I, Einarson TR, Koren G. Malformation rates in children of women with untreated epilepsy: a meta-analysis. Drug Saf. 2004; 27:197–202.

10. Veiby G, Daltveit AK, Engelsen BA, Gilhus NE. Pregnancy, delivery, and outcome for the child in maternal epilepsy. Epilepsia. 2009; 50:2130–39.

11. Nikfar S, Abdollahi M, Moretti ME, Magee LA, Koren G. Use of proton pump inhibitors during pregnancy and rates of major malformations: a meta-analysis. Dig. Dis. Sci. 2002; 47:1526–29.

12. Pasternak B, Hviid A. Use of proton-pump inhibitors in early pregnancy and the risk of birth defects. N. Engl. J. Med. 2010; 363:2114–23.

13. Gill SK, O'Brien L, Koren G. The safety of histamine 2 (H2) blockers in pregnancy: a meta-analysis. Dig. Dis. Sci. 2009; 54:1835–38.

14. Matok I, Gorodischer R, Koren G, Sheiner E, Wiznitzer A, et al. The safety of H2-blockers use during pregnancy. J. Clin. Pharmacol. 2010; 50:81–87.

15. Walfisch A, Al-maawali A, Moretti ME, Nickel C, Koren G. Teratogenicity of angiotensin converting enzyme inhibitors or receptor blockers. J. Obstet. Gynaecol. 2011; 31:465–72.

16. Li DK, Yang C, Andrade S, Tavares V, Ferber JR. Maternal exposure to angiotensin converting enzyme inhibitors in the first trimester and risk of malformations in offspring: a retrospective cohort study. BMJ. 2011; 343:d5931.

17. Koren G, Nava-Ocampo AA, Moretti ME, Sussman R, Nulman I. Major malformations with valproic acid. Can. Fam. Physician. 2006; 52:441–42.

18. Jentink J, Loane MA, Dolk H, Barisic I, Garne E, et al. Valproic acid monotherapy in pregnancy and major congenital malformations. N. Engl. J. Med. 2010; 362:2185–93.

19. Banach R, Boskovic R, Einarson T, Koren G. Long-term developmental outcome of children of women with epilepsy, unexposed or exposed prenatally to antiepileptic drugs: a meta-analysis of cohort studies. Drug Saf. 2010; 33:73–79.

20. Meador KJ, Baker GA, Browning N, Clayton-Smith J, Combs-Cantrell DT, et al. Cognitive function at 3 years of age after fetal exposure to antiepileptic drugs. N. Engl. J. Med. 2009; 360:1597–605.

21. Pastuszak AL, Levy M, Schick B, Zuber C, Feldkamp M, et al. Outcome after maternal varicella infection in the first 20 weeks of pregnancy. N. Engl. J. Med. 1994; 330:901–5.

22. Enders G, Miller E, Cradock-Watson J, Bolley I, Ridehalgh M. Consequences of varicella and herpes zoster in pregnancy: prospective study of 1739 cases. Lancet. 1994; 343:1548–51.

Chapter 4: Bias against the null hypothesis in retrospective registries of gestational drug exposure.

This chapter has been accepted for publication (in press):

Etwel F, Koren G. Bias against the Null Hypothesis in Retrospective Registries of Gestational Drug Exposure. J Obstet Gynaecol Can. 2016.

4.1. Introduction.

Typically, medications for use in humans are introduced to the market supported by reproductive animal data, which are often not predictive of the risk of human malformation. Furthermore, in pre-marketing clinical trials, accidental exposures to a medication during pregnancy are typically very rare [1]. However, because 50% of all pregnancies are unplanned [2], large numbers of women are exposed inadvertently to medications in early pregnancy. Moreover, many pregnant women suffer from conditions that require continued treatment during pregnancy. When a new drug enters the market, case reports of fetal exposure begin to emerge, but unless a very highly teratogenic signal and a unique phenotype are evident (such as was noted with thalidomide or isotretinoin) [3], it takes years before a prospective cohort study of first trimester fetal exposure becomes available.

Historically, most information about the risks of drugs in pregnancy has arisen from findings of spontaneous adverse event reports (case reports). This mechanism of passive surveillance has been well-described [4] and is advantageous in the identification of a rare or unusual fetal outcome. A major limitation of retrospective case series is the lack of denominator data, which precludes estimation of the size of risk with use of the drug compared to the risk in the general population.

Retrospective registries of exposure during pregnancy (enrolment in which follows notification by families or physicians after the pregnancy outcome is known) are typically established by drug companies as part of the regulatory process and their contents are often reported in the peer-reviewed literature.

The main concern regarding the interpretation of findings in these registries is that families with malformed children exposed to a given drug in pregnancy, or their physicians, will be more likely to report the malformation to registries than families with healthy children prenatally exposed to the same drug [5]. However, there is little information available on the precision of the estimates from such registries. In 1999, our group documented that the rate of major malformation associated with the antifungal itraconazole was 13% in the retrospective report collected by the manufacturer, but was only 3.2% in the prospective report collected by the same company [5]. Since then, however, the hypothesis that retrospective registries are biased towards higher rates of malformations has not been further confirmed.

Because most medications are not teratogenic [6], a potentially false teratogenic signal may elicit anxiety and may lead women not to treat serious medical conditions. In at least one class of drugs (the statins), a report of adverse fetal outcomes based on retrospective surveillance [7] led to high levels of anxiety. However, the adverse fetal outcomes were later shown in a meta-analysis of prospective studies not to be associated with exposure to statins [8].

The objective of the present study was to compare the rates of major congenital malformations reported in retrospective and prospective registries for the same drug to quantify the potential bias of retrospective reports.

4.2. Methods.

We performed a search of the electronic database PubMed from inception to December 31 2013 for all available full English texts, using the following search terms: “retrospective pregnancy registry”, “prospective pregnancy registry”, “reporting bias”, “drug company”, “drug registry”, and combining them with

"congenital malformations" or "embryopathy". In addition, several pregnancy registry annual reports that were documented by drug companies and received by the Motherisk program at the Hospital for Sick Children in Toronto were reviewed for the period 1984 to 2011. Motherisk regularly receives these reports upon their release.

For this analysis, we included published articles and registry reports that provided data on rates of major malformations in the offspring of women who were exposed to the specific drug during the first trimester of pregnancy, derived from both retrospective and prospective registries for the same drug.

The following information was recorded from the registries for each drug: the total number of major malformations among live born infants; the number of stillbirths or terminated pregnancies (the numerator); and the total number of reported live births, stillbirths, elective pregnancy terminations, and miscarriages (the denominator).

The reported rates of major malformations in the retrospective and prospective reports from the same registry for the same drug were compared using Fisher's exact test. Odds ratios and 95% confidence intervals were also calculated. The distribution of malformations in each report was compared to the normal distribution of birth defects reported in the United States, in order to identify whether there was a specific pattern of malformations [9].

4.3. Results.

The electronic search identified a total of 1316 published articles. After removing all animal studies, case reports, controlled observational studies, and review articles without original data, 122 articles were reviewed in detail. Five drugs or classes of drug identified in peer-reviewed published articles fulfilled the

inclusion criteria (itraconazole, fluoxetine, acyclovir, statins, and mefloquine) [5, 10-13]. Three drugs from drug company annual reports also met the inclusion criteria (quetiapine, quadrivalent human papillomavirus vaccine, and montelukast sodium). In all cases, the rates of major malformations after exposure to these drugs were significantly higher in data reported retrospectively than in data reported prospectively (Table 4). For all drugs studied, estimates of major congenital malformation rates from retrospective registries were higher than from prospective registries; the median bias was higher by a factor of 4.18 ± 1.23 (range 2.13 to 5.97). For six of these drugs the breakdown of malformations was available in the reports, and there was no specific pattern of malformations in any of them (data not shown). Details of each of the eight drug registries can be found in the Appendix 3.

Table 4. Rates of major malformation in the eligible registries.

Drug name	Retrospective data		Prospective data		Increased rate*	*P* for difference	OR (95% CI)
	No. of cases with malformation / total No. of cases	Rate (%) (95% CI)	No. of cases with malformation/ total No. of cases	Rate (%) (95% CI)			
Quetiapine	20/253	7.91 (5.2 to 11.9)	6/224	2.68 (1.2 to 5.8)	2.95	0.014	3.12 (1.23 to 7.91)
Quadrivalent human papillomavirus vaccine	12/261	4.60 (2.6 to 7.9)	24/1113	2.16 (1.4 to 3.2)	2.13	0.049	2.19 (1.08 to 4.43)
Montelukast Na	11/66	16.67 (9.5 to 27.6)	8/250	3.20 (1.6 to 6.3)	5.21	<0.001	6.05 (2.32 to 15.75)
Itraconazole	17/166	10.24 (6.5 to 15.9)	5/199	2.51 (1.0 to 5.9)	4.08	0.003	4.43 (1.60 to 12.27)
Fluoxetine	89/426	20.89 (17.3 to 25.0)	23/658	3.50 (2.3 to 5.2)	5.97	<0.001	7.29 (4.52 to 11.75)
Acyclovir	7/31	22.58 (11.2 to 40.4)	5/101	4.95 (2.1 to 11.3)	4.56	0.007	5.60 (1.63 to 19.19)
Statins	13/91	14.29 (8.5 to 23.1)	6/158	3.80 (1.7 to 8.2)	3.76	0.005	4.22 (1.55 to 11.54)
Mefloquine	29/115	25.22 (18.1 to 33.9)	38/717	5.30 (3.9 to 7.2)	4.76	<0.001	6.03 (3.54 to 10.27)

*Increased rate is equal to the retrospective risk percentage divided by prospective risk percentage

4.4. Discussion.

In all available registries with both prospective and retrospective collection of data for the same drug, we found consistently higher rates of congenital malformations in the reports based on retrospective data collection. The powerful impact of reporting the results of a retrospective data collection became evident when Edison and Muenke claimed (based on case reports) that statins increased teratogenic risk [7]; this claim supported a category X labelling ("the risks involved in use of the drug in pregnant women clearly outweigh potential benefits") for this class of drugs by the Food and Drug Administration in the United States. However, an increasing number of prospective controlled cohort studies and a meta-analysis have failed to show such an association [8]. Retrospective data collection appears therefore to result in reporting bias; this reporting bias can lead to high levels of anxiety and misperception among women and their health professionals, leading even to the termination of otherwise wanted pregnancies [6].

This significant reporting bias of registries with spontaneous retrospective data collection does not apply to controlled retrospective observational studies, such as case-control or retrospective cohort studies. Typically, in controlled retrospective studies the measurement of exposure to drugs is not correlated with the measurement of outcomes, contrary to drug companies' reports [14].

Because, in all cases, the bias against the null hypothesis in retrospective registries was evident and consistent, an additional hypothesis may be considered. In conditions in which the retrospective registry for a particular drug does not show malformation rates above the expected baseline of 3% to 5%, the drug is unlikely to be associated with a clinically significant increase in teratogenic risk.

To begin examining this hypothesis, we considered the effects of thiopurines, which are widely used in the management of inflammatory bowel disease and other autoimmune conditions. The first retrospective study of teratogenic effects associated with this drug class reported 31 exposed cases, with only one case with malformation, which was attributed to another drug [15]. Hence the malformation rate associated with thiopurines identified in this retrospective registry was in the range of the 3-5% baseline risk. Agreeing with this study, two separate meta-analyses of all prospective thiopurine studies suggested that maternal exposure to this class of medications was not associated with increased teratogenic risk [16, 17]. This hypothesis will have to be confirmed by additional research.

4.5. Conclusion.

Our findings confirm that when data related to malformations associated with drug use in pregnancy are collected in registries retrospectively, studies based on these data have a major and consistent bias against the null hypothesis. This bias must be considered when interpreting the findings of such studies.

4.6. References.

1. Shields KE, Wiholm BE, Hostelley LS, Striano LF, Arena SR, Sharrar RG. Monitoring outcomes of pregnancy following drug exposure: a company-based pregnancy registry program. Drug Saf 2004;27:353–67.

2. Finer LB, Zolna MR. Unintended pregnancy in the United States: incidence and disparities. Contraception 2006; 84:478–85.

3. McBride WG. Thalidomide and congenital abnormalities. Lancet 1961;2:1358.

4. Kennedy D, Goldman S, Lillie R. Spontaneous reporting in the United States. In Strom B, ed., Pharmacoepidemiology, 3rd ed, England: John Wiley & Sons; 2000: pp.151–74.

5. Bar-Oz B, Moretti ME, Mareels G, Van Tittelboom T, Koren G. Reporting bias in retrospective ascertainment of drug-induced embryopathy. Lancet 1999;354:1700–1.

6. Koren G, Pastuszak A, Ito S. Drugs in pregnancy. N Engl J Med 1998;338:1128–37.

7. Edison RJ, Muenke M. Central nervous system and limb anomalies in case reports of first-trimester statin exposure. N Engl J Med 2004;350:1579–82.

8. Zarek J, Koren G: The fetal safety of statins; a systematic review and meta analysis. J Obstet Gynaecol Can 2014;36:596–9.

9. James LM. Maps of birth defects occurrence in the U.S., Birth Defects Monitoring Program (BDMP)/CPHA, 1970-1987. Teratology 1993;48:551–646.

10. Goldstein DJ, Corbin LA, Sundell KL. Effects of first-trimester fluoxetine exposure on the newborn. Obstet Gynecol 1997;89:713–8.

11. Andrews EB, Yankaskas BC, Cordero JF, Schoeffler K, Hampp S. Acyclovir in pregnancy registry: six years; experience. The Acyclovir in Pregnancy Registry Advisory Committee. Obstet Gynecol 1992;79:7–13.

12. Pollack PS, Shields KE, Burnett DM, Osborne MJ, Cunningham ML, Stepanavage ME. Pregnancy outcomes after maternal exposure to simvastatin and lovastatin. Birth Defects Res A Clin Mol Teratol 2005;73:888–96.

13. Schlagenhauf P, Blumentals WA, Suter P, Regep L, Vital-Durand G, Schaerer MT, Boutros MS, et al. Pregnancy and fetal outcomes after exposure to mefloquine in the pre- and periconception period and during pregnancy. Clin Infect Dis 2012;54:e124-31.

14. Mann CJ. Observational research methods. Research design II: cohort, cross sectional, and case control studies. Emerg Med J 2003;20:54–60.

15. Schardein JL. Chemically induced birth defects. Marcel Dekker , New York, 2nd edition.1993.

16. Akbari M, Shah S, Velayos FS, Mahadevan U, Cheifetz AS. Systematic review and meta-analysis on the effects of thiopurines on birth outcomes from female and male patients with inflammatory bowel disease. Inflamm Bowel Dis 2013;19:15–22.

17. Hutson JR, Matlow JN, Moretti ME, Koren G. The fetal safety of thiopurines for the treatment of inflammatory bowel disease in pregnancy. J Obstet Gynaecol Can 2013;33:1–8.

Chapter 5: The fetal safety of cetirizine: An observational cohort study and meta-analysis.

This chapter has been published previously:

Etwel F, Djokanovic N, Moretti ME, Boskovic R, Martinovic J, Koren G. The fetal safety of cetirizine: an observational cohort study and meta-analysis. J Obstet Gynaecol. 2014 Jul;34(5):392-9.

5.1. Introduction.

Second-generation antihistamines (astemizole, loratadine, cetirizine, and fexofenadine) provide symptomatic relief of allergic disorders without the adverse effects of first-generation antihistamines, mostly CNS and anticholinergic effects. Second-generation antihistamines are preferred particularly by patients with a higher risk for development of adverse effects, including sedation and impairment sleep architecture [1, 2].

Cetirizine hydrochloride (Reactine)®, a nonprescription selective, second-generation histamine (H1) receptor antagonist, is a major active metabolite of hydroxyzine (first-generation antihistamines) with anti-allergic, antihistaminic and anti-inflammatory effects. Cetirizine is the most potent antihistamine available and more effective than loratadine and other H1 receptor antagonists in inhibiting histamine induced wheal response (swelling) and flare response (vasodilation) [3]. Due to its high potency, cetirizine may be appropriate for most severe allergy symptoms that are unresponsive to other antihistamines. It has a rapid onset, a long duration of activity and low potential for interaction with drugs that are metabolized by the hepatic cytochrome P450 system [3, 4].

It is estimated that up to 20%-30% of women of childbearing age experience allergic rhinitis and 4%-7% suffer from asthma during pregnancy, making them two of the most common groups of medical conditions that complicate pregnancy. The symptoms may vary from mild (sneezing, itching), which commonly can be relieved by avoiding the source of allergy, to severe nasal obstruction that may require pharmacotherapy [5]. Product labels state that medications for allergic rhinitis should be avoided during pregnancy due to lack of fetal safety data [6] and because half of all pregnancies are unplanned [7], this may lead to fetal exposure to antihistamine before a woman knows she is pregnant.

No teratogenic effects were detected with oral cetirizine doses of 60, 188 and 133 times the maximum clinically studied human dose in mice, rats and rabbits, respectively [8]. However, the animal studies are not necessarily indicative of adverse effects during pregnancy at clinically relevant doses and are not always predictive of human response. There are limited human studies reporting cetirizine exposure in pregnancy and pregnancy outcomes. A small prospective, comparative study conducted by Motherisk following 39 mothers exposed to cetirizine (37 in the first trimester) did not find differences in pregnancy outcomes between the exposed and comparison groups [9]. The most recent data were from the Berlin teratogen information service, with 196 women exposed in any trimester (11% in the first trimester), also not showing increased risk of birth defects or other adverse outcomes [10]. A recent review from the Food and Drug Administration with the American College of Allergy, Asthma, Immunology and American College of Obstetrics and Gynecology stated that cetirizine should be considered mainly for the second and third semester of pregnancy as a second generation agent for allergic condition in pregnancy if first generation antihistamine agents are not tolerable [11]. These statements suggest to patients and health care providers that the fetal safety of cetirizine is still questioned. Because half of all pregnancies are unplanned, this type of message may increase anxiety among many exposed women.

Hence, our study objectives were as follows: the primary objective was to determine whether cetirizine hydrochloride exposure during the first trimester of human pregnancy is associated with an increased rate of major birth defects above the baseline rate of 2-5% in the general population. The secondary objective was to determine the rates of spontaneous abortions, stillbirths, birth weight and neonatal complications following cetirizine hydrochloride exposure.

5.2. Methods.

This study includes an observational cohort study and a meta-analysis of all available studies to date.

5.2.1. Observational cohort study.

5.2.1.1. Study design.

This was a prospectively collected observational cohort study.

5.2.1.2. Settings.

This study was conducted at the Motherisk Program located at the Hospital for Sick Children in Toronto. The Motherisk Program is a counseling service that provides pregnant, breastfeeding women, and health professionals information on the safety and risks of exposures to prescription and over-the-counter medications, natural health products, chemicals, radiation, and infectious diseases [12]. Women who called the Motherisk Program between January 1, 2004 and December 31, 2007 were enrolled.

5.2.1.3. Study participants.

Three groups of women were recruited. The first group included pregnant women exposed to cetirizine hydrochloride during the first trimester of pregnancy. The second group included women who called the general Motherisk line about exposure to non-teratogenic agents (control group). The control group was matched to the study group (first group) according to maternal age at the time of conception (± 2 years) and gestational age at the time of first call to Motherisk (± 2 weeks). The third group included pregnant women exposed to cetirizine hydrochloride during the second and/or third trimester of pregnancy (disease matched non first trimester exposure).

5.2.1.4. Inclusion criteria.

1. Women who contacted Motherisk regarding information on the safety or risk of using cetirizine hydrochloride for the treatment of seasonal allergic rhinitis, chronic idiopathic urticaria and any other allergy at any stage of pregnancy.

2. Women who contacted Motherisk regarding information on the safety of vitamins or other non-teratogenic exposures (i.e. Tylenol, etc.) at any stage of their pregnancy.

5.2.1.5. Exclusion criteria.

1. Women who were exposed to teratogenic agents (e.g. anticonvulsants, isotretinoin, warfarin).

2. Women with medical conditions that may be associated with birth defects or any pregnancy complications (e.g. diabetes, alcohol abuse).

5.2.1.6. Data collection.

All women in the study group were recruited when they first contacted the Motherisk program regarding the safety of cetirizine hydrochloride during pregnancy. At the time of their first call, the study protocol was explained and oral informed consent was obtained. At this initial call, we collected demographic data, general health information, and information on exposure to any drugs used concomitantly with a special focus on the details about cetirizine exposure (dose, duration and adverse effects) on a previously developed structural questionnaire. The women were re-interviewed 6 months or more after delivery to obtain outcome data using standardized follow-up forms. In addition, the mothers' pre-pregnancy weight and weight at the time of delivery were recorded. Subsequent to the completion of the pregnancy follow-up and in order to confirm medical details of the babies' health, it was necessary to obtain the verbal permission of the

women to send a letter to their children's health care providers. The letters were sent to the caller's physicians (family physicians or pediatricians) for verification of the information obtained from the mothers. The doctors were asked to complete questionnaires and the state of general health of the babies as well as major and minor malformations. The follow-up procedures have been approved by Sick Children Hospital's Research Ethics Committee.

5.2.1.7. Statistical analysis.

Outcome end points of interest were compared among those exposed to cetirizine in the first trimester group and control groups (not exposed to cetirizine and non-first trimester exposed to cetirizine) with the Student's t test with Bonferroni correction, Chi-square or the Fisher's exact test whenever suitable.

5.2.2. Meta-analysis.

A systematic review and meta-analyses were conducted on all observational cohort studies published (including the current cohort study) that address the effect of cetirizine on pregnancy outcomes and cohort studies that had hydroxyzine as an exposed group. Cetirizine is an active carboxylic acid metabolite of hydroxyzine (first generation antihistamine) [13]. Combining the extracted cetirizine studies and hydroxyzine studies in one meta-analysis is therefore biologically plausible and will increase the sample size to estimate the fetal safety of cetirizine and hydroxyzine during pregnancy.

5.2.2.1. Search strategy.

A systematic review was performed to retrieve all published articles involving cetirizine or hydroxyzine exposure during pregnancy. This review followed the guidelines of the Meta-Analysis of Observational Studies in Epidemiology (MOOSE) group [14]. Searches were conducted using electronic

databases for possibly relevant articles that were published in any language up to December 2012. Included databases were PubMed, SCOPUS, EMBASE and TOXLINE. The literature was searched using drug names (Cetirizine, Cetirizine Dihydrochloride, Zyrtec, Reactine, Zirtek, Voltric, Cetirizine AL, Hydroxyzine, Vistaril, Durrax or Atarax) and drug categories based on pharmacological action (Histamine H1 Antagonists, H1 Antihistaminic, H1 Receptor Blockaders, Non-Sedating Histamine H1 Antagonists, Second Generation H1 Antagonists or first Generation H1 Antagonists).

Subsequently, these terms were combined with various MeSH categories (including pregnancy, pregnancy complications, abnormalities, embryonic and fetal development, maternal exposure, teratogens) and keywords (including birth defect, abnormality, malformation, embryopathy). The reference lists of all retrieved studies, including reviews, were examined for articles not identified by the search strategy.

5.2.2.2. Study selection.

Studies were included in the meta-analysis if they met the following criteria:

1- Observational cohort studies, but not case control, or case reports.

2- Studies that reported the incidence of malformation and/or other pregnancy outcomes in the offspring of women who were or were not exposed to cetirizine during pregnancy (use of a control or comparison group).

3- Sufficient data to calculate the Odd ratios.

4- Reported data that was not included in a later report by the same group of investigators (to prevent duplication of overlapping reports).

5- A sample size larger than 10.

6- Human studies only.

Two authors reviewed the studies to determine whether or not they met inclusion criteria.

5.2.2.3. Data extraction and analysis.

A data extraction form was used to collect the information from each study. This information included: first author, year of publication, study design, study location, years of study and outcome measures. Outcome measures were extracted for either the cetirizine or hydroxyzine exposed group and the control group and these measures included the whole group number, live birth number, malformed cases, spontaneous and therapeutic abortion cases, and prematurity (< 37 weeks gestation) cases. All the data was arranged in 2x2 tables to calculate the odd ratios. A random-effects meta-analysis model was used to combine the risk data for malformation and other pregnancy outcomes by using a statistical program called Comprehensive Meta-Analysis Version 2.0. Odd rations and 95% confidence intervals were calculated. Heterogeneity of effects was assessed using the Q statistic. Three meta-analyses were conducted to assess the risk of major malformations, one for cetirizine studies, one for hydroxyzine studies, and another one for combined hydroxyzine and cetirizine studies. Two meta-analyses were performed for the other pregnancy outcomes (spontaneous abortion and prematurity).

5.3. Results.

5.3.1. Cetirizine cohort study.

The cohort study included 78 pregnancies exposed to cetirizine during the first trimester (with or without second or third exposure), 56 pregnancies exposed to cetirizine during second and/or third trimester (no first trimester exposure), and

134 pregnancies exposed to non teratogenic drugs. Mean of maternal age of the three groups were similar (between 32 and 33 years).

In the group exposed to cetirizine during first trimester, there were a total of 73 live births counting three sets of twins, five spontaneous abortions, one therapeutic abortion, two fetal deaths, and eleven premature births. Two cases with major malformation were reported in the cetirizine first trimester exposed group: The first case was a child born with a hip dysplasia and the other case was Down's syndrome, detected at 14 weeks and ending in therapeutic abortion at 18-19 weeks. Two cases with minor malformation were reported in the same group: periventricular leukomalacia was diagnosed in the same child that was born with a hip dysplasia, and the second case was a child born with a tongue tie.

In the group exposed to cetirizine during only the second and/or third trimester, there were 57 live births including one set of twins (all the pregnancies ended with a live birth), and four premature cases. There were no major malformations and one minor malformation. The child with the minor malformation was born with esophageal sphincter, which was not fully formed until 4-5 months. This defect runs in the paternal family.

In our control group, there were 128 live births including one set of twins, seven spontaneous abortions, and three premature cases. In this control group, there were three major malformations and one minor malformation. One of the major malformations was Trisomy 13 found in one of the spontaneously aborted fetuses. The other two major malformations were infants born with undescended testes associated with exstrophy of the bladder and right inguinal hernia. The minor malformation case was an infant born with an umbilical hernia.

Two comparisons were made, one between the group exposed to cetirizine during first trimester and the control group (Table 5.1) and the other between the group exposed to cetirizine during the first trimester and the exposed group to cetirizine during second or third trimester (Table 5.2). There was no difference in the rates of major or minor malformation, live births, spontaneous or therapeutic abortions, still births, and rates of cesarean or neonatal distress among the groups. However, there were significant differences between the exposed group to cetirizine during first trimester and the control group in rates of prematurity (P = 0.001), birth weight (P = 0.01) and gestational age at birth (P = 0.006). These differences were not detected between the group exposed to cetirizine in the first trimester and the group exposed to cetirizine during the second or third trimesters. Moreover, sub analysis revealed that offspring of women with asthma, twins and smoking cases receiving cetirizine exhibited significantly lower birth weights and rates of prematurity. After excluding them from the study group, these differences from the control group disappeared (Table 5.3).

Table 5.1. Outcomes of pregnancies in fetuses that had first Trimester cetirizine exposure and control group.

Outcome	Cetirizine T1 group	Control group	*P*
Live birth	73/81	128/135	0.27
Spontaneous abortion	5/81	7/135	0.77
Therapeutic abortion	1/81	0/135	0.38
Stillbirth	2/81	0/135	0.14
Major malformations [a]	2/76	3/129[b]	1.00
Minor malformations [a]	2/76	1/128	0.61
Birth weight, g [c]	3,317±704	3,547± 532	< 0.01
Gestational age [c]	39±2.56	40±1.59	< 0.006
Prematurity [c]	11/73	3/128	< 0.001
Cesarean section [c]	24/70	22/94	0.16
Neonatal distress [c]	9/73	9/128	0.21

[a]: Of live birth, therapeutic abortion and stillbirth.

[b]: Of live birth, therapeutic abortion, stillbirth and one case of spontaneous abortion.

[c]:Of live birth.

Table 5.2. Outcomes of pregnancies in fetuses that had first trimester cetirizine exposure and in fetus exposed to cetirizine during second and/or third trimester.

Outcome	Cetirizine T1 group	Cetirizine non T1 group	*P*
Live birth	73/81	57/57	0.02
Spontaneous abortion	5/81	0/57	0.08
Therapeutic abortion	1/81	0/57	1.00
Stillbirth	2/81	0/57	0.51
Major malformations [a]	2/76	0/57	0.51
Minor malformations [a]	2/76	1/57	1.00
Birth weight, g [b]	3,317±704	3,462±558	0.21
Gestational age [b]	39±3	39±2	0.27
Prematurity [b]	11/73	4/57	0.18
Cesarean section [b]	24/70	16/56	0.57
Neonatal distress [b]	9/73	11/57	0.33

[a]: Of live birth, therapeutic abortion and stillbirth.

[b]: Of live birth.

Table 5.3. Outcomes of pregnancies in fetuses that had first trimester cetirizine exposure and control group after removing of asthma, twins and smoking cases.

Outcome	Cetirizine T1 Group	Control Group	*P*
Live Birth	56/64	119/126	0.15
Birth weight, g [a]	3,449±695	3,560±512	0.24
Gestational age [a]	39±3	40±1	0.11
Prematurity [a]	5/56	3/119	0.11

[a]: Of live birth

5.3.2. Meta-analyses.

The electronic search identified 1500 literature titles. After removing all duplicates and reviewing titles and/or abstracts to exclude the animal studies, non-cohort studies, review articles without original data, and articles that addressed pregnancy outcomes of antihistamines exposure other than cetirizine or hydroxyzine, twelve articles were reviewed in detail. Ten studies fulfilled the inclusion criteria for the meta-analysis including the current cohort study. The other four were excluded because one of them had no healthy control groups [15], one was a case control study [16], one study focused only on one type of malformation [17], and one study was published only as an abstract [18] and the same data were included in a peer reviewed publication [9] (Figure 5.1).

Of the ten studies included in our meta-analyses, four studies included women on hydroxyzine [19, 20, 21, 22], four studies were on cetirizine [10, 23, 24] and two studies were on hydroxyzine and cetirizine [9, 25] (Table 5.4). All the included studies measured the risk of major malformations, whereas five studies assessed the risk of spontaneous abortion [19, 9, 24, 10, current study], and two studies also assessed prematurity [10, current study]. The Q-statistic for heterogeneity of effects was not significant for any of the analyses ($P > 0.05$).

Figure 5.1. Diagram for search strategy and study selection for the meta-analyses.

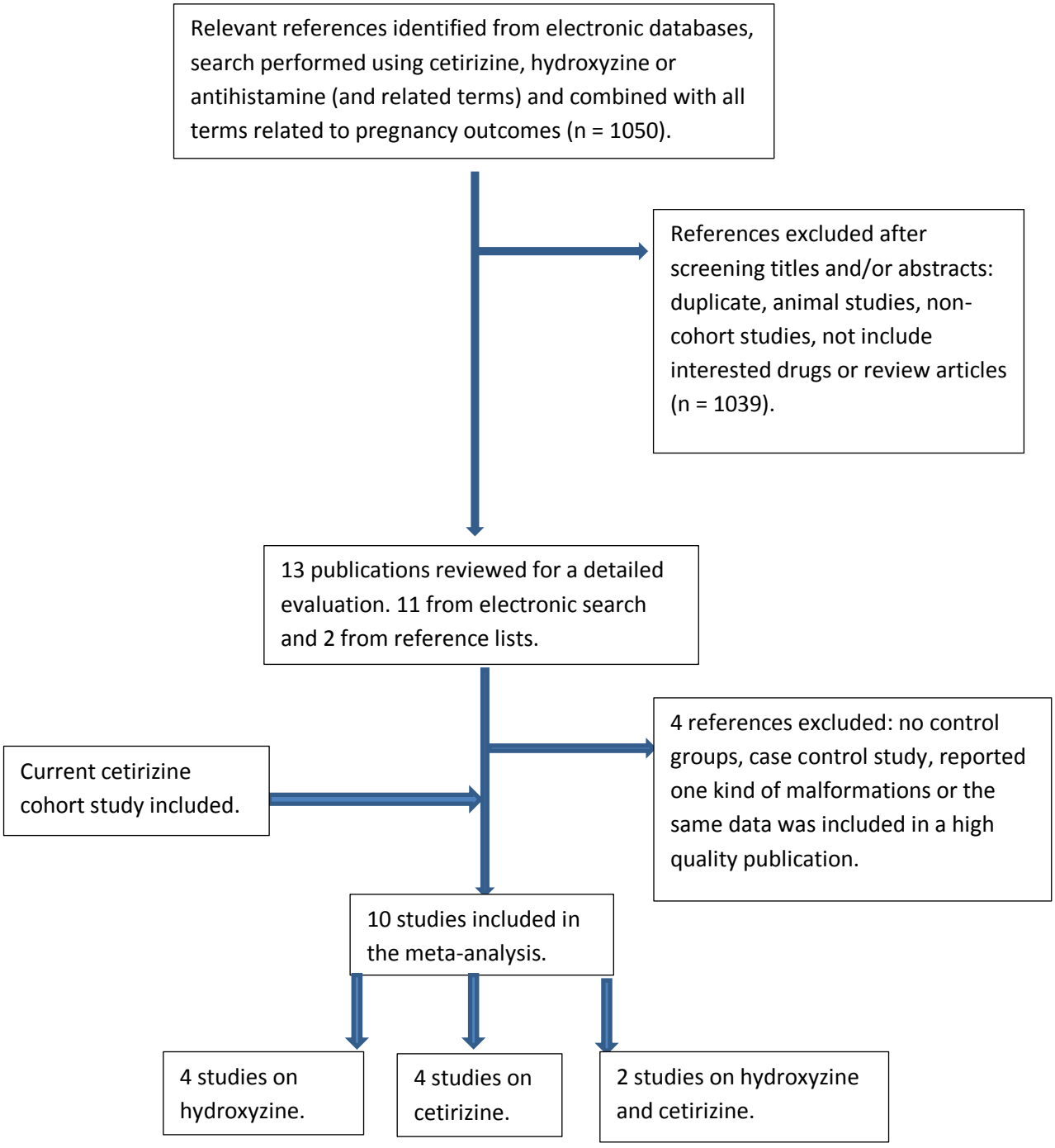

Relevant references identified from electronic databases, search performed using cetirizine, hydroxyzine or antihistamine (and related terms) and combined with all terms related to pregnancy outcomes (n = 1050).
References excluded after screening titles and/or abstracts: duplicate, animal studies, non-cohort studies, not include interested drugs or review articles (n = 1039).
13 publications reviewed for a detailed evaluation. 11 from electronic search and 2 from reference lists.
4 references excluded: no control groups, case control study, reported one kind of malformations or the same data was included in a high quality publication.
Current cetirizine cohort study included.
10 studies included in the meta-analysis.
4 studies on hydroxyzine.
4 studies on cetirizine.
2 studies on hydroxyzine and cetirizine.

Table 5.4. Characteristics of the nine studies included in the meta-analyses.

Study name (year)	Medication name	No. of exposed	No. of controls	Pregnancy outcome evaluated	Publication type
Erez (1971) [19]	Hydroxyzine	79	36	Malformed, SA	Peer review journal
Heinonen (1977) [21]	Hydroxyzine	50	50,232	Malformed	Book
Briggs (1994) [20]	Hydroxyzine	828	228,273	Malformed	Book (personal communication), control group published by Schatz & Petitti
Schatz (1997) [25]	Hydroxyzine, cetirizine	76	82	Malformed	Guest editorial
Einarson (1997) [9]	Hydroxyzine, cetirizine	92	92	Malformed, SA	Peer review journal
Kallen (2002) [23]	cetirizine	917	402,628	Malformed	Peer review journal
Diav-Citrin (2003) [22]	Hydroxyzine	33	844	Malformed	Peer review journal
Paulus (2004) [24]	cetirizine	123	470	Malformed, SA	Abstract
Weber (2008) [10]	cetirizine	177	1,521	Malformed, SA, P	Peer review journal
Current study	Cetirizine	76	129	Malformed, SA, P	Peer review journal

SA: Spontaneous Abortion, P: Prematurity

5.3.2.1. Meta-analysis of hydroxyzine studies that assessed the risk of major malformations.

Data from six studies with a total of 1,082 women exposed to hydroxyzine and 279,480 unexposed controls were included in the meta-analysis. The risk for congenital malformations in the offspring of women exposed to hydroxyzine was not higher than those in the controls that were not exposed to hydroxyzine (OR 1.21; 95% CI 0.92-1.59) (Figure 5.2).

5.3.2.2 Meta-analysis of cetirizine studies that assessed the risk of major malformations.

Six potentially relevant cetirizine studies fulfilled the inclusion criteria for the meta-analysis, two of which were excluded because they reported two zero events in the exposed and in the control groups (odd ratio could not be calculated from these studies) [9, 25]. A total of 1,293 exposed and 404,748 unexposed controls from the remaining four studies were included in the meta-analysis. The odds ratio (95% CI) for incidence of abnormalities after exposure to cetirizine was 1.26 (0.93-1.69) (Figure 5.3).

5.3.2.3. Meta-analysis of combined hydroxyzine and cetirizine studies that assessed the risk of major malformations.

This meta-analysis combined all hydroxyzine and cetirizine studies, with a total of 2,448 exposed and 684,305 unexposed controls. The odds ratio (95% CI) for incidence of abnormalities after exposure to hydroxyzine or cetirizine was 1.23 (1.01-1.51), which was marginally significant (Figure 5.4).

5.3.2.4. Meta-analysis of hydroxyzine and cetirizine studies that assessed the risk of spontaneous abortion.

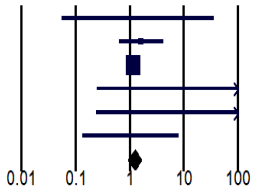
Five studies examined the risk of spontaneous abortion for pregnant women exposed to hydroxyzine or cetirizine, and were included in the meta-analysis (total of 598 exposed and 2,491 unexposed controls). The odd ratio (95% CI) for incidence of spontaneous abortion after exposure to hydroxyzine or cetirizine was 1.09 (0.77-1.53). (Figure 5.5).

5.3.2.5 Meta-analysis of cetirizine studies that assessed the risk of prematurity.

Only two studies reported prematurity in the cetirizine- exposed and control groups, including the current cohort study and there were no hydroxyzine studies that examined the risk of prematurity (total of 233 exposed and 1,640 unexposed controls). The odd ratio (95% CI) for incidence of prematurity after exposure to cetirizine was 1.47 (0.31-7.01) (Figure 5.6).

Figure 5.2. Odds ratios and 95% confidence intervals for malformations in offspring of women using hydroxyzine during pregnancy versus control groups.

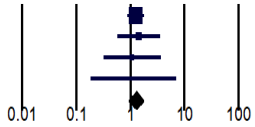
Study name (year)	Statistics for each study			Events / Total		Odds ratio and 95% CI	Relative weight
	Odds ratio	Lower limit	Upper limit	Exposed	Control		
Erez (1971)	1.39	0.06	34.98	1 / 75	0 / 34		0.71
Heinonen (1977)	1.61	0.64	4.06	5 / 50	3243 / 50232		8.70
Briggs (1994)	1.15	0.86	1.54	48 / 828	11579 / 228273		87.19
Schatz (1997)	5.36	0.25	115.00	2 / 43	0 / 44		0.79
Einarson (1997)	5.19	0.24	110.82	2 / 53	0 / 53		0.79
Diav-Citrin (2003)	1.02	0.13	7.79	1 / 33	25 / 844		1.81
	1.21	0.92	1.59				



Test of heterogeneity: P-value = 0.808

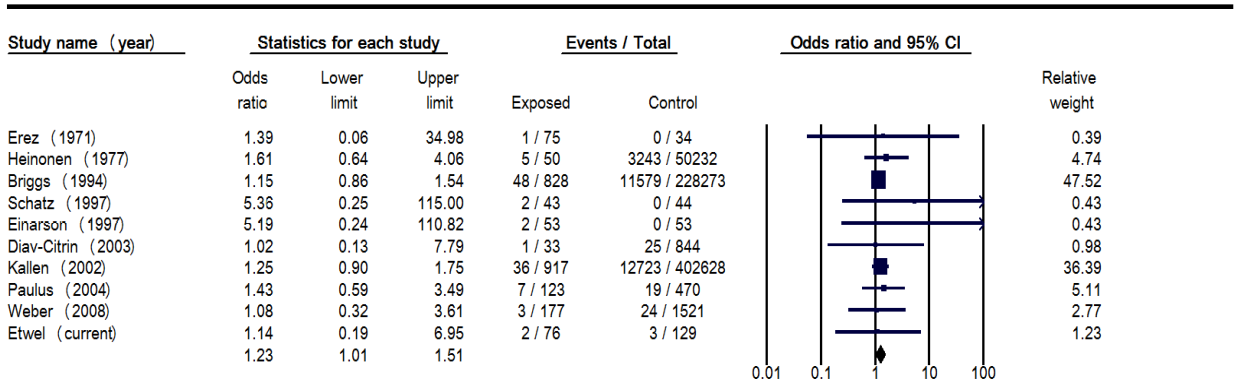
Figure 5.3. Odds ratios and 95% confidence intervals for malformations in offspring of women using cetirizine during pregnancy versus control groups.

Study name （year）	Statistics for each study			Events / Total		Odds ratio and 95% CI	Relative weight
	Odds ratio	Lower limit	Upper limit	Exposed	Control		
Kallen （2002）	1.25	0.90	1.75	36 / 917	12723 / 402628		79.97
Paulus （2004）	1.43	0.59	3.49	7 / 123	19 / 470		11.24
Weber （2008）	1.08	0.32	3.61	3 / 177	24 / 1521		6.08
Etwel （current）	1.14	0.19	6.95	2 / 76	3 / 129		2.71
	1.26	0.93	1.69				



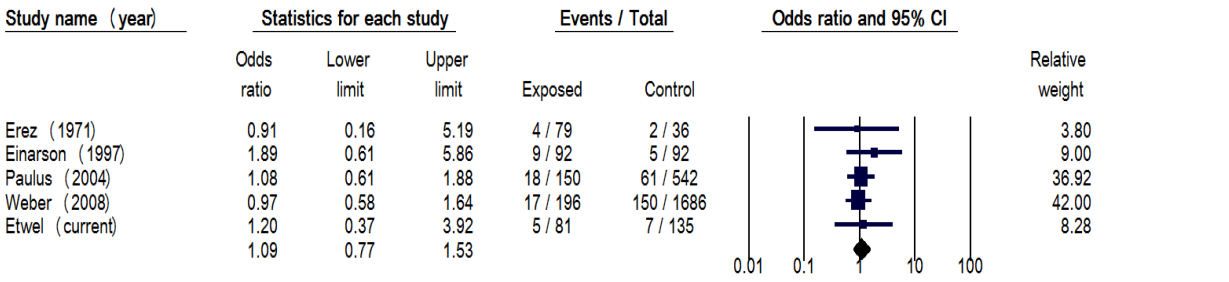
Test of heterogeneity: P-value = 0.984

Figure 5.4. Odds ratios and 95% confidence intervals for malformations in offspring of women using hydroxyzine or cetirizine during pregnancy versus control groups.



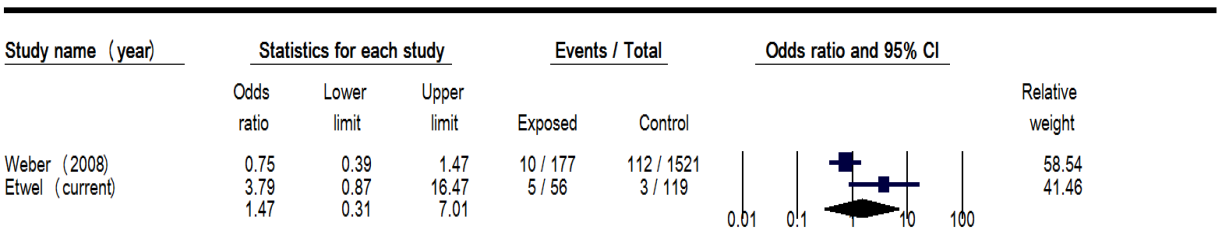
Test of heterogeneity: P-value = 0.981

Figure 5.5. Odds ratios and 95% confidence intervals for spontaneous abortion for women using hydroxyzine or cetirizine during pregnancy versus control groups.



Test of heterogeneity: P-value = 0.886

Figure 5.6. Odd ratios and 95% confident intervals for prematurity in offspring of women using cetirizine during pregnancy versus control groups.



Test of heterogeneity: P-value = 0.050

5.4. Discussion.

5.4.1. Cetirizine cohort study.

The results of our controlled cohort study suggest that this antihistamine, when taken during organogenesis, is not associated with an increased risk of major malformations or spontaneous abortions when compared to non teratogenic medications taken by healthy pregnant women. Importantly, this association was also not present when compared to disease- matched women who took cetirizine after the organogenesis period. These results agree with several other human studies, that there is no risk above the baseline for major malformations when cetirizine is taken during pregnancy [9, 10].

There were statistically significant differences between the first trimester (T1) exposed group and the control group in the rate of twins (P = 0.03) and maternal asthma (P = 0.0007). These two confounders are strongly associated with prematurity [26, 27]. The control group was not matched for smoking, and smoking is also a known cause of prematurity [28]. We analyzed the data after removing these confounders and examined whether the association between cetirizine exposure during organogenesis and prematurity is still evident. After removing all twins, asthma and smoking cases from both groups, the association between cetirizine and prematurity (P = 0.11), birth weight (P = 0.24), and gestational age at birth (P = 0.11) disappeared (Table 5.3). Just as important, the association between prematurity and cetirizine use during the first trimester is not present when we compared it with a disease –match group. These data highlight the importance of controlling for confounding by indication in pregnancy outcome studies.

Our cohort study has limited power to show increased teratogenic risk. Approximately 200 cases and an equal number of matched comparisons are needed

to detect (with a power of 80% and a =0.05) a five- fold increased teratogenic risk above the baseline of 3%. To overcome this hurdle, we increased the sample size by combining the previous human studies in one meta-analysis and estimated the major malformations and other pregnancy outcomes.

5.4.2. Meta-analyses.

The meta-analyses suggest that the use of hydroxyzine or cetirizine does not appear to be associated with a major increased risk for malformations. Although the meta-analysis combining hydroxyzine and cetirizine studies shows a marginal association of an increased risk for major malformation (odd ratio 1.31; 95% CI 1.01-1.51), this result may not be clinically significant. In particular, as shown in our cohort study, there may be confounding by indication, as several studies suggested an increased risk of malformation in offspring of asthmatic patients.

These results were compatible with a case control study published in 2009 [16] that looked at the association between birth defects and antihistamine use during early pregnancy. Moreover, a non-interventional observational cohort study published in 1998 [15] followed women who were exposed to newly marketed drugs at that time, one of them being cetirizine. The results showed that the proportion of live infants with a congenital abnormality born to mothers exposed to newly marketed drugs in the first trimester was similar to the percentage of congenital anomalies in the general population (no congenital cases reported after cetirizine exposure).

Furthermore, secondary analyses of other pregnancy outcomes showed no apparent increased risk for spontaneous abortions , prematurity or after exposure to cetirizine or hydroxyzine.

5.5. Conclusion.

In conclusion, based on the current cohort study and meta-analysis, cetirizine is not associated with an increased risk of major malformations or other adverse fetal outcomes. The study highlights the importance of control for confounding by indication, in this case asthma, which may adversely affect pregnancy outcomes irrespective of cetirizine use.

5.6. References.

1. Casale TB, Blaiss MS, Gelfand E, Gilmore T, Harvey PD, Hindmarch I, et al. First do no harm: Managing antihistamine impairment in patients with allergic rhinitis. J Allergy Clin Immunol. 2003 May;111(5):S835-42.

2. Morgan MM, Khan DA, Nathan RA. Treatment for allergic rhinitis and chronic idiopathic urticaria: Focus on oral antihistamines. Ann Pharmacother. 2005 Dec;39(12):2056-64.

3. Golightly LK, Greos LS. Second-generation antihistamines: Actions and efficacy in the management of allergic disorders. Drugs. 2005;65(3):341-84.

4. Curran MP, Scott LJ, Perry CM. Cetirizine: A review of its use in allergic disorders. Drugs. 2004;64(5):523-61.

5. Keles N. Treatment of allergic rhinitis during pregnancy. Am J Rhinol. 2004 Jan-Feb;18(1):23-8.

6. Mazzotta P, Loebstein R, Koren G. Treating allergic rhinitis in pregnancy. safety considerations. Drug Safety. 1999 Apr;20(4):361-75.

7. Finer LB, Zolna MR. Unintended pregnancy in the united states: Incidence and disparities, 2006. Contraception. 2011 Nov;84(5):478-85.

8. Kamijima M, Sakai Y, Kinoshita K. Reproductive and developmental toxicity studies of cetirizine in rats and rabbits. Clin Report. 1994;28:1877-903.

9. Einarson A, Bailey B, Jung G, Spizzirri D, Baillie M, Koren G. Prospective controlled study of hydroxyzine and cetirizine in pregnancy. Ann Allergy Asthma Immunol. 1997 Feb;78(2):183-6.

10. Weber-Schoendorfer C, Schaefer C. The safety of cetirizine during pregnancy. A prospective observational cohort study. Reproductive Toxicology. 2008;26(1):19-23.

11. Blaiss MS, Food and Drug Administration (U.S.), ACAAI-ACOG(American College of Allergy, Asthma, and Immunology and American College of Obstetricians and Gynecologists.). Management of rhinitis and asthma in pregnancy. Ann Allergy Asthma Immunol. 2003 Jun;90(6 Suppl 3):16-22.

12. Koren G, Feldman Y, Shear N. Motherisk--a new approach to antenatal counselling of drug/chemical exposure. Vet Hum Toxicol. 1986 Dec;28(6):563-5.

13. Barnes CL, McKenzie CA, Webster KD, Poinsett-Holmes K. Cetirizine: A new, nonsedating antihistamine. Ann Pharmacother. 1993 Apr;27(4):464-70.

14. Stroup DF, Berlin JA, Morton SC, Olkin I, Williamson GD, Rennie D, et al. Meta-analysis of observational studies in epidemiology: A proposal for reporting. meta-analysis of observational studies in epidemiology (MOOSE) group. JAMA. 2000 Apr 19;283(15):2008-12.

15. Wilton LV, Pearce GL, Martin RM, Mackay FJ, Mann RD. The outcomes of pregnancy in women exposed to newly marketed drugs in general practice in england. Br J Obstet Gynaecol. 1998 Aug;105(8):882-9.

16. Gilboa SM, Strickland MJ, Olshan AF, Werler MM, Correa A. Use of antihistamine medications during early pregnancy and isolated major malformations. Birth Defects Research Part A - Clinical and Molecular Teratology. 2009;85(2):137-50.

17. Carter PL, Montague JC,Jr, Buffalo MD. Preliminary data relative to the correlation of medications taken during the first trimester of pregnancy and subsequent cleft palate. Folia Phoniatr (Basel). 1980;32(4):298-308.

18. Einarson A; Spizziri D; Berkovich M; Einarson T; Koren G. Prospective study of hydroxyzine use in pregnancy. Reprod Toxicol. 1993,7(6):640.[Abstract].

19. Erez S, Schifrin BS, Dirim O. Double-blind evaluation of hydroxyzine as an antiemetic in pregancy. J Reprod Med. 1971 Jul;7(1):35-7.

20. Briggs GG, Freeman RK, Yaffe SJ, Ovid Technologies I. Drugs in pregnancy and lactation. Baltimore: Williams and Wilkins, 1994.

21. Heinonen OP, Slone D, Shapiro SS. Birth defects and drugs in pregnancy. Littleton, Mass.: Publishing Sciences Group; 1976.

22. Diav-Citrin O, Shechtman S, Aharonovich A, Moerman L, Arnon J, Wajnberg R, et al. Pregnancy outcome after gestational exposure to loratadine or antihistamines: A prospective controlled cohort study. J Allergy Clin Immunol. 2003 Jun;111(6):1239-43.

23. Kallen B. Use of antihistamine drugs in early pregnancy and delivery outcome. J Matern Fetal Neonatal Med. 2002 Mar;11(3):146-52.

24. W. Paulus, S. Schloemp, K. Sterzik and F. Stoz,. Pregnancy outcome after exposure to cetirizine/levocetirizine in the first trimester—a prospective controlled study, . reprod toxicol, 19 (2) (2004), p. 258 [abstract]. Reproductive Toxicology. 2004;19(2):239-60 [Abstract].

25. Schatz M, Petitti D. Antihistamines and pregnancy. Ann Allergy Asthma Immunol. 1997 Feb;78(2):157-9.

26. Murphy VE, Namazy JA, Powell H, Schatz M, Chambers C, Attia J, et al. A meta-analysis of adverse perinatal outcomes in women with asthma. BJOG. 2011 Oct;118(11):1314-23.

27. Refuerzo JS. Impact of multiple births on late and moderate prematurity. Semin Fetal Neonatal Med. 2012 Jun;17(3):143-5.

28. Shah NR, Bracken MB. A systematic review and meta-analysis of prospective studies on the association between maternal cigarette smoking and preterm delivery. Am J Obstet Gynecol. 2000 Feb;182(2):465-72.

Chapter 6: The risk of adverse pregnancy outcome after first trimester exposure to H1 antihistamines: a systematic review and meta-analysis.

This chapter has been accepted for publication (in press)

Etwel F, Faught LH, Rieder MJ, Koren G. The risk of adverse pregnancy outcome after first trimester exposure to H1 antihistamines: a systematic review and meta-analysis. Drug Safety 2016.

6.1. Introduction.

Antihistamines (AHs) are among the most commonly prescribed drugs during pregnancy, with approximately 15% of pregnant women reporting the use of over-the-counter or prescribed AHs at some point during their pregnancy, particularly during the first trimester [1-3]. AHs are classified as either H1 or H2 with reference to the relative selectivity of their targeting receptor. H1-AHs are used for the treatment of nausea and vomiting during pregnancy (NVP), which occurs in approximately 85% of pregnancies as well as for the symptomatic relief of asthma, urticaria, allergy, the common cold, and other relatively minor conditions [4, 5]. H2-AHs are used to treat indigestion and acid reflux [6]. H1-AH exposure typically occurs most commonly during the first trimester, while H2-AH exposure is more common thereafter as NVP tends to present during the first trimester and resolve in the early second trimester and gastric symptoms usually appear later in pregnancy [7]. Given the large number of pregnant women exposed to H1-AHs during the first trimester - a critical time for fetal development - there is a compelling need to examine any potential risks arising from their use during pregnancy.

The gold standard of clinical research is the double blind randomized placebo control trial. Unfortunately, this approach is ethically unacceptable when studying drug safety in pregnancy due to the possibility of exposing pregnant mothers and their unborn babies to potentially harmful treatments [8]. Consequently, the most practical approach to quantify the safety and risks of H1-AH exposure during the first trimester is to conduct a systematic review and meta-analysis of all available observational cohort and case control studies of exposed mothers [9]. Previous research on the safety of H1-AH use during pregnancy does exist in the form of meta-analyses. Unfortunately the information from these

studies is contradictory, outdated or specific to one type of H1-AH, leaving questions surrounding the safety of all available H1-AHs.

The first meta-analysis conducted over 20 years ago included all the studies available at that time and concluded that H1-AHs can safely be used during pregnancy [10]. However, upon re-analysis of all this data, a second group produced contradictory results with respect to the cohort studies meta-analysis, in which they showed an increased risk of major malformation in those exposed to H1-AHs. The same study demonstrated no increased risk when analyzing case control studies or the studies that focused only on doxylamine [11]. Other studies have included meta-analyses on doxylamine when used for treatment of NVP [12-14], loratidine, hydroxyzine, and cetirizine [15, 16], each generating reassuring results. The primary objective of the present study is to determine whether H1-AHs, used in the treatment of any condition during the first trimester of pregnancy, are associated with an increased rate of major malformation. Secondary objectives include assessing the safety of H1-AHs used specifically for the treatment of NVP as well as examining the effect of H1-AHs on other pregnancy outcomes, including spontaneous abortions, prematurity, stillbirth, and low birth weight, following first trimester exposure.

6.2. Methods.

A systematic review and meta-analyses were conducted on all observational cohort and case control studies published that addressed the effect of H1-AH on pregnancy outcomes.

6.2.1. Search strategy.

Following the guidelines of PRISMA [17], a systematic review was performed to retrieve all published articles involving H1-AH exposure during

pregnancy. Electronic databases including PubMed and EMBASE were searched from inception till 10 January 2016 for relevant articles published in any language. Search strategies are presented in Appendix 5. Subsequently, the reference lists of all collected studies were reviewed for articles not previously identified by the search strategy.

6.2.2. Study selection.

Any published human study that met the following criteria was included in the meta-analyses:

1- Observational cohort or case control studies that clearly confirmed in the original article the exposure to H1-AH during first trimester and those studies that had enough data to select the group that had first trimester exposure.

2- Studies that had sufficient data to select only major malformation and/or other pregnancy outcomes in the offspring of women who were exposed to one or more types of H1-AH during the first trimester of pregnancy and were compared to a control group, where the control consisted of women who were not exposed to any drug throughout their entire pregnancy and/or women who were exposed to drugs other than specific H1-AHs under study.

3- Studies that provided sufficient data to calculate the odds ratios (ORs) and the 95% confidence intervals (95% CI).

4- Studies with a sample size larger than 10.

5- Studies that focused only on medications used for therapeutic purposes.

6- Updated studies by the same group of investigators on the same type of H1-AHs were selected to prevent duplication of overlapping reports.

Two authors (FE and MJR) screened the titles and abstracts of all studies identified by the electronic search to determine whether or not they met inclusion criteria to evaluate the full text. Full text of likely studies for eligibility was reviewed by the two authors (FE and LHF). Disagreements were resolved by a third author (GK).

6.2.3. Data extraction.

Information from each study was collected with the use of a data extraction form (Appendix 6). Information collected included the drug name of H1-AH , first author, year of publication, journal name, study design, study location, year of study, whether exposure occurred during the first trimester, type of control, and outcome measures.

Outcome measures were extracted for both the exposed and control groups and included a number of the following: pregnant women, live births, major malformations (any structural defect that caused significant medical, surgical, or cosmetic problems) [18], preterm infants (infants born alive before 37 weeks' gestation), spontaneous abortions (miscarriage; loss of pregnancy before 20 weeks' gestation), stillbirths (fetal death after 20 weeks ' gestation), and low birth weight infants (live born infants of less than 2,500 g (5 pounds 8 ounces))[19]. The original studies must have used the same terminology for the outcome measures and/or its definitions in order to be included. Also, if detailed information about malformation was reported in the original study, screening of the major malformation was performed. Authors of included studies were not contacted to obtain data not reported in their original publication.

All the data were arranged in 2x2 tables to calculate the ORs and 95% CI and all the outcomes were considered binary outcomes (the adverse outcomes had

two possible outcomes: all or nothing). When calculating ratios for spontaneous abortions, as well as for stillbirth, the denominator was the total number of pregnancies. For prematurity, low birth weight, and major malformation outcomes, ratios were calculated using live birth when this information was available. Stillbirths, elective, therapeutic and/or spontaneous abortions that were diagnosed as major malformations were all counted as major malformation cases.

6.2.4. Data analysis.

Risk data for malformation and other pregnancy outcomes collected in all studies was combined with the Comprehensive Meta-Analysis Version 2.0, using a random-effects model. Both odds ratios and 95% confidence intervals were calculated for each outcome. Heterogeneity among studies was assessed using the Q statistic, which was then quantified by I2. A significant Q statistic ($P<0.05$), represents a high degree of variance among the studies analyzed. An associated I2 value between 0% to 40% might not be important, while between 30% to 60% may represent moderate heterogeneity, between 50% to 90% may represent substantial heterogeneity, and between 75% to 100% considerable heterogeneity [20]. Four separate meta-analyses were conducted to assess the risk of major malformations:

1) All H1-AH exposed cohort studies.

2) Sensitivity analysis of H1-AH cohort studies excluding studies where the comparison group may have had some H1-AH exposure other than the drug under investigation.

3) H1-AHs only used for NVP.

4) All H1-AH case control studies.

Four separate meta-analyses were performed for all other collected pregnancy outcomes (prematurity, spontaneous abortion, stillbirth, and low birth weight).

6.2.5. Analysis of potential publication bias.

Funnel plots were generated using the Duval and Tweedie's trim and fill method for each meta-analysis where the number of studies included was greater than 10. Funnel plots were visually inspected in order to assess for publication bias. The number of unpublished studies (K) that were potentially omitted from the primary analysis was determined; if evidence of publication bias existed (K>0), adjusted point estimates (ORs) were calculated based on the number of omitted studies [21].

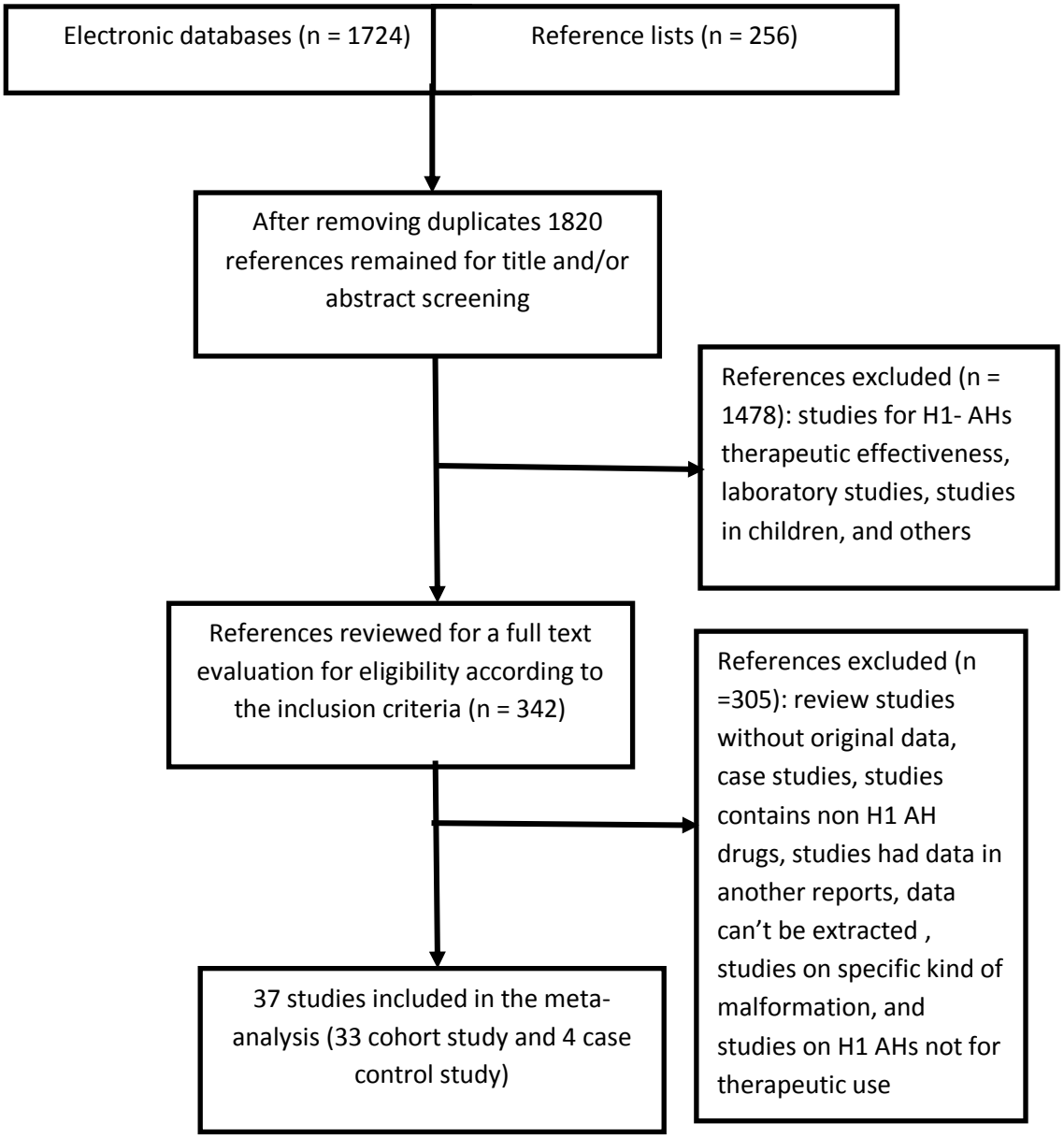
6.2.6. Quality assessment.

The Newcastle-Ottawa assessment scale for cohort studies and case control studies was used to evaluate the quality of included studies in the meta-analyses [22]. The evaluation focused on the following three categories: the selection category ranged from 0–4 points, the comparability category ranged from 0–2 points, and the outcome category ranged from 0–3 points (the outcome category was for cohort studies and for case control studies was called the exposure category). The scale range was from 0 to 9. For the comparability category, controlling for maternal age was the most important factor, as studies were given 1 point for this factor and if any other factors such as nicotine consumption, drinking alcohol, diabetes, hypertension, previous abortions and/or previous malformed children were controlled for, they received 2 points. For the outcome category for the cohort studies, the follow up period for major malformation had to be at least 6 months.

6.3. Results.

An electronic search of all databases identified a total of 1724 manuscripts (Figure 6.1). After removing all duplicate publications and excluding animal studies, and studies on drug effectiveness, 342 articles were reviewed in detail. A total of 37 studies (33 cohort studies [7, 16, 23-52] and 4 case control studies [3, 53-55] fulfilled the inclusion criteria for the meta-analysis (Table 6 for characteristics of the included studies (Appendix 7)).

Figure 6.1. Flow chart for study selection for the meta-analyses.


Electronic databases (n = 1724)
Reference lists (n = 256)
After removing duplicates 1820 references remained for title and/or abstract screening
References excluded (n = 1478): studies for H1- AHs therapeutic effectiveness, laboratory studies, studies in children, and others
References reviewed for a full text evaluation for eligibility according to the inclusion criteria (n = 342)
References excluded (n =305): review studies without original data, case studies, studies contains non H1 AH drugs, studies had data in another reports, data can't be extracted , studies on specific kind of malformation, and studies on H1 AHs not for therapeutic use
37 studies included in the meta-analysis (33 cohort study and 4 case control study)

The average quality score of the included studies was 6.08 (± 1.95 (SD)) out of 9 according to the Newcastle-Ottawa assessment scale. All the included studies assessed major malformations or major malformation plus other pregnancy outcomes, except one cohort study that did not assess major malformations but rather spontaneous abortions and prematurity [7].

6.3.1. Meta-analysis of cohort studies assessing risk of major malformations.

Data from 32 cohort studies [16, 23-52] with a total of 49,635 women exposed to H1-AH and 1,302,596 unexposed controls were included in this meta-analysis. The risk of major malformation in the offspring of women exposed to H1-AHs was not higher than that of the control population (OR 1.07; 95% CI 0.98-1.16) (Figure 6.2). The Q-statistic for heterogeneity of effects was not significant ($P > 0.05$, $I2 < 25\%$) and there was no evidence of publication bias in this analysis (Figure 6.3).

6.3.2. Sensitivity analysis of the meta-analysis of cohort studies assessing risk of major malformations.

Four cohort studies were excluded [30, 32, 41, 45], since exposure of the control group to an H1-AH not under investigation could not be ruled out for these studies. For example, specific H1-AHs were being studied and compared to a control group, which may have been exposed to antiemetic drugs that have H1-AHs such as doxylamine. The remaining 28 cohort studies [16, 23-29, 31, 33-40, 42-44, 46-52] with a total of 21,427 women exposed to H1-AHs and 449,939 unexposed controls were included in the sensitivity analysis. The risk of major malformation in the offspring of women exposed to H1-AHs was not higher than that of the control groups (OR 1.01; 95% CI 0.90-1.12) (Figure 6.4). The Q-statistic for heterogeneity of effects was not significant ($P > 0.05$, $I2 = 0\%$) and there was no evidence of publication bias in this analysis.

6.3.3. Meta-analysis of cohort studies assessing risk of major malformations for H1-AHs used to treat NVP.

There were 18 cohort studies [23-29, 31, 33-37, 44, 46, 50, 52] that studied H1-AHs as an antiemetic. A total of 27,243 women who were exposed to H1-AHs for treatment of NVP and 441,623 unexposed controls were included in this meta-analysis. The risk of major malformation in the offspring of women exposed to H1-AHs when used for the treatment of NVP was not higher than the control group (OR 0.95; 95% CI 0.87-1.05) (Figure 6.5). There was no indication of difference in risk of major malformations in the offspring of women exposed to H1-AHs compared to control. The Q-statistic for heterogeneity of effects was not significant ($P > 0.05$, $I2 = 0\%$) and there was no evidence of publication bias in this analysis.

6.3.4. Meta-analysis for case control studies assessing risk of major malformations.

Four case control studies [3, 53-55] fulfilled the inclusion criteria; a total of 7,270 women exposed to H1-AHs and 90,336 unexposed controls were included in the meta-analysis. The risk of major malformation in the offspring of women exposed to H1-AHs was not higher than that of the control group (OR 1.05; 95% CI 0.90-1.23) (Figure 6.6). The Q-statistic for heterogeneity of effects was not significant ($P > 0.05$, $I2 = 0\%$).

6.3.5. Meta-analysis of cohort studies assessing the risk of prematurity.

Nine cohort studies reported prematurity outcomes [7, 16, 29, 42-44, 47, 51, 52] for a total of 1,799 H1-AHs exposed women and 9,156 unexposed controls. The odds ratio (95% CI) for the incidence of prematurity after exposure to H1-AHs was 0.96 (0.76-1.20). The Q-statistic for heterogeneity of effects was not significant ($P > 0.05$) with the I2 value was between 25%-50% which indicates low heterogeneity (Figure 6.7).

6.3.6. Meta-analysis of cohort studies assessing the risk of spontaneous abortion.

Thirteen cohort studies reported spontaneous abortion outcomes [7, 16, 23, 27, 39, 40, 42, 47-52] for a total of 2,522 H1-AH exposed women and 7,276 unexposed controls. The odds ratio (95% CI) for the incidence of spontaneous abortion after exposure to H1-AHs was 1.00 (0.83-1.20) (Figure 6.8). The Q-statistic for heterogeneity of effects was not significant ($P > 0.05$, $I2 = 0\%$), and there was no evidence of publication bias in this analysis.

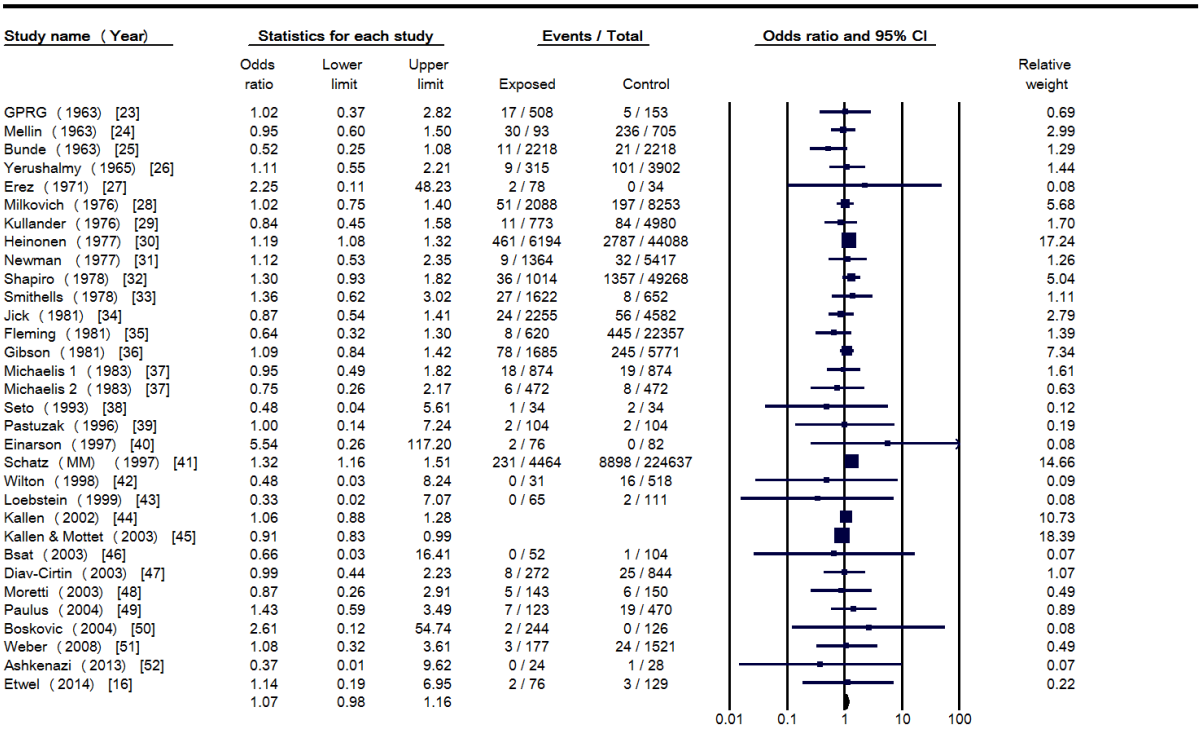
6.3.7. Meta-analysis of cohort studies assessing the risk of stillbirth.

Eight cohort studies reported stillbirth outcomes [7, 16, 23, 39, 40, 43, 48, 50] for a total of 1,571 H1-AH exposed women and 3,328 unexposed controls. The odds ratio (95% CI) for the incidence of stillbirth after exposure to H1-AHs was 1.23 (0.48-3.18). The Q-statistic for heterogeneity of effects was not significant ($P > 0.05$, $I2 < 25\%$) (Figure 6.9).

6.3.8. Meta-analysis of cohort studies assessing the risk of low birth weight.

Three cohort studies reported low birth weight outcomes [43, 50, 52] for a total of 265 H1-AH exposed women and 384 unexposed controls. The odds ratio (95% CI) for the incidence of low birth weight after exposure to H1-AHs was 1.20 (0.63-2.29). The Q-statistic for heterogeneity of effects was not significant ($P > 0.05$, $I2 = 0\%$) (Figure 6.10).

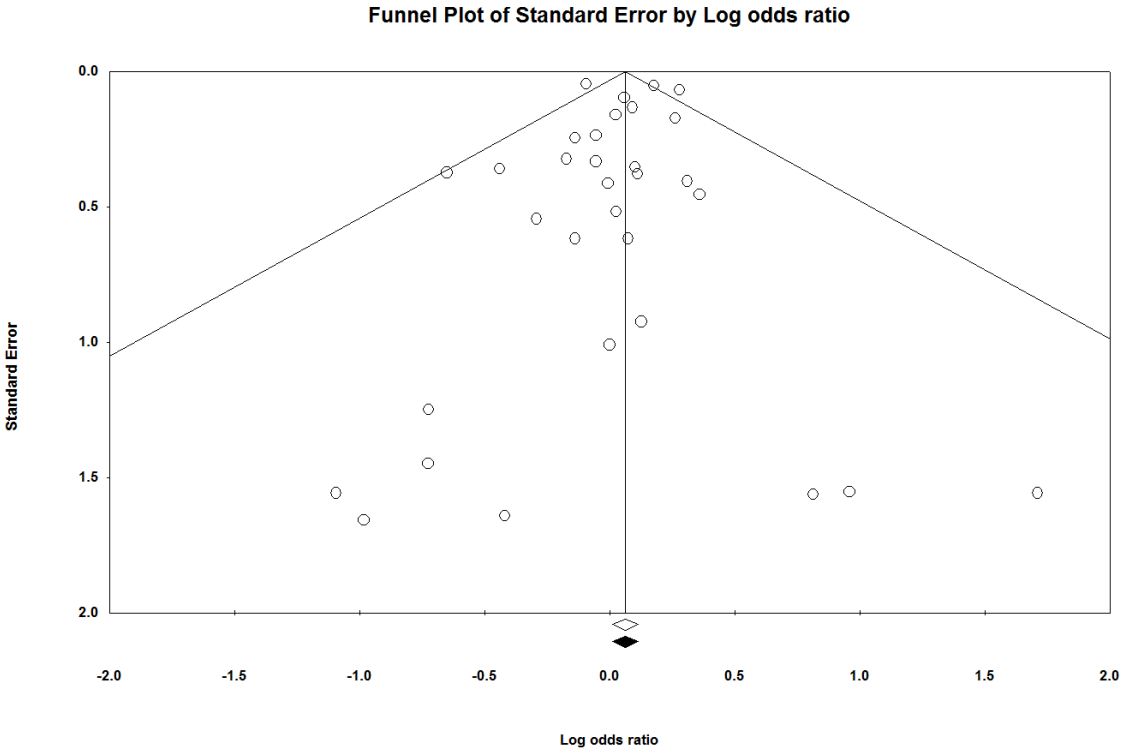
Figure 6.2. Forest plots of all H1-AHs cohort studies that assessed the risk of major malformations.



Total odds ratio (95% CI) = 1.07 (0.98- 1.16)

Test of heterogeneity: Q = 40.35, df = 31 (P = 0.12), I^2 = 23.16%

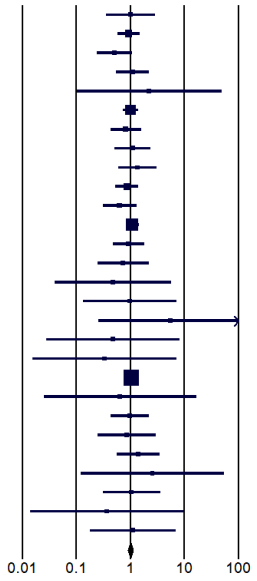
Figure 6.3. Publication bias using funnel plot for meta-analysis of all H1-AHs cohort studies that assessed the risk of major malformations.



Open circles represent the included studies, the open rhombus is the observed measure of effect, and the closed rhombus is the adjusted measure of effect.

Figure 6.4. Forest plots of H1-AHs cohort studies excluding the studies that the comparison group may have some H1-AHs exposure that assessed the risk of major malformations (sensitivity analysis).

Study name (Year)	Statistics for each study			Events / Total		Odds ratio and 95% CI	Relative weight
	Odds ratio	Lower limit	Upper limit	Exposed	Control		
GPRG (1963) [23]	1.02	0.37	2.82	17 / 508	5 / 153		1.17
Mellin (1963) [24]	0.95	0.60	1.50	30 / 93	236 / 705		5.63
Bunde (1963) [25]	0.52	0.25	1.08	11 / 2218	21 / 2218		2.24
Yerushalmy (1965) [26]	1.11	0.55	2.21	9 / 315	101 / 3902		2.51
Erez (1971) [27]	2.25	0.11	48.23	2 / 78	0 / 34		0.13
Milkovich (1976) [28]	1.02	0.75	1.40	51 / 2088	197 / 8253		12.36
Kullander (1976) [29]	0.84	0.45	1.58	11 / 773	84 / 4980		3.00
Newman (1977) [31]	1.12	0.53	2.35	9 / 1364	32 / 5417		2.18
Smithells (1978) [33]	1.36	0.62	3.02	27 / 1622	8 / 652		1.90
Jick (1981) [34]	0.87	0.54	1.41	24 / 2255	56 / 4582		5.20
Fleming (1981) [35]	0.64	0.32	1.30	8 / 620	445 / 22357		2.43
Gibson (1981) [36]	1.09	0.84	1.42	78 / 1685	245 / 5771		17.67
Michaelis 1 (1983) [37]	0.95	0.49	1.82	18 / 874	19 / 874		2.83
Michaelis 2 (1983) [37]	0.75	0.26	2.17	6 / 472	8 / 472		1.06
Seto (1993) [38]	0.48	0.04	5.61	1 / 34	2 / 34		0.20
Pastuzak (1996) [39]	1.00	0.14	7.24	2 / 104	2 / 104		0.31
Einarson (1997) [40]	5.54	0.26	117.20	2 / 76	0 / 82		0.13
Wilton (1998) [42]	0.48	0.03	8.24	0 / 31	16 / 518		0.15
Loebstein (1999) [43]	0.33	0.02	7.07	0 / 65	2 / 111		0.13
Kallen (2002) [44]	1.06	0.88	1.28				33.06
Bsat (2003) [46]	0.66	0.03	16.41	0 / 52	1 / 104		0.12
Diav-Cirtin (2003) [47]	0.99	0.44	2.23	8 / 272	25 / 844		1.84
Moretti (2003) [48]	0.87	0.26	2.91	5 / 143	6 / 150		0.82
Paulus (2004) [49]	1.43	0.59	3.49	7 / 123	19 / 470		1.52
Boskovic (2004) [50]	2.61	0.12	54.74	2 / 244	0 / 126		0.13
Weber (2008) [51]	1.08	0.32	3.61	3 / 177	24 / 1521		0.82
Ashkenazi (2013) [52]	0.37	0.01	9.62	0 / 24	1 / 28		0.11
Etwel (2014) [16]	1.14	0.19	6.95	2 / 76	3 / 129		0.37
	1.01	0.90	1.12				

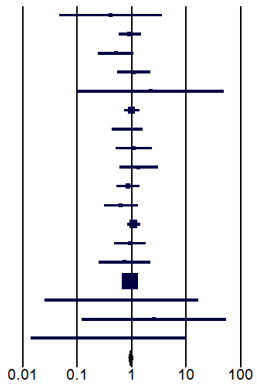


Total odds ratio (95% CI) = 1.01 (0.90- 1.12)

Test of heterogeneity: Q = 11.19, df = 27 (P = 1.00), I^2 = 0.00%

Figure 6.5. Forest plots of H1-AHs used to treat NVP cohort studies that assessed the risk of major malformations.

Study name (Year)	Statistics for each study			Events / Total		Odds ratio and 95% CI	Relative weight
	Odds ratio	Lower limit	Upper limit	Exposed	Control		
GPRG (1963) [23]	0.42	0.05	3.64	1 / 72	5 / 153		0.19
Mellin (1963) [24]	0.95	0.60	1.50	30 / 93	236 / 705		4.18
Bunde (1963) [25]	0.52	0.25	1.08	11 / 2218	21 / 2218		1.67
Yerushalmy (1965) [26]	1.11	0.55	2.21	9 / 315	101 / 3902		1.87
Erez (1971) [27]	2.25	0.11	48.23	2 / 78	0 / 34		0.10
Milkovich (1976) [28]	1.02	0.75	1.40	51 / 2088	197 / 8253		9.19
Kullander (1976) [29]	0.84	0.45	1.58	11 / 773	84 / 4980		2.23
Newman (1977) [31]	1.12	0.53	2.35	9 / 1364	32 / 5417		1.62
Smithells (1978) [33]	1.36	0.62	3.02	27 / 1622	8 / 652		1.42
Jick (1981) [34]	0.87	0.54	1.41	24 / 2255	56 / 4582		3.86
Fleming (1981) [35]	0.64	0.32	1.30	8 / 620	445 / 22357		1.80
Gibson (1981) [36]	1.09	0.84	1.42	78 / 1685	245 / 5771		13.14
Michaelis 1 (1983) [37]	0.95	0.49	1.82	18 / 874	19 / 874		2.10
Michaelis 2 (1983) [37]	0.75	0.26	2.17	6 / 472	8 / 472		0.79
Kallen (2002) [44]	0.94	0.83	1.07				55.57
Bsat (2003) [46]	0.66	0.03	16.41	0 / 52	1 / 104		0.09
Boskovic (2004) [50]	2.61	0.12	54.74	2 / 244	0 / 126		0.10
Ashkenazi (2013) [52]	0.37	0.01	9.62	0 / 24	1 / 28		0.08
	0.95	0.87	1.05				

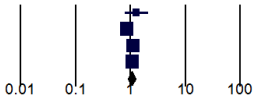


Total odds ratio (95% CI) = 0.95 (0.87- 1.05)

Test of heterogeneity: Q = 8.42, df = 17 (P = 0.96), I^2= 0.00%

Figure 6.6. Forest plots of H1-AHs case control studies that assessed the risk of major malformations.

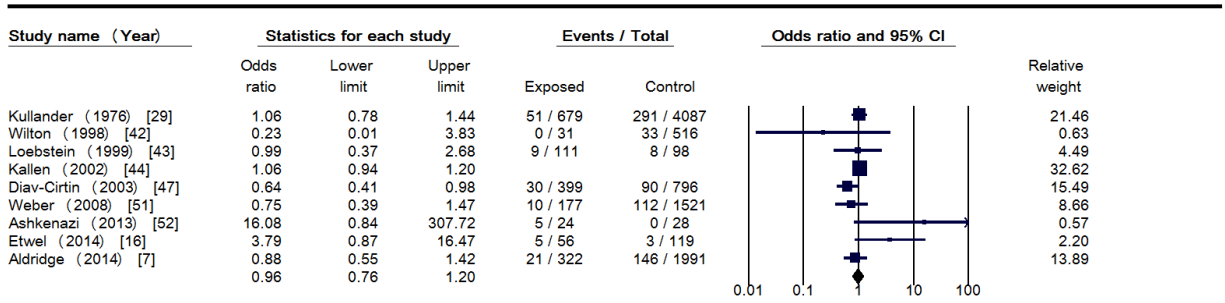
Study name （Year）	Statistics for each study			Events / Total		Odds ratio and 95% CI	Relative weight
	Odds ratio	Lower limit	Upper limit	Exposed	Control		
Eskenazi （1982） [53]	1.29	0.80	2.08	22 / 122	333 / 2293		8.31
Czeizel （2005） [54]	0.88	0.81	0.95	914 / 2640	21929 / 58354		31.29
Gilboa （2009） [55]	1.14	1.02	1.28	1064 / 1582	7975 / 12420		29.21
Li （2013） [3]	1.10	1.01	1.20	1969 / 2926	11244 / 17269		31.20
	1.05	0.90	1.23				



Total odds ratio (95% CI) = 1.05 (0.90- 1.23)

Test of heterogeneity: Q = 21.01, df = 3 (P = 1.00), I^2 = 0.00%

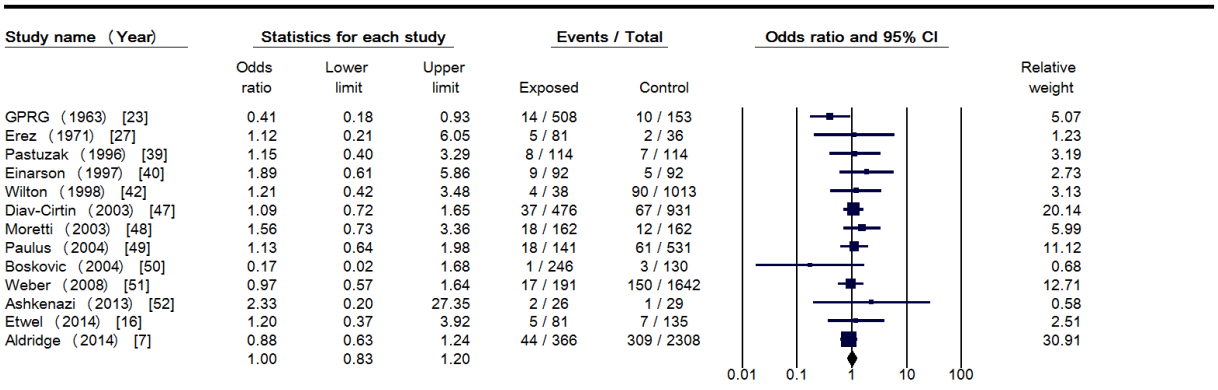
Figure 6.7. Forest plots of H1-AHs cohort studies that assessed the risk of prematurity.



Total odds ratio (95% CI) = 0.96 (0.76- 1.20)

Test of heterogeneity: Q = 13.64, df = 8 (P = 0.09), I^2 = 41.36%

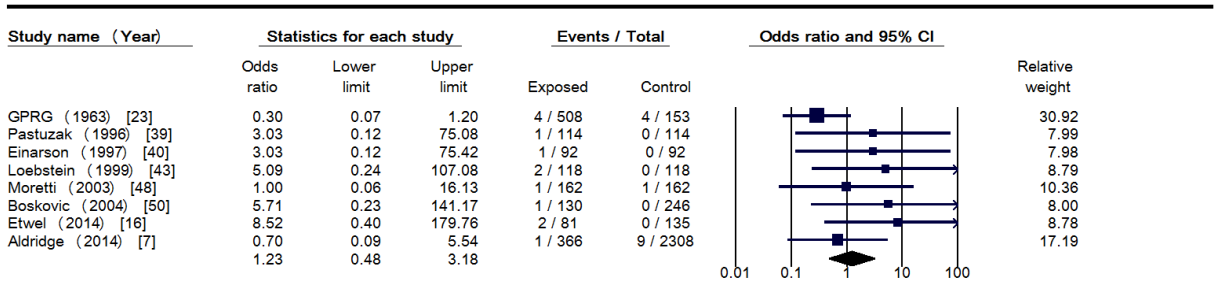
Figure 6.8. Forest plots of H1-AHs cohort studies that assessed the risk of spontaneous abortion.



Total odds ratio (95% CI) = 1.00 (0.83- 1.20)

Test of heterogeneity: Q = 10.93, df = 12 (P = 0.54), I^2 = 0.00%

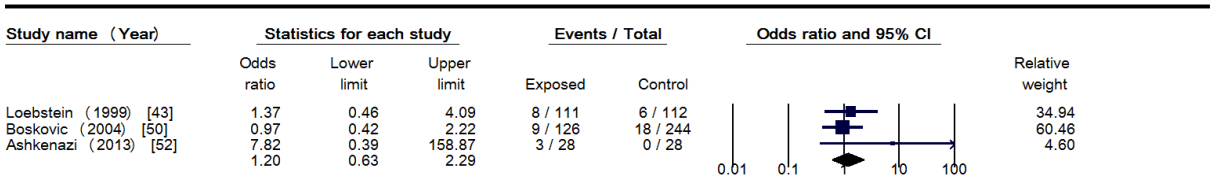
Figure 6.9. Forest plots of H1-AHs cohort studies that assessed the risk of stillbirth.



Total odds ratio (95% CI) = 1.23 (0.48- 3.18)

Test of heterogeneity: Q = 8.05, df = 7 (P = 0.33), I^2 = 13.09%

Figure 6.10. Forest plots of H1-AHs cohort studies that assessed the risk of low birth weight



Total odds ratio (95% CI) = 1.20 (0.63- 2.29)

Test of heterogeneity: Q = 1.81, df = 2 (P = 0.41), I^2 = 0.00%

6.4. Discussion.

This study provides a quantitative estimate of the risk of adverse pregnancy outcomes following first trimester exposure to H1-AHs. The study of H1-AHs in pregnancy, which includes developing a clear picture of the safety and possible risks associated with their use, is important given their wide use by pregnant women. As a consequence of being available both over the counter and as a prescription medication, intentional and accidental exposures are frequent in the first and subsequent trimesters. To date, the majority of research on H1-AH exposure in pregnancy has been reassuring, providing evidence suggesting there is no increased risk associated with their use. Some conflicting studies, however, do exist, including several original research studies and one meta-analysis [11]. This highlights the need for an up-to-date review and analysis of H1-AH safety in pregnancy that includes all new available research studies as well as those previously reviewed.

Four different meta-analyses were conducted by us to address the potential effects of H1-AHs with respect to major malformations. As we collected studies with two different designs, cohort and case control, meta-analyses were conducted separately for each type.

The first meta-analysis included all 32 available cohort studies, which met the study inclusion criteria. The results of this meta-analysis contradict the most recently published meta-analysis on H1-AH safety based on cohort studies, published in 2014 by Chin et al. [11]. However, this particular meta-analysis included only 11 studies compared to our 33, with their most recent study being in 1993 [38] whereas ours being in 2014 [16] The lack of randomized studies on H1-AH in pregnancy, similar to lack of randomized studies of virtually all drug classes

in pregnancy, increases the risk of bias and hence, the need to carefully assess such potential bias.

Of potential importance, given our inclusion criteria, there were three studies which were excluded from our analysis but whose results add to the H1-AH safety profile [56-58]. In these studies H1-AH use was not therapeutic but rather as an overdose or abuse. The results of these studies that showed no association between H1-AH use and major malformation are important as the exposure of these pregnant women was to much higher doses than what would have been experienced in therapeutic use. These findings further corroborate our hypothesis that H1-AHs are not teratogenic.

Following our analysis of all 32 cohort studies, we performed sensitivity analyses, trying to address some potential bias, by excluding those studies where members of the control group might have been exposed to an H1-AH other than the drug under investigation. As an example, if meclizine safety was being assessed and compared to a group of women not exposed to meclizine (but where they could have been exposed to drugs other than meclizine such as doxylamine), we could not ascertain that women in the control group were not exposed to a different H1-AH, unless clearly stated by the authors. While still able to provide us with some safety information, this was not an ideal control group for our analysis. The results of our sensitivity analysis were consistent with the original analysis where no association between H1-AH use and major malformation was found.

We have also carried out a separate analysis looking at studies where H1-AHs were used to treat NVP. A total 18 of our 32 cohort studies looked specifically at H1-AH used for the treatment of NVP. The results showed no increase in the risk of major malformations in the offspring of women exposed to

H1-AHs used to treat NVP compared to control. There is ample evidence that NVP renders protective effects on pregnancy outcome, with mothers who experience morning sickness having better birth outcomes, including reduced risks of spontaneous abortion, preterm birth, malformation, and children with higher IQ [59]. Our analysis for this potential bias by indication ruled out a significant bias.

Four case control studies met the inclusion criteria and were included in this meta-analysis. Although there were many additional case control studies available in the literature, they could not be included in our meta-analysis since they each assessed the risk of only one type of malformation. The results of the case control study analysis did not show an increase in risk of major malformation following H1-AHs use. Since, the meta-analyses for cohort studies and case control studies conducted in this current study cannot rule out an increased risk for specific congenital anomalies, this limitation should be addressed in future research.

To our knowledge, this is the first meta-analysis addressing pregnancy outcomes other than major malformations, following exposure to any H1-antihisimines. These included the potential effect of H1-AHs on prematurity, spontaneous abortion, low birth weight and stillbirth.

In looking at prematurity, nine cohort studies were available with outcomes. Individually all nine studies showed no increase in the risk of prematurity following H1-AH exposure and therefore, as expected, the combined results of the meta-analysis also found no increased risk for prematurity.

When evaluating the effects of H1-AH on spontaneous abortion, 13 studies were available with outcomes and included in the meta-analysis. Individually and combined, these studies showed no increased risk of spontaneous abortion following H1-AHs exposure. Importantly, results of the initial analysis

demonstrated evidence of publication bias that required adjusting the overall estimate to overcome the presence of bias. After the adjustment, the overall result did not change, indicating there is no increased risk of spontaneous abortion associated with H1-AH use. This particular meta-analysis is important because animal models assessing the safety of H1-AHs during pregnancy have suggested that they may negatively impact the implantation process [60]. Reassuringly, this meta-analysis shows no increased risk of spontaneous abortion following therapeutic use of this medication.

Eight cohort studies assessed stillbirth following H1-AH exposure. The eight studies both individually and when combined showed no increase in the risk of stillbirth between exposed and control groups. Since the original studies did not examine the cause of stillbirth in these groups, we cannot rule out the association between the H1-AH use and stillbirth outcome.

Three studies assessed low birth weight, not showing increased risk. Low birth weight is caused either by preterm birth or by stunted growth for gestational age, or a combination of both. Being small for gestational age can be due to intrauterine growth restriction secondary to many possible factors [61]. There is a lack of data in the original studies as for the causes of low birth weight, and more studies will be needed to draw firm conclusions.

The available studies, none of which is randomized, performed over half a century should increase one's vigilance for potential sources of bias. Only about half of the included studies controlled for maternal age and/or other confounders such as nicotine and alcohol consumption, previous abortions, diabetes, gravidity and/or parity [3, 7, 16, 25, 37-40, 43, 44, 46, 48, 50, 52, 54, 55]. However, their results are not distinctively different from those which controlled for these

variables. Two included studies measured H1-AHS use during the first trimester as prescriptions filled, without any assurance that the pregnant women actually took the medications [33, 42]. Eleven studies were conducted in teratology information services [16, 38-40, 43, 44-52]. This can potentially introduce a selection bias as there is evidence demonstrating that pregnant women of low socioeconomic class do not use such services with the same frequency as women of higher socioeconomic status and thus the study population may not be generalizable [62]. Data collected from different studies may be subjected to bias as different studies had different reporting standards and different classifications of the outcome. However, each included study utilized the same standards for its cases and control groups, so that the estimated risk in each study may not be affected. There are also sources of potential bias by indication. NVP has been shown to confer more favorable pregnancy outcome. As shown above, we have addressed this potential source of bias by analyzing separately only the studies where AH were given for morning sickness, and this analysis does not suggest an apparent bias in overall malformation rates. Similarly, AH are sometimes used for the allergic component of asthma among women, and here a bias against the null hypothesis may be created by less favorable pregnancy outcome in asthmatic women [63]. However, available studies did not specify asthma as a diagnosis and hence this potential source of bias cannot be ruled out.

6.5. Conclusion.

In conclusion, based on our updated meta-analyses, with very large sample sizes, H1-AHs do not appear to be associated with an increased risk of major malformation or other adverse fetal outcomes. Despite methodological limitations and potential sources of unresolved bias, this study may provide important information to both pregnant women and their health care providers regarding the

safety of H1-AH use during early pregnancy. At the present time, these meta-analyses cannot rule out an increased risk for specific congenital malformations.

Compliance with ethical standards: This systematic review of published manuscripts meets the criteria for ethical standards.

Funding: No sources of funding were used to assist in the preparation of this study

Conflict of interest: Fatma Etwel, Lauren H Faught, Michael J Rieder and Gideon Koren have no conflicts of interest that are directly relevant to the content of this study.

6.6. References.

1. Stephansson O, Granath F, Svensson T, Haglund B, Ekbom A, Kieler H. Drug use during pregnancy in Sweden - assessed by the Prescribed Drug Register and the Medical Birth Register. Clin Epidemiol. 2011;3:43-50.

2. Werler MM, Mitchell AA, Hernandez-Diaz S, Honein MA. Use of over the-counter medications during pregnancy. Am J Obstet Gynecol. 2005;193:771–777.

3. Li Q, Mitchell AA, Werler MM, Yau WP, Hernández-Díaz S. Assessment of antihistamine use in early pregnancy and birth defects. J Allergy Clin Immunol Pract. 2013;1(6):666-74.

4. Woolhouse M. Complementary medicine for pregnancy complications. Aust Fam Physician. 2006;35(9):695.

5. Bousquet J, Godard P, Michel FB. Antihistamines in the treatment of asthma. Eur Respir J. 1992;5(9):1137-42.

6. Soll AH, Walsh JH. Regulation of gastric acid secretion. Annu Rev Physiol. 1979;41:35-53.

7. Aldridge TD, Hartmann KE, Michels KA, Velez Edwards DR. First-trimester antihistamine exposure and risk of spontaneous abortion or preterm birth. Pharmacoepidemiol Drug Saf. 2014;23(10):1043-50.

8. Shields KE, Wiholm BE, Hostelley LS, Striano LF, Arena SR, Sharrar RG. Monitoring outcomes of pregnancy following drug exposure: a company-based pregnancy registry program. Drug Saf. 2004;27:353-67.

9. Etwel F, Hutson JR, Madadi P, Gareri J, Koren G. Fetal and perinatal exposure to drugs and chemicals: novel biomarkers of risk. Annu Rev Pharmacol Toxicol. 2014;54:295-315.

10. Seto A, Einarson T, Koren G. Pregnancy outcome following first trimester exposure to antihistamines: meta-analysis. Am J Perinatol. 1997;14(3):119-24.

11. Chin JW, Gregor S, Persaud N. Re-analysis of safety data supporting doxylamine use for nausea and vomiting of pregnancy. Am J Perinatol. 2014 Sep;31(8):701-10.

12. McKeigue PM, Lamm SH, Linn S, Kutcher JS. Bendectin and birth defects: I. A meta-analysis of the epidemiologic studies. Teratology. 1994;50(1):27-37.

13. MacMahon B. More on Bendectin. JAMA. 1981;246(4):371-2.

14. Einarson TR, Leeder JS, Koren G. A method for meta-analysis of epidemiological studies. Drug Intell Clin Pharm. 1988 Oct;22(10):813-24.

15. Schwarz EB, Moretti ME, Nayak S, Koren G. Risk of hypospadias in offspring of women using loratadine during pregnancy: a systematic review and meta analysis. Drug Saf. 2008;31(9):775-88.

16. Etwel F, Djokanovic N, Moretti ME, Boskovic R, Martinovic J, Koren G. The fetal safety of cetirizine: an observational cohort study and meta-analysis. JObstet Gynaecol. 2014;34(5):392-9.

17. Moher D, Liberati A, Tetzlaff J, Altman DG; PRISMA Group. Reprint—preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. Phys Ther. 2009 Sep;89(9):873-80.

18. Rasmussen SA, Olney RS, Holmes LB, Lin AE, Keppler-Noreuil KM, Moore CA; National Birth Defects Prevention Study. Guidelines for case classification for the National Birth Defects Prevention Study. Birth Defects Res A Clin Mol Teratol. 2003 Mar;67(3):193-201.

19. De Neubourg D, van Duijnhoven NT, Nelen WL, D'Hooghe TM. Dutch translation of the ICMART-WHO revised glossary on ART terminology. Gynecol Obstet Invest. 2012;74(3):233-48.

20. Higgins JPT, Green S (editors). Cochrane Handbook for Systematic Reviews of Interventions Version 5.1.0 [updated March 2011]. The Cochrane Collaboration, 2011. Available from www.cochrane-handbook.org.

21. Duval S, Tweedie R. Trim and fill: A simple funnel-plot-based method of testing and adjusting for publication bias in meta-analysis. Biometrics. 2000;56(2):455-63.

22. Stang A. Critical evaluation of the Newcastle-Ottawa scale for the assessment of the quality of nonrandomized studies in meta-analyses. Eur J Epidemiol. 2010 Sep;25(9):603-5.

23. General Practitioner Research Group. General Practitioner Clinical Trials: DRUGS IN PREGNANCY SURVEY. Practitioner 1963;191:775-780.

24. Mellin G, Katzenstein M. Meclozine and fetal abnormalities. Lancet 1963;26:222-223.

25. Bunde CA, Bowles DM. A technique for controlled survey of case records. Curr Ther Res Clin Exp. 1963;5:245-248.

26. Yerushalmy J, Milkovich L. Evaluation of the teratogenic effect of meclizine in man. Am J Obstet Gynecol. 1965;93(4):553-62.

27. Erez S, Schifrin BS, Dirim O. Double-blind evaluation of hydroxyzine as an antiemetic in pregnancy. J Reprod Med. 1971;7(1):35-7.

28. Milkovich L,vandenBerg BJ. An evaluation of the teratogenicity of certain antinauseant drugs. Am J Obstet Gynecol. 1976;125(2): 244-248.

29. Kullander S, Källén B. A prospective study of drugs and pregnancy. II. Anti-emetic drugs. Acta Obstet Gynecol Scand. 1976;55(2):105-11.

30. Heinonen OP, Slone D, Shapiro S. Birth defects and drugs in pregnancy. Littleton MA: PSG Publishing; 1977.

31. Newman NM, Correy JF, Dudgeon GI. A survey of congenital abnormalities and drugs in a private practice. Aust N Z J Gynaecol. 1977;17:156-159.

32. Shapiro S, Kaufman DW, Rosenberg L, Slone D, Monson RR, Siskind V, Heinonen OP. Meclizine in pregnancy in relation to congenital malformations. Br Med J. 1978;1(6111):483.

33. Smithells RW, Sheppard S. Teratogenicity testing in humans: a method demonstrating safety of bendectin. Teratology. 1978; 17(1):3-135.

34. Jick H, Holmes LB, Hunter JR, Madsen S, Stergachis A. First trimester drug use and congenital disorders. JAMA. 1981;246(4): 343-346.

35. Fleming DM, Knox JDE,Crombie DL. Debendox in early pregnancy and fetal malformation. Br Med J (Clin Res Ed). 1981;283(6284):99-101.

36. Gibson GT, Colley DP, McMichael AJ, Hartshorne JM. Congenital anomalies in relation to the use of doxylamine/dicyclomine and other antenatal factors: an ongoing prospective study. Med J Aust. 1981;1(8):410-414.

37. Michaelis J, Michaelis H, Glück E, Koller S. Prospective study of suspected associations between certain drugs administered during early pregnancy and congenital malformations. Teratology. 1983;27(1):57-64.

38. Seto A, Einarson T, Koren G. Evaluation of brompheniramine safety in pregnancy. Reprod Toxicol. 1993;7(4):393-5.

39. Pastuszak A, Schick B, D'Alimonte D, Donnenfeld A, Koren G. The safety of astemizole in pregnancy. J Allergy Clin Immunol. 1996;98(4):748-50.

40. Einarson A, Bailey B, Jung G, Spizzirri D, Baillie M, Koren G. Prospective controlled study of hydroxyzine and cetirizine in pregnancy. Ann Allergy Asthma Immunol. 1997;78(2):183-6.

41. Schatz M, Petitti D. Antihistamines and pregnancy. Ann Allergy Asthma Immunol. 1997;78(2):157-9.

42. Wilton LV, Pearce GL, Martin RM, Mackay FJ, Mann RD. The outcomes of pregnancy in women exposed to newly marketed drugs in general practice in England. Br J Obstet Gynaecol. 1998;105(8):882-9.

43. Loebstein R, Lalkin A, Addis A, Costa A, Lalkin I, Bonati M, Koren G. Pregnancy outcome after gestational exposure to terfenadine: A multicenter, prospective controlled study. J Allergy Clin Immunol. 1999;104(5):953-6.

44. Källén B. Use of antihistamine drugs in early pregnancy and delivery outcome. J Matern Fetal Neonatal Med. 2002;11(3):146-52.

45. Källén B, Mottet I. Delivery outcome after the use of meclozine in early pregnancy. Eur J Epidemiol. 2003;18(7):665-9.

46. Bsat FA, Hoffman DE, Seubert DE. Comparison of three outpatient regimens in the management of nausea and vomiting in pregnancy. J Perinatol. 2003;23(7):531-5.

47. Diav-Citrin O, Shechtman S, Aharonovich A, Moerman L, Arnon J, Wajnberg R, Ornoy A. Pregnancy outcome after gestational exposure to loratadine or antihistamines: a prospective controlled cohort study. J Allergy Clin Immunol. 2003;111(6):1239-43.

48. Moretti ME, Caprara D, Coutinho CJ, Bar-Oz B, Berkovitch M, Addis A, Jovanovski E, Schüler-Faccini L, Koren G. Fetal safety of loratadine use in the first trimester of pregnancy: a multicenter study. J Allergy Clin Immunol. 2003;111(3):479-83.

49. Paulus W, Schloemp S, Sterzik K, Stoz F. Pregnancy outcome after exposure to cetirizine/levocetirizine in the first trimester-a prospective controlled study. Reprod Toxicol. 2004;19 (2):258 [Abstract].

50. Boskovic R, Rudic N, Danieliewska-Nikiel B, Navioz Y, Koren G. Is lack of morning sickness teratogenic? A prospective controlled study. Birth Defects Res A Clin Mol Teratol. 2004;70(8):528-30.

51. Weber-Schoendorfer C, Schaefer C. The safety of cetirizine during pregnancy. A prospective observational cohort study. Reprod Toxicol. 2008;26(1):19-23.

52. Ashkenazi-Hoffnung L, Merlob P, Stahl B, Klinger G. Evaluation of the efficacy and safety of bi-daily combination therapy with pyridoxine and doxylamine for nausea and vomiting of pregnancy. Isr Med Assoc J. 2013;15(1):23-6.

53. Eskenazi B, Bracken MB. Bendectin (Debendox) as a risk factor for pyloric stenosis. Am J Obstet Gynecol. 1982;144(8):919-24.

54. Czeizel AE, Vargha P. A case-control study of congenital abnormality and dimenhydrinate usage during pregnancy. Arch Gynecol Obstet. 2005;271(2):113-8.

55. Gilboa SM, Strickland MJ, Olshan AF, Werler MM, Correa A; National Birth Defects Prevention Study. Use of antihistamine medications during early pregnancy and isolated major malformations. Birth Defects Res A Clin Mol Teratol. 2009;85(2):137-50.

56. Petik D, Acs N, Bánhidy F, Czeizel AE. A study of the potential teratogenic effect of large doses of promethazine used for a suicide attempt by 32 pregnant women. Toxicol Ind Health. 2008;24(1-2):87-96.

57- Little BB, Snell LM, Breckenridge JD, Knoll KA, Klein VR, Gilstrap LC. Effects of T's and blues abuse on pregnancy outcome and infant health status. Am J Perinatol. 1990;7(4):359 62.

58. Chasnoff IJ, Hatcher R, Burns WJ, Schnoll SH. Pentazocine and tripelennamine ('T's and blue's'): effects on the fetus and neonate. Dev Pharmacol Ther. 1983;6(3):162-9.

59. Koren G, Madjunkova S, Maltepe C. The protective effects of nausea and vomiting of pregnancy against adverse fetal outcome--a systematic review. Reprod Toxicol. 2014;47:77-80.

60. Matsuyama K, Ichikawa T, Nitta Y, Ikoma Y, Ishimura K, Horio S, Fukui H. Localized expression of histamine H1 receptors in syncytiotrophoblast cells of human placenta. J Pharmacol Sci. 2006;102(3):331-7.

61. Zohdi V, Sutherland MR, Lim K, Gubhaju L, Zimanyi MA, Black MJ. Low birth weight due to intrauterine growth restriction and/or preterm birth: effects on nephron number and long-term renal health. Int J Nephrol. 2012;2012:136942.

62. Ornoy A, Mastroiacovo P. More on data from teratogen information systems (TIS). Teratology. 2000 May;61(5):327-8.

63. Liu S, Wen SW, Demissie K, Marcoux S, Kramer MS. Maternal asthma and pregnancy outcomes: a retrospective cohort study. Am J Obstet Gynecol. 2001 Jan;184(2):90-6.

Chapter 7: When original positive studies of novel therapies are subsequently nullified; cumulative meta-analyses in preeclampsia.

This chapter has been published previously:

Etwel F, Koren G. When positive studies of novel therapies are subsequently nullified: cumulative meta-analyses in preeclampsia. Clin Invest Med. 2015 Oct 7;38(5):E274-83.

7.1. Introduction.

Typically, after laboratory and experimental animal investigations, novel therapeutic modalities are introduced to humans through case reports and small non-randomized, prospective studies. These are subsequently followed by randomized, double blinded, placebo controlled trials (RCTs), which eventually allow researchers to define whether the modality is sufficiently effective and safe over an existing gold standard. The lag time between the first published RCT and a decision by the medical and scientific communities to accept or reject a new modality, can be relatively long. During this period, scientific communications through editorials, commentaries, letters to editors and lectures, are vehicles that may convince clinicians to use or not to use the new treatment.

In the area of maternal-fetal medicine, new therapeutic options are few and far between [1]. The ethical challenge of exposing a developing fetus to drugs can cause delays and hesitations among clinicians and drug companies alike. Yet, not treating serious maternal conditions can also adversely affects the unborn child and puts the pregnant patient at risk of morbidity and mortality[2].

Usually, when clinician-investigators complete an RCT of a novel modality, they attempt to publish the results in high citation impact journals, as these assure wide professional and public dissemination, in addition to increased likelihood of future grant funding and professional promotion [3]. However, in more than a few cases, the first, high impact factor publications suggesting a significantly favorable effect had been followed by negative trials.

The objective of the present study was to examine changes over time in the pooled effect size of RCT published on the protective effects of antioxidants and

low dose aspirin against preeclampsia, a common and serious obstetric complication [4], and identify determinants that may affect these changes.

7.2. Methods.

We used two recently published meta-analyses of RCTs examining the protective effects of antioxidant treatment and those of low dose aspirin against preeclampsia [5-6]. The two selected meta-analyses were subjected to methodological quality assessment using the AMSTAR method [7]. The assessment of multiple systematic reviews' (AMSTAR) is a tool containing an 11 point questionnaire with each point having four possible answers. The AMSTAR quality assessment tool falls into three ranges, High (9-11), Medium (5-8), and Low (0-4) [8]. The quality assessment was applied to the studies in the original meta-analyses to ensure that the studies selected were of good quality and their analyses included appropriate inclusion and exclusion criteria to avoid clinical and methodological heterogeneity and to control for internal validity.

In both meta-analyses, some papers were “positive” in terms of protective effect (defined by us as RR below 0.9) (RR of 0.9 suggests a 10% protective effect which was considered by us a reasonable minimum) and some were negative (RR equal or above 0.9). The overall result of the antioxidant meta-analysis was “no protective effect” (negative). In the meta-analysis of low dose aspirin, the overall result was marginally protective. These two meta-analyses were subjected by us to cumulative meta-analysis and correlation studies.

7.2.1. Cumulative meta-analysis.

We conducted cumulative meta-analyses of the selected meta-analyses (without conducting new meta-analyses and adding new studies) to detect a possible time-dependent effect. The cumulative analysis route displays results

accumulated over time: that is, the second row presents a summary analysis comprising the first two studies; the third row presents a summary analysis comprising the first three studies, and so on through the final row. When the data are arranged by year of publication this shows the effect measure (relative risk) that could have been achieved at any point in time with each new study's arrival; furthermore, the changes in the final conclusion can be examined over time. For the cumulative meta-analyses the Comprehensive Meta-Analysis Version 2.0 (Biostat, Engelwood NJ) was used. Heterogeneity among studies was assessed using the Q statistic and the I-squared test. If the Q statistic (P value) is <0.05, it represents a high degree of variance among the studies analyzed, and the results are quantified by I-squared values. I-squared between 25%-50% signifies low heterogeneity, between 50%-75% moderate heterogeneity, and >75% signifies high heterogeneity [9]. Publication bias was analyzed using Funnel plots for detecting the presence of gray literature and assessing its impact on the analysis. The number of unpublished studies (K) that were possibly absent from our analysis was determined; if evidence of publication bias occurred (K>0), then adjusted point estimates (RR) were calculated based on the number of omitted studies (K) using the Duval and Tweedie's trim-and-fill method.

7.2.2. Correlation studies.

We correlated the journal's impact factor, citation number of each paper by using Google scholar during May 2015 and their sample size, with the RR of the study's primary results. In the case of antioxidants, we also correlated the journal's impact factor with the quality of the paper, by using the Cochrane Collaboration method [10]. This method demonstrates which articles satisfied all quality assessment criteria (no risk of bias) and which articles have not satisfied all quality assessment criteria. Comparison of continuous variables was conducted by the

Mann Whitney U test, and correlations between variables were calculated by the Spearman method.

7.3. Results.

The two meta-analyses included in this review were subjected to quality assessment, where the total AMSTAR scores were high (where both studies had maximum scores of 11). This suggests that the studies included in the meta-analysis are not subjected to clinical and methodological heterogeneity that may affect the validity of the overall results. The conducted cumulative meta-analysis did not show statistically significant heterogeneity since all the *P* values of the Q tests were less than 0.05 and the I-square were less than 50%.

7.3.1. Antioxidant studies.

The first RCT included in the antioxidant meta-analysis was published in 1994 and the most recent one was published in 2011. The sample sizes of the 15 included studies ranged between 60-9969 subjects, the journals' citation impact factor ranged between 0.60-54.42, and the number of citations of each paper between 13- 857 (between 2.17/year and 53.56/year) (Table 7.1).

The median sample size of the positive trials (median 267) was tenfold smaller than the sample size of the negative trials (median 2120) (*P*=0.017). There was a significant positive correlation between study size and RR (rho=0.74; *P* =0.0016). There was no significant correlation between RR and citation number (rho=0.239), or between RR and the journal's impact factor (rho=0.332). In contrast, the journal's impact factor significantly correlated with the number of citations per year (rho=0.82; *P* =0.00016). Overall, the impact factor of the journal did not correlate with the quality of the papers as measured by Biberio- Salle et al [5] by using the Cochrane Collaboration method. Three studies that fulfilled all

quality criteria (25, 26, 30), with no risk of bias, had a RR of more than 0.9 (0.97, 1.20 and 1.03), showing no protective effect of antioxidant on pre-eclampsia.

During the first 12 years, in 5 studies, the cumulative meta-analysis revealed that there was a seeming significant protective effect of antioxidant vs. placebo on the rates of preeclampsia. This effect gradually diminished and was nullified by larger studies by 2006 (Figure 7.1). After the analysis of the publication bias using the funnel plot, the analysis detected five missing studies (Figure 7.5) and after incorporating the studies in the analysis the overall results shows more non protective effect of the antioxidant (RR=0.91 in the original meta-analysis, vs. RR= 1.01 adjusted meta-analysis) (Table 7.3).

7.3.2. Low dose aspirin studies.

The first RCT included in the low dose aspirin meta-analysis was published in 1986 and the most recent one was published in 2012. The sample sizes of the 14 included studies ranged between 44 and 8257 subjects, the journals' citation impact factor ranged between 1.41 -54.42, and the number of citations of each paper was between 9-482 (0.43/year-16.62/year) (Table 7.2).

The median sample size of the positive trials measuring IUGR (median 72) was 15 fold smaller than the sample size of the negative trials (median 3019) (P=0.006). Similar trends were seen for preterm delivery and rates of preeclampsia (Table7.2). There was a trend toward significant positive correlation between study size and RR for IUGR (rho=; P =0.06). There was no correlation between RR and citation number, or between RR and the journal's impact factor. In contrast, the journal's impact factor significantly correlated with the number of citations per year (rho=0.55; P =0.05).

The cumulative meta-analysis revealed that during the first 8-11 years, there was a significant protective effect of low dose aspirin vs. placebo on the rates of IUGR, prematurity and preeclampsia. This effect gradually diminished and was either nullified or remained marginally significant which was caused by larger studies starting in 2006 (Figures7.2- 7.4). After the analysis of the publication bias using the funnel plot, the analysis detected five to six missing studies and after incorporating the studies in the three meta-analyses the overall results showed marginally more significance. (from RR=0.77 in the original preeclampsia meta-analysis to RR= 0.83 in the adjusted meta-analysis; from RR=0.86 in the original preterm birth meta-analysis to RR= 0.98 in the adjusted meta-analysis; and from RR=0.80 in the original IUGR meta-analysis to RR=1.93 in the adjusted meta-analysis) (Table 7.3).

Table 7.1. Details of the RCTs included in the meta-analysis of antioxidants for preventing preeclampsia.

Study Name	Year	Sample size	RR	Journal name (impact factor)	Number of citation	Citation per year
Han [20]	1994	100	0.10	Chinese Med J (2.34)	46	2.19
Chappell [21]	1999	283	0.46	Lancet (39.20)	857	53.56
Sharma [22]	2003	100	0.48	Int J Gynecol Obstet (1.56)	68	5.67
Steyn [23]	2003	200	1.00	J Obstet Gynaecol (0.60)	53	4.42
Beazley [24]	2005	251	0.92	Am J Obstet Gynecol (3.97)	137	13.70
Poston [25]	2006	1877	0.97	Lancet (39.20)	57	6.33
Rumbold [26]	2006	2395	1.20	NEJM (54.42)	398	44.22
Rumiris [27]	2006	60	0.24	Hypertens Preg (1.19)	80	8.89
Spinnato [28]	2007	159	0.88	Obstet Gynecol (4.80)	105	13.13
Banerjee [29]	2009	1355	0.99	J Obstet Gynecol Res (0.90)	13	2.17
Villar [30]	2009	707	1.03	BJOG (1.56)	113	18.83
McCance [31]	2010	2363	0.81	Lancet (39.20)	55	11.00
Roberts [32]	2010	749	1.07	NEJM (54.40)	201	40.20
Xu [33]	2010	9969	1.04	Am J Obstet Gynecol (3.97)	89	17.80
Vadillo [34]	2011	444	0.75	BMJ (16.30)	80	20.00

Table 7.2. Details of the RCTs included in the meta-analysis of low dose aspirin for preeclampsia.

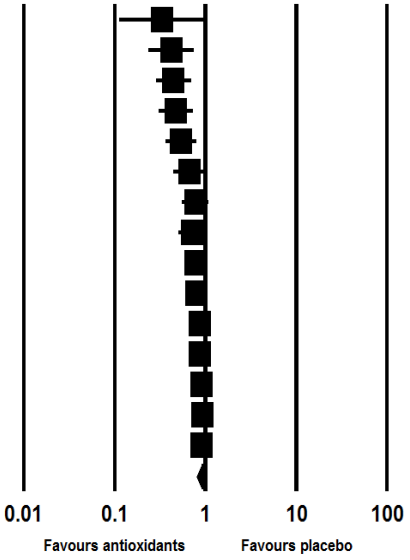
Study name	Year	RR (sample size)			Journal name (impact factor)	Number of citations	Citation per year
		Preeclampsia	Preterm birth	IUGR			
Wallenburg [35]	1986	0.07 (44)	0.12 (44)	0.73 (44)	Lancet (39.21)	482	16.62
Benigni [36]	1989	-	0.38 (33)	0.31 (33)	NEJM (54.42)	249	9.58
Schiff [37]	1989	0.13 (65)	0.31 (66)	0.30 (65)	NEJM (54.42)	314	12.08
McParland [38]	1990	0.11 (100)	-	1.08 (100)	Lancet (39.21)	160	6.40
Vinnika [39]	1993	0.84 (197)	-	0.46 (197)	BJOG (3.86)	61	2.77
Caspi [40]	1994	0.19 (47)	0.75 (47)	0.52 (94)	Am J Reprod Immunol (2.67)	9	0.43
CLASP [41]	1994	0.88 (7974)	0.90 (7974)	0.90 (8257)	Lancet (39.21)	72	3.43
Hermida [42]	1997	0.43 (100)	0.20 (100)	0.50 (100)	Hypertension (7.63)	49	2.72
Gallery [43]	1997	-	0.65(108)	-	Hypertension Pregnancy (1.41)	20	1.11
MFMU [44]	1998	0.90 (2503)	0.93 (2503)	1.19 (2503)	NEJM (54.42)	467	27.47
Grab [45]	2000	1.43 (43)	-	-	Ultrasound Obstet Gynnecol (3.85)	26	1.73
Vainio [46]	2002	0.20 (86)	-	0.33 (86)	BJOG (3.86)	151	11.62
Yu [47]	2003	0.95(554)	0.90 (554)	0.90 (554)	Ultrasound Obstet Gynnecol (3.85)	100	8.33
Villa [48]	2012	0.72(121)	-	0.33 (121)	BJOG (3.86)	49	16.33
Ayala [49]	2012	0.94(350)	0.38 (350)	0.49 (350)	Chronobiol Int (2.88)	26	8.67

Table 7.3. Duval and Tweedie's Trim and Fill summary data for analysis of publication bias in the two meta-analyses.

			Random Effects Model			
		Studies Trimmed	Point Esti-mate	Lower Limit	Upper Limit	Q-Value
Anti-Oxidant & Preeclampsia	**Observed Values**		0.91	0.80	1.03	23.55
	Adjusted Values	5	1.01	0.94	1.16	46.05
Low Dose Aspirin & Preeclampsia	**Observed Values**		0.77	0.62	0.95	19.95
	Adjusted Values	5	0.83	0·64	1.07	35.61
Low Dose Aspirin & Preterm Birth	**Observed Values**		0.86	0.76	0.98	13.37
	Adjusted Values	5	0·89	0.76	1.05	25.76
Low Dose Aspirin & IUGR	**Observed Values**		0.80	0.65	0.99	19.00
	Adjusted Values	6	1.93	0.74	1.18	33.97

Figure 7.1. Cumulative chronological meta-analysis of risk ratio in RCTs investigating the effectiveness of antioxidants for preeclampsia.

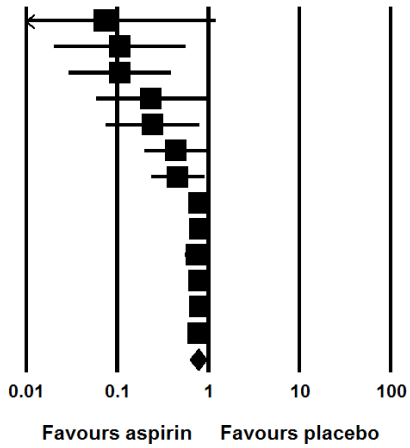
Study name (Year)	Cumulative n	Cumulative statistics			Cumulative risk ratio (95% CI)	Relative weight
		Point	Lower limit	Upper limit		
Han (1994)	100	0.34	0.11	0.98		1.28
Chappell (1999)	383	0.42	0.24	0.74		4.33
Sharma (2003)	632	0.44	0.29	0.69		7.14
Steyn (2003)	834	0.47	0.31	0.72		7.76
Beazley (2005)	934	0.54	0.37	0.79		9.80
Poston (2006)	3329	0.67	0.44	1.00		23.99
Rumbold (2006)	5206	0.77	0.56	1.07		31.22
Rumiris (2006)	5266	0.72	0.51	1.01		31.98
Spinnato (2007)	5973	0.77	0.59	1.01		39.85
Banerjee (2009)	6132	0.80	0.63	1.02		42.92
Villar (2009)	7487	0.86	0.71	1.05		56.98
McCance (2010)	8236	0.86	0.73	1.02		65.86
Roberts (2010)	18205	0.91	0.79	1.05		82.33
Xu (2010)	20568	0.93	0.81	1.05		91.04
Vadillo (2011)	21012	0.91	0.80	1.03		100.00
		0.91	0.80	1.03		



Test of heterogeneity: df = 14 (P = 0.049) ; I-squered = 41.071%

Figure 7.2. Cumulative chronological meta-analysis of preeclampsia of RCTs of women at risk of preeclampsia who took either aspirin or placebo.

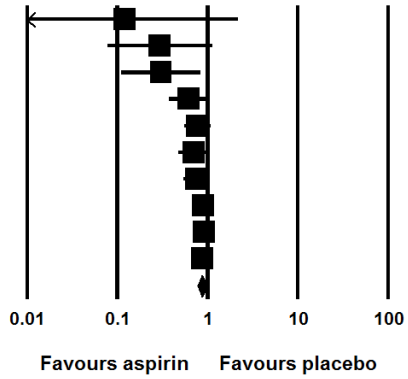
Study	Cumulative statistics					Cumulative risk ratio (95% CI)	Relative weight
	Point	Lower limit	Upper limit	Aspirin	Placebo		
Wallenburg 1986	0.07	0.00	1.20	0 / 21	7 / 23		0.56
Schiff 1989	0.11	0.02	0.55	1 / 55	14 / 54		1.59
McParland 1990	0.11	0.03	0.38	2 / 103	24 / 106		2.65
Viinikka 1993	0.23	0.06	0.93	11 / 200	35 / 206		7.99
Caspi 1994	0.24	0.08	0.79	11 / 224	37 / 229		8.48
CLASP 1994	0.44	0.20	0.96	278 / 4216	339 / 4211		36.49
Hermida 1997	0.46	0.24	0.90	281 / 4266	346 / 4261		38.94
MFMU 1998	0.78	0.61	1.02	507 / 5520	596 / 5510		66.77
Grab 2000	0.80	0.63	1.02	510 / 5542	598 / 5531		68.26
Vainio 2002	0.74	0.56	0.98	512 / 5585	608 / 5574		70.22
Yu 2003	0.80	0.64	1.00	561 / 5861	660 / 5852		87.44
Villa 2012	0.80	0.65	0.99	569 / 5922	671 / 5912		92.76
Ayala 2012	0.77	0.62	0.95	580 / 6098	693 / 6086		100.00
	0.77	0.62	0.95				



Test of heterogeneity: df = 12 (P = 0.068) ; I-squered = 39.841%

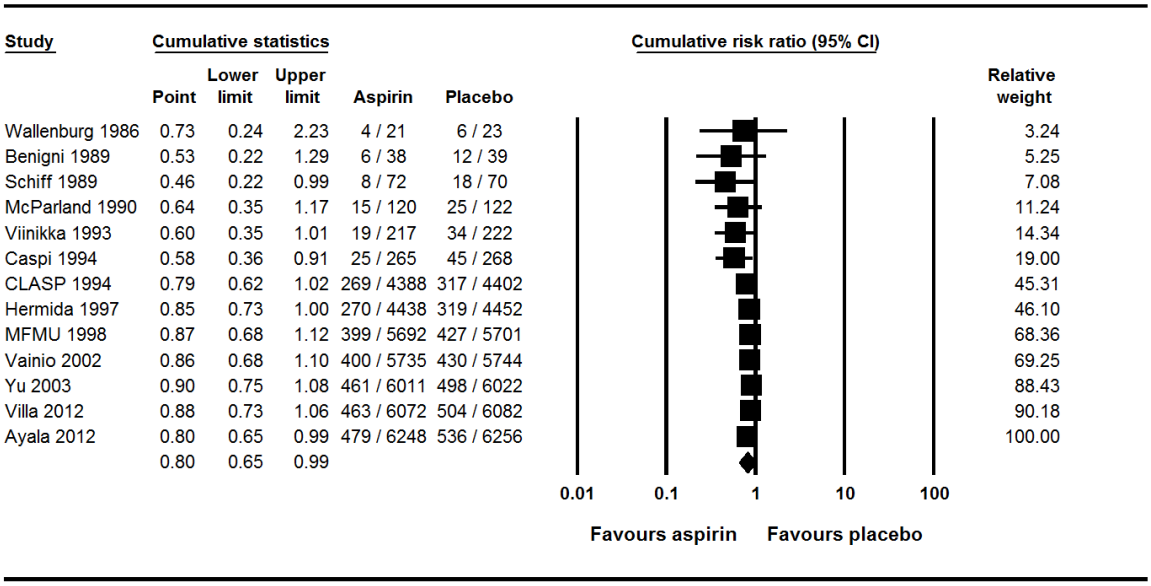
Figure 7.3. Cumulative chronological meta-analysis of preterm birth of RCTs of women at risk of preeclampsia who took either aspirin or placebo.

Study	Cumulative statistics					Cumulative risk ratio (95% CI)	Relative weight
	Point	Lower limit	Upper limit	Aspirin	Placebo		
Wallenburg 1986	0.12	0.01	2.12	0 / 21	4 / 23		0.18
Benigni 1989	0.30	0.08	1.11	2 / 38	9 / 39		0.85
Schiff 1989	0.30	0.11	0.82	4 / 72	15 / 71		1.49
Caspi 1994	0.61	0.38	0.99	15 / 96	29 / 94		6.08
CLASP 1994	0.77	0.55	1.07	701 / 4088	790 / 4076		44.32
Hermida 1997	0.71	0.48	1.04	702 / 4138	795 / 4126		44.66
Gallery 1997	0.74	0.55	1.01	708 / 4196	803 / 4176		46.15
MFMU 1998	0.89	0.81	0.98	1210 / 5450	1340 / 5425		84.44
Yu 2003	0.90	0.84	0.97	1277 / 5726	1415 / 5703		97.93
Ayala 2012	0.86	0.76	0.98	1284 / 5902	1435 / 5877		100.00
	0.86	0.76	0.98				



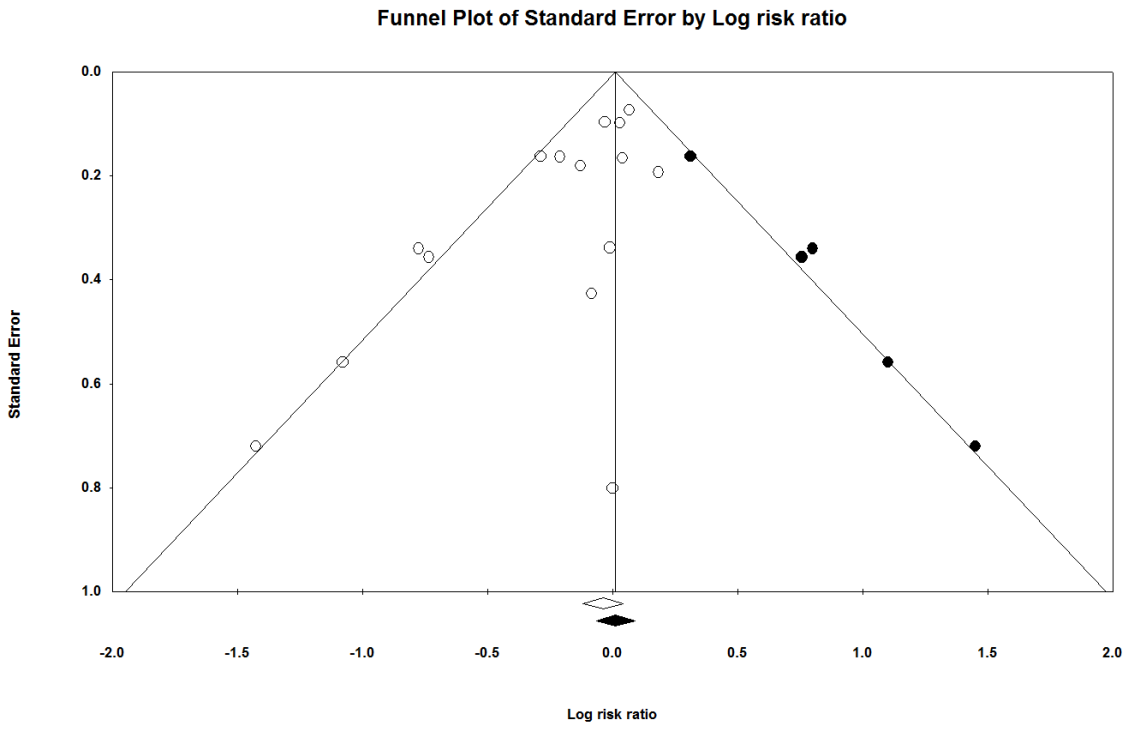
Test of heterogeneity: df = 9 (P = 0.146) ; I-squered = 32.676%

Figure 7.4. Cumulative chronological meta-analysis of IUGR sorted by the year of publication for trials of women at risk of preeclampsia who took either aspirin or placebo.



Test of heterogeneity: df = 12 (P = 0.089) ; I-squered = 36.830%

Figure 7.5. Publication bias using funnel plot of preeclampsia prevention by antioxidant meta-analysis (open circles are the original studies and closed circles are the added studies during adjustment).


Funnel Plot of Standard Error by Log risk ratio
Standard Error
0.0
0.2
0.4
0.6
0.8
1.0
-2.0
-1.5
-1.0
-0.5
0.0
0.5
1.0
1.5
2.0
Log risk ratio

7.4. Discussion.

Studies published in high impact factor journals are cited significantly more. Naturally, this leads physicians to encounter them more often and possibly assume that these studies reflect the right clinical answer. Similar to our findings, a recent study has shown that the impact factor predicts 59% of the variation in citations of systematic reviews. However, the distribution of citations was obviously skewed [11]. Importantly, our study shows that high impact factor journals do not exhibit higher likelihood of predicting a correct answer, which implies that the higher citation impact does not translate into more correct clinical impact. The fact that they are cited substantially more may thus create a reporting bias. For example, the second published study by Chappel et al. on antioxidants [21] showed a 54 percent reduction in risk of preeclampsia with antioxidants and exhibited the highest number of citations to date (n=857), which may impact clinicians' decision in considering patient care. Yet, this study did not predict correctly the overall effect of antioxidants, which is null.

What are the reasons that first studies often declare a dramatic effect only to be nullified later? Our study offers several plausible explanations:

Firstly, as shown by our results, studies with smaller sample sizes are more likely to be biased against the null hypothesis and suggest positive results. Small studies are easier to execute and bring to completion, so naturally they are more likely to be published first. As major journals are seeking novel discoveries, they may be more inclined to accept such papers, and hence, to be exposed to the risk of bias against the null hypothesis.

Secondly, there is a serious and systematic bias against the null hypothesis in the publication process [12, 13]. We and others have shown that negative studies

(i.e. those not showing a significant effect) are less likely to be submitted by their authors [14], less likely to be accepted for scientific presentations or publication by journals [13, 15], less likely to be quoted by the lay media, [16] and less likely to be cited in the peer review literature [17]. When these effects are combined, a serious bias can be produced, potentially creating a spurious positive effect. As years go by, larger studies are published, and the slow-to-be-accepted negative papers manage to find their way to PubMed –Medline [18], hence the original spurious positive effect may be diluted and finally nullified. This is further shown by the assessment of publication bias, suggesting that there are five to six studies that have not been published and after incorporating the missing studies the adjusted overall effect shows further nullification of the signal.

With respect to Impact Factor, it has been and continues to be the leading journal quality indicator despite recognized weaknesses such as the effects of self-citation, review articles, the total number of articles published, and English language bias [3]. Related to the issues addressed in the present study, a recent evaluation of 13 trauma journals concluded that the impact factor of a journal was a poor measure of the clinical relevance of its papers. Specifically, the authors found that high impact journals did not address clinical research in surgery and when they did, there was a delay before such papers were cited [18].

In the case of preeclampsia, the biological plausibility of either antioxidants or low dose aspirin in experimental models has led to great therapeutic hopes, which can probably explain the enthusiasm that accompanied the first positive studies. With both antioxidants and low dose aspirin, the cumulative chronological meta-analyses revealed that during the first years, there was a seeming significant protective effect on the rates of preeclampsia and its complications. With both modalities, the suggested protective effects gradually disappeared and were either

nullified or remained marginally significant by larger, later studies. Our study highlights the importance of the cumulative meta-analysis as a powerful evaluation tool, which has been used more and more often to decide when additional RCTs are no longer needed therefore suggesting an effect has been proven beyond reasonable doubt [19].

The fact that initial papers in high impact journals did not predict better the clinical utility of antioxidants or low dose aspirin for preeclampsia is consistent with the finding that there were no consistent differences in quality between high impact journals vs. papers in less prestigious journals. This highlights one of the criticisms against the citation impact factor, as it may be informative about the overall quality of the journal, but not of specific papers [11].

7.5. Conclusion.

In conclusion, initial studies, often published in high citation impact factor journals, are cited significantly more times, but do not exhibit higher likelihood of predicting a correct answer. Studies with smaller sample sizes are more likely to be biased against the null hypothesis and as such, cumulative meta-analysis is an effective tool to predict potential bias against the null hypothesis.

7.6. References.

1. Koren G. Treating the mother, protecting the unborn: the motherisk approach. Pediatr Pharmacol Ther. 2013 Jan;18(1):4-7.

2. Lyerly AD, Faden RR.Mothers Matter: Ethics and Research during Pregnancy. Virtual Mentor. 2013 Sep 1;15(9):775-8.

3. Kianifar H, Sadeghi R, Zarifmahmoudi L. Comparison Between Impact Factor, Eigenfactor Metrics, and SCimago Journal Rank Indicator of Pediatric Neurology Journals. Acta Inform Med. 2014 Apr;22(2):103-6.

4. Gordin D, Forsblom C, Groop PH, Teramo K, Kaaja R. Risk factors of hypertensive pregnancies in women with diabetes and the influence on their future life. Ann Med. 2014 Jul 21:1-5.

5. Salles AM, Galvao TF, Silva MT, Motta LC, Pereira MG. Antioxidants for preventing preeclampsia: a systematic review. ScientificWorldJournal. 2012;2012:243476.

6. Henderson JT, Whitlock EP, O'Connor E, Senger CA, Thompson JH, Rowland MG. Low-dose aspirin for prevention of morbidity and mortality from preeclampsia: a systematic evidence review for the U.S. Preventive Services Task Force. Ann Intern Med. 2014 May 20;160(10):695-703.

7. Shea BJ, Grimshaw JM, Wells GA, Boers M, Andersson N, Hamel C, Porter AC, Tugwell P, Moher D, Bouter LM. Development of AMSTAR: a measurement tool to assess the methodological quality of systematic reviews. BMC Med Res Methodol. 2007 Feb 15;7:10.

8. Sequeira-Byron P, Fedorowicz Z, Jagannath VA, Sharif MO. An AMSTAR assessment of the methodological quality of systematic reviews of oral healthcare interventions published in the Journal of Applied Oral Science (JAOS). J Appl Oral Sci. 2011 Oct;19(5):440-7.

9. Higgins JP, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. BMJ. 2003; 327(7414):557-60.

10. J. Higgins and S. Green, Eds., Cochrane Handbook for Systematic Reviews of Interventions, Version 5.1.0 edition, 2011, http://www.cochrane-handbook.org.

11. Royle P, Kandala NB, Barnard K, Waugh NSyst Rev. Bibliometrics of systematic reviews: analysis of citation rates and journal impact factors. Sys Rev.2013 Sep 12;2:74.

12. Koren G. Bias against the null hypothesis in maternal-fetal pharmacology and toxicology. Clin Pharmacol Ther. 1997 Jul;62(1):1-5.

13. Koren G, Graham K, Shear H, Einarson T. Bias against the null hypothesis: the reproductive hazards of cocaine. Lancet. 1989 Dec 16;2(8677):1440-2.

14. Easterbrook PJ, Berlin JA, Gopalan R, Matthews DR. Publication bias in clinical research. Lancet. 1991 Apr 13;337(8746):867-72.

15. Jannot AS, Agoritsas T, Gayet-Ageron A, Perneger TV. Citation bias favoring statistically significant studies was present in medical research. J Clin Epidemiol. 2013 Mar;66(3):296-301.

16. Koren G, Klein N. Bias against negative studies in newspaper reports of medical research. JAMA. 1991 Oct 2;266(13):1824-6.

17. Koren G, Nickel C.Perpetuating fears: bias against the null hypothesis in fetal safety of drugs as expressed in scientific citations. J Popul Ther Clin Pharmacol. 2011;18(1):e28-32.

18. Kodumuri P, Ollivere B, Holley J, Moran CG. The impact factor of a journal is a poor measure of the clinical relevance of its papers. Bone Joint J. 2014 Mar;96-B(3):414-9.

19. Lau J, Antman EM, Jimenez-Silva J, Kupelnick B, Mosteller F, Chalmers TC. Cumulative meta-analysis of therapeutic trials for myocardial infarction. N Engl J Med. 1992 Jul 23;327(4):248-54.

20. Han L, Zhou SM. Selenium supplement in the prevention of pregnancy induced hypertension. Chin Med J (Engl). 1994 Nov;107(11):870-1.

21. Chappell LC, Seed PT, Briley AL, Kelly FJ, Lee R, Hunt BJ, Parmar K, Bewley SJ, Shennan AH, Steer PJ, Poston L. Effect of antioxidants on the occurrence of pre-eclampsia in women at increased risk: a randomised trial.Lancet. 1999 Sep 4;354(9181):810-6.

22. Sharma JB, Kumar A, Kumar A, Malhotra M, Arora R, Prasad S, Batra S. Effect of lycopene on pre-eclampsia and intra-uterine growth retardation in primigravidas. Int J Gynaecol Obstet. 2003 Jun;81(3):257-62.

23. Steyn PS, Odendaal HJ, Schoeman J, Stander C, Fanie N, Grové D. A randomised, double-blind placebo-controlled trial of ascorbic acid supplementation for the prevention of preterm labour. J Obstet Gynaecol. 2003 Mar;23(2):150-5.

24. Beazley D, Ahokas R, Livingston J, Griggs M, Sibai BM. Vitamin C and E supplementation in women at high risk for preeclampsia: a double-blind, placebo controlled trial. Am J Obstet Gynecol. 2005 Feb;192(2):520-1.

25. Poston L, Briley AL, Seed PT, Kelly FJ, Shennan AH; Vitamins in Pre-eclampsia (VIP) Trial Consortium. Vitamin C and vitamin E in pregnant women at risk for pre-eclampsia (VIP trial): randomised placebo-controlled trial. Lancet. 2006 Apr 8;367(9517):1145-54.

26. Rumbold AR, Crowther CA, Haslam RR, Dekker GA, Robinson JS; ACTS Study Group. Vitamins C and E and the risks of preeclampsia and perinatal complications. N Engl J Med. 2006 Apr 27;354(17):1796-806.

27. Rumiris D, Purwosunu Y, Wibowo N, Farina A, Sekizawa A. Lower rate of preeclampsia after antioxidant supplementation in pregnant women with low antioxidant status. Hypertens Pregnancy. 2006;25(3):241-53.

28. Spinnato JA 2nd, Freire S, Pinto E Silva JL, Cunha Rudge MV, Martins-Costa S, Koch MA, Goco N, Santos Cde B, Cecatti JG, Costa R, Ramos JG, Moss N, Sibai BM. Antioxidant therapy to prevent preeclampsia: a randomized controlled trial. Obstet Gynecol. 2007 Dec;110(6):1311-8.

29. Banerjee S, Jeyaseelan S, Guleria R. Trial of lycopene to prevent pre-eclampsia in healthy primigravidas: results show some adverse effects. J Obstet Gynaecol Res. 2009 Jun;35(3):477-82.

30. Villar J, Purwar M, Merialdi M, Zavaleta N, Thi Nhu Ngoc N, Anthony J, De Greeff A, Poston L, Shennan A; WHO Vitamin C and Vitamin E trial group. World Health Organisation multicentre randomised trial of supplementation with vitamins C and E among pregnant women at high risk for pre-eclampsia in populations of low nutritional status from developing countries. BJOG. 2009 May;116(6):780-8..

31. McCance DR, Holmes VA, Maresh MJ, Patterson CC, Walker JD, Pearson DW, Young IS; Diabetes and Pre-eclampsia Intervention Trial (DAPIT) Study Group. Vitamins C and E for prevention of pre-eclampsia in women with type 1 diabetes (DAPIT): a randomised placebo-controlled trial. Lancet. 2010 Jul 24;376(9737):259-66.

32. Roberts JM, Myatt L, Spong CY, Thom EA, Hauth JC, Leveno KJ, Pearson GD, Wapner RJ, Varner MW, Thorp JM Jr, Mercer BM, Peaceman AM, Ramin SM, Carpenter MW, Samuels P, Sciscione A, Harper M, Smith WJ, Saade G, Sorokin Y, Anderson GB; Eunice Kennedy Shriver National Institute of Child Health and Human Development Maternal-Fetal Medicine Units Network. Vitamins C and E to prevent complications of pregnancy-associated hypertension. N Engl J Med. 2010 Apr 8;362(14):1282-91.

33. Xu H, Perez-Cuevas R, Xiong X, Reyes H, Roy C, Julien P, Smith G, von Dadelszen P, Leduc L, Audibert F, Moutquin JM, Piedboeuf B, Shatenstein B, Parra-Cabrera S, Choquette P, Winsor S, Wood S, Benjamin A, Walker M, Helewa M, Dubé J, Tawagi G, Seaward G, Ohlsson A, Magee LA, Olatunbosun F, Gratton

R, Shear R, Demianczuk N, Collet JP, Wei S, Fraser WD; INTAPP study group. An international trial of antioxidants in the prevention of preeclampsia (INTAPP). Am J Obstet Gynecol. 2010 Mar;202(3):239.e1-239.

34. Vadillo-Ortega F, Perichart-Perera O, Espino S, Avila-Vergara MA, Ibarra I, Ahued R, Godines M, Parry S, Macones G, Strauss JF. Effect of supplementation during pregnancy with L-arginine and antioxidant vitamins in medical food on pre-eclampsia in high risk population: randomised controlled trial. BMJ. 2011 May 19;342:d2901.

35. Wallenburg HC, Dekker GA, Makovitz JW, Rotmans P. Low-dose aspirin prevents pregnancy-induced hypertension and pre-eclampsia in angiotensin-sensitive primigravidae. Lancet. 1986 Jan 4;1(8471):1-3.

36. Benigni A1, Gregorini G, Frusca T, Chiabrando C, Ballerini S, Valcamonico A, Orisio S, Piccinelli A, Pinciroli V, Fanelli R, et al. Effect of low-dose aspirin on fetal and maternal generation of thromboxane by platelets in women at risk for pregnancy-induced hypertension. N Engl J Med. 1989 Aug 10;321(6):357-62.

37. Schiff E, Peleg E, Goldenberg M, Rosenthal T, Ruppin E, Tamarkin M, Barkai G, Ben-Baruch G, Yahal I, Blankstein J, et al. The use of aspirin to prevent pregnancy-induced hypertension and lower the ratio of thromboxane A2 to prostacyclin in relatively high risk pregnancies. N Engl J Med. 1989 Aug 10;321(6):351-6.

38. McParland P, Pearce JM, Chamberlain GV. Doppler ultrasound and aspirin in recognition and prevention of pregnancy-induced hypertension. Lancet. 1990 Jun 30;335(8705):1552-5.

39. Viinikka L1, Hartikainen-Sorri AL, Lumme R, Hiilesmaa V, Ylikorkala O. Low dose aspirin in hypertensive pregnant women: effect on pregnancy outcome and prostacyclin-thromboxane balance in mother and newborn. Br J Obstet Gynaecol. 1993 Sep;100(9):809-15.

40. Caspi E1, Raziel A, Sherman D, Arieli S, Bukovski I, Weinraub Z. Prevention of pregnancy-induced hypertension in twins by early administration of low-dose aspirin: a preliminary report. Am J Reprod Immunol. 1994 Jan;31(1):19-24.

41. CLASP: a randomised trial of low-dose aspirin for the prevention and treatment of pre-eclampsia among 9364 pregnant women. CLASP (Collaborative Low-dose Aspirin Study in Pregnancy) Collaborative Group. Lancet. 1994 Mar 12;343(8898):619-29.

42. Hermida RC, Ayala DE, Iglesias M, Mojón A, Silva I, Ucieda R, Fernández JR. Time-dependent effects of low-dose aspirin administration on blood pressure in pregnant women. Hypertension. 1997 Sep;30(3 Pt 2):589-95.

43. Gallery ED, Ross MR, Hawkins M, et al. Low-dose aspirin in high-risk pregnancy? Hypertension Pregnancy. 1997;16(2):229-38.

44. Caritis S, Sibai B, Hauth J, Lindheimer MD, Klebanoff M, Thom E, VanDorsten P, Landon M, Paul R, Miodovnik M, Meis P, Thurnau G. Low-dose aspirin to prevent preeclampsia in women at high risk. National Institute of Child Health and Human Development Network of Maternal-Fetal Medicine Units. N Engl J Med. 1998 Mar 12;338(11):701-5.

45. Grab D, Paulus WE, Erdmann M, Terinde R, Oberhoffer R, Lang D, Muche R, Kreienberg R. Effects of low-dose aspirin on uterine and fetal blood flow during pregnancy: results of a randomized, placebo-controlled, double-blind trial. Ultrasound Obstet Gynecol. 2000 Jan;15(1):19-27.

46. Vainio M, Kujansuu E, Iso-Mustajärvi M, Mäenpää J. Low dose acetylsalicylic acid in prevention of pregnancy-induced hypertension and intrauterine growth retardation in women with bilateral uterine artery notches. BJOG. 2002 Feb;109(2):161-7.

47. Yu CK1, Papageorghiou AT, Parra M, Palma Dias R, Nicolaides KH; Fetal Medicine Foundation Second Trimester Screening Group. Randomized controlled trial using low-dose aspirin in the prevention of pre-eclampsia in women with abnormal uterine artery Doppler at 23 weeks' gestation. Ultrasound Obstet Gynecol. 2003 Sep;22(3):233-9.

48. Villa PM, Kajantie E, Räikkönen K, Pesonen AK, Hämäläinen E, Vainio M, Taipale P, Laivuori H; PREDO Study group. Aspirin in the prevention of pre-eclampsia in high-risk women: a randomised placebo-controlled PREDO Trial and a meta-analysis of randomised trials. BJOG. 2013 Jan;120(1):64-74.

49. Ayala DE1, Ucieda R, Hermida RC. Chronotherapy with low-dose aspirin for prevention of complications in pregnancy. . Chronobiol Int. 2013 Mar;30(1-2):260-79.

Chapter 8: General discussion.

8.1. Discussion of research findings.

Although nonrandomized observational studies are of lower quality than randomized studies, nonrandomized studies have some advantages, as they are conducted over a long period of time with the number of participants increasing cumulatively [1]. The almost total lack of randomized control studies of drug therapy during pregnancy makes observational studies the main source of evidence in teratology, where data can be synthesized from multiple large prescription databases. This has led to a more recent form of cohort publications based on large numbers of patients and exposures, albeit retrospective in nature. These studies' strengths lie in their large size, but the problem is that the larger cohort studies will take many years to accumulate and the rate of association of new drugs used by pregnant women and abnormalities will still need to be estimated in the meantime. In contrast, small cohort studies conducted by different countries and research centers will be published years before the large cohort studies can finish collecting their data. The meta-analysis of small nonrandomized cohort studies from different sources, but with the same teratogenic questions, can give us early estimates about drug use during pregnancy, and one can reassure pregnant women who have taken the drug before they knew they are pregnant or pregnant women who need to use a certain drug are acting on reliable information.

The current thesis shows, with sufficient evidence how similar are the conclusions based on the meta-analysis of small studies and those from very large cohort studies in estimating teratogenicity [2]. This is obvious, especially with untreated epilepsy, proton pump inhibitors and H2 blockers being safe, while valproic acid may cause malformations.

This study enables researchers, clinicians, drug companies and regulators to trust the meta-analyses of small underpowered controlled studies in identifying the signals of new drugs used during pregnancy earlier in the course of marketing.

Retrospective registries can be useful as we estimate, based on the available data, that there is a 4.5 fold amplification factor in retrospective reports compared to prospective studies of the same compounds by the same company for risk of congenital malformations from drug exposure during pregnancy [3]. If the retrospective registries lead to stable increases in teratogenic signal, this must also mean that, if the retrospective cohort does not show malformation rates above the expected baseline of 3-5%, the drug is probably not associated with increased teratogenic risk.

Meta-analysis of observational studies is an effective tool to answer unresolved research questions in teratology. Meta-analysis has an advantage that gives more statistical power as similar results from different studies on the same research question are combined. Meta-analysis also allows a more truthful representation of different populations than is delivered by the specific study estimators.

The use of meta-analysis can generate new hypotheses and insight on drug safety during pregnancy, such as the case of H1 antihistamine in treatment of NVP. 18 of our 33 cohort studies looked specifically at H1 antihistamine use for the treatment of NVP and were included in our analysis. The results showed no association between H1 antihistamine use as an antiemetic and major malformations, which is consistent with previous studies. Yet, with an odds ratio (0.95) and lower limit of the confidence interval (0.87) less than one, our results also may suggest a trend towards a protective effect of antihistamines [4].

However, several studies have shown that NVP itself has a protective effect on the unborn baby, with mothers who experience morning sickness having better birth outcomes including reduced risks of spontaneous abortion, preterm birth, birth defects, and children with higher IQ [5]. There are two well-accepted theories of how NVP may play a role in better pregnancy outcomes. The first is that the presence of NVP in pregnancy may prevent the ingestion of harmful teratogens, either through lack of appetite or physical sickness, therefore protecting the unborn fetus [6]. The second theory suggests that NVP is secondary to high hormone levels associated with viable pregnancies, and therefore the symptoms themselves are not protective, merely they are a negative side effect. Based on the latter theory rather than being protective, NVP could then be an indication of a more optimal pregnancy, which would result in better outcomes [7]. However one particular negative effect of NVP can be the inability to obtain adequate nutrition as a result of emesis or lack of desire to eat. Women who are treated for their NVP will not suffer the consequences of inadequate nutrition. It is therefore reasonable to assume that those women who suffer from NVP (i.e. have optimal hormone levels for the best outcomes) and are able to obtain optimal nutrition (because they used antiemetic treatment), which could explain the protective effects of antihistamines both reported by us and other studies. However, more research studies, with carefully planned control groups, are required in order to definitively answer questions surrounding these possible protective effects previously observed.

This study provides important information to both pregnant women and their health care providers regarding the safety and risk of H1 antihistamines use during this sensitive time. Although our conclusions are based on a large number of studies, many others exist which also address fetal safety after exposure to H1 antihistamines. These particular studies suffer from methodological issues that

made data extraction for meta-analysis difficult and/or impossible. Much of this is the result of issues presented when studying the safety of medication in pregnancy. However, more observational controlled studies, with more consistent methodology are necessary in order to best assess the safety and risks associated with not only H1-AH use in pregnancy, but all medication in general.

Cumulative meta-analysis in drug therapy is an essential tool used in predicting when the direction of the conclusion starts to change, from effective to no effect may even cross to negative effect, after introducing more trials. The problem is that the primary studies that discover the new remedy usually suffer from limited sample size. These studies are often published in high impact factor journals and are highly cited causing the medical community to trust their preliminary results. The cumulative meta-analysis of RCTs published on the protective effects of antioxidants and low dose aspirin against preeclampsia clearly shows the potential bias against the null hypothesis [8]. This study should encourage researchers who perform new trials on drug therapy to conduct cumulative meta-analysis of all previous trials to give the medical community more insight of the direction of the overall results.

8.2. Methodological challenges in observational studies included in meta-analyses.

Although many observational control studies so far have addressed fetal safety after exposure to specific chemicals, they suffer from methodological issues including lack of important information such that performing meta-analyses is challenging. Therefore, there is a need for clear guidelines for researchers who perform observational studies to help other researchers in including them in meta-analyses to increase the sample size and reduce clinical heterogeneity that is often

present in the exposed groups, control groups (comparison group) and in pregnancy outcomes measures.

8.2.1. Limitations in the exposed group.

The limitations in the exposed group are that the gestational age and duration, daily dose and the drug indication are not defined or reported consistently. In addition, the exposed group should optimally be free of conditions that are known to cause adverse pregnancy outcomes, except for the disease that is being treated by the drug under study; however, many studies fail to practice and/or report such exclusion.

8.2.2. Limitations in the control group.

The comparison (or control) group must be composed of healthy volunteers who have not been exposed to the specific drug under study and/or the same pharmacological class of the drug under study. Ideally they must not have been exposed to chemicals that may cause unwanted fetal effects. In addition, the control group must be free of any disease that may affect pregnancy outcome. The control group and the exposed group should optimally be matched for maternal age and, if possible, for other confounders such as nicotine and alcohol consumption, diabetes, gravidity, parity, previous abortions and/or malformations in previous pregnancies.

The above conditions are not followed and/or reported by some studies. In some studies, a disease matched control group is needed because the control group has the same disease as the exposed group but does not take medications, the control group is treated for the condition with another class of drugs, or the control group takes the medication under study but not during the first trimester of pregnancy.

8.2.3. Limitations in the pregnancy outcomes.

Regarding malformation, birth defects should be classified as major or minor and the pediatrician who follows up on a case should confirm the severity of the malformation. When reporting the major malformation, it is important that researchers not just give the number of each kind of malformation because sometimes one case has more than one kind of malformation and this may cause a multiple counting of malformations; instead, researchers should also indicate the number of exposed subjects and how many experienced major malformations. Because some research studies did not follow these guidelines, this caused exclusion of the studies from our meta-analysis.

Most of the studies examined the association between prenatal exposure to drugs and major birth defects and they did not study other adverse pregnancy outcomes, such as spontaneous abortions, prematurity, stillbirth, infant death, perinatal death, therapeutic abortions, elective abortion, low birth weight, abnormal head-circumference and abnormal Apgar score. These outcomes are equally important to address teratogenic potential.

The reasons behind in utero death may be due to birth defects or due to other causes reasons; however, these incidents are inconsistently reported. Information on abortions should clarify if they were spontaneous, therapeutic or elective.

8.2.4. Limitations in general.

All the important raw data, such as pregnancy numbers, live births, the gestational ages at birth, the number of premature births, rates of spontaneous abortions and all deaths; should be reported, but some studies did not report all necessary information. Moreover, often updated studies included old data

published in the past without referencing it, and this could have caused double counting or an overlap when performing meta-analysis.

It is evident that in case control studies the cases and controls are selected based on the presence of congenital anomalies. Sometimes we are missing important data necessary to conduct meta-analysis.

Case control Studies are retrospective and observational. We first identify the group that has the health outcome of interest (malformations, which in case control study are cases). Then we identify a group that did not have the health outcome of interest (malformations), which in case control study are controls. Then we determine whether or not the participants from each group had a particular exposure to H1 antihistamine in the past. Table 8 provides the information needed to conduct the meta-analysis for the cohort study.

In the case control group, the following information is provided:

1- Number of cases (as defined by the case control study that has the outcome) which are equal to A+C.

2- Number of controls (as defined by case control study that has no outcome) which are equal to B+D.

Some case control studies did not provide all the necessary information needed, such as A or C and B or D. This caused an exclusion of these studies from meta-analysis that may provide important evidence of the fetal safety/risk of the drug under study.

Table 8. Information needed to conduct the meta-analysis for cohort study.

	Malformation cases	Total number
Exposed group	A	A+B
Control group	C	C+D

A: Malformation cases that are exposed to the drug under study.

B: Normal cases that are exposed to the drug under study.

C: Malformation cases that are not exposed to the drug under study.

D: Normal cases that are not exposed to the drug under study.

8.2.5 Limitations due to study bias.

Study bias is an error that leads to false positive or negative estimation of the risk. Bias can have different sources from study design to the process of publication [9]. There is always a challenge to decrease the sources of bias and increase the generalizability of the data. But, due to the limited data on drug fetal safety in the literature and lack of RCTs, observational studies have to be included in meta-analyses even though they did not always control for potential bias. This is one of the big limitations of meta-analyses conducted in this field.

Bias by indication is one of these limitations, where the disease itself that is intended to be treated by the drug under study, may cause or protect from the unwanted pregnancy outcome. This bias can be overcome by proper study design by having a disease- matched control and if one has large pool of studies, one can conduct a sensitivity analysis that includes only observational studies where confounders were adequately addressed in the study design and analysis.

Maternal fetal research deals with observational studies where the population is usually pregnant women who voluntarily chose to use the drug under study or accidently took the drug before they knew that they were pregnant. The population under study may include women with health insurance, good education and/or previous unwanted pregnancy outcome, who will seek help from the medical community regarding drug safety during pregnancy [10]. A selection bias may then be generated and the data may not be easily generalized. This limitation must be acknowledged in the original studies by reporting the demographic characteristics of the included population and proper matching for variables such as previous adverse pregnancy outcome between the exposed and the control groups.

There is always a chance that the meta-analysis missed some studies because of publication bias, where studies with significant results are more likely to be published in the literature than studies with results that are not statistically significant. Generating a funnel plot is a common way to address publication bias but if the number of included studies is less than ten then this method cannot be used due to lack of sensitivity [11]. Missing studies are not always due to publication bias. There are other sources that may lead to missing studies such as language bias where non-English studies are more likely to be missed, citation bias where studies with non-significant results are less cited by other papers and more likely to be missed, and availability bias where studies with keywords that did not match the search terms are more likely to be missed [12].

These limitations cannot be avoided most of the time since the original data were collected by different groups of researchers with different criteria. Generating widely acceptable guidelines for observational studies on specific topics on fetal drug safety can help limit possible biases and may help achieve more precise estimates of risk/safety of fetal drug exposure during pregnancy.

8.3. Conclusion.

The following points are the general conclusions of the included studies in this thesis:

1- Meta-analysis of smaller studies appears to generate correct signal in estimating human teratogenicity years before large and methodologically superior cohort studies are published [2].

2- The present study confirms a major and consistent bias against the null hypothesis in retrospective registry studies which needs to be considered when interpreting such data. Spontaneous reporting is highly selective toward adverse

events, as families with normal pregnancy outcomes are less likely to report them [3].

3- Overall, cetirizine is not associated with a clinically important increase in risk of major malformations or other adverse fetal outcomes. Confounding of results due to use of the drug for asthma must be considered [13].

4- Based on our meta-analyses, which include a large number of studies, H1 antihistamines are not associated with an increased risk of major malformation or other adverse fetal outcomes. This study provides important information to both pregnant women and their health care providers regarding the safety and risk of H1 antihistamine use during this sensitive time [4].

5- Initial intervention studies, often published in high impact factor journals, are cited significantly more times but do not exhibit a higher likelihood of predicting a correct long term answer. As such, cumulative meta-analysis is an effective tool in predicting potential bias against the null hypothesis and the need for additional studies [8].

8.4. Area of future research.

In the area of therapeutics in pregnancy, we need to continue to study the safety and efficacy of drugs specific to this population. Conducting small cohort studies in different parts of the world and combining them in meta-analyses is an effective tool for this purpose. Furthermore, conducting cumulative meta-analyses is important whenever new studies emerge in an effort to continue to examine the safety and efficacy of therapies in pregnancy, or deciding that sufficient data already exists.

8.5. References.

1) Bluhm R. Some observations on observational research. Perspect Biol Med 2009; 52:252-263.

2) Etwel F, Hutson JR, Madadi P, Gareri J, Koren G. Fetal and perinatal exposure to drugs and chemicals: novel biomarkers of risk. Annu Rev Pharmacol Toxicol. 2014;54:295-315.

3) Etwel F, Koren G. Bias against the Null Hypothesis in Retrospective Registries of Gestational Drug Exposure. J Obstet Gynaecol Can. 2016.

4) Etwel F, Faught LH, Rieder MJ, Koren G. The risk of adverse pregnancy outcome after first trimester exposure to H1 antihistamines: a systematic review and meta-analysis. Drug Safety 2016.

5) Koren G, Madjunkova S, Maltepe C. The protective effects of nausea and vomiting of pregnancy against adverse fetal outcome--a systematic review. Reprod Toxicol. 2014;47:77-80.

6) Profet M. Pregnancy sickness as adaptation: a deterrent to maternal ingestion of teratogens. In:Barkow JH, Cosmides L, Tooby J, editors. The adapted mind: evolutionary psychology and the generation of culture. New York: Oxford University

7) Forbes S. Pregnancy sickness and parent-offspring conflict over thyroid function. J Theor Biol. 2014 Aug 21;355:61-7.

8) Etwel F, Koren G. When positive studies of novel therapies are subsequently nullified: cumulative meta-analyses in preeclampsia. Clin Invest Med. 2015 Oct 7;38(5):E274-83.

9) Gerhard T. Bias: considerations for research practice. Am J Health Syst Pharm. 2008 Nov 15;65(22):2159-68.

10) Ornoy A, Mastroiacovo P. More on data from teratogen information systems (TIS). Teratology. 2000 May;61(5):327-8.

11) Sterne JA, Gavaghan D, Egger M. Publication and related bias in meta-analysis: power of statistical tests and prevalence in the literature. J Clin Epidemiol.2000 Nov;53(11):1119-29.

12) Borenstein M, Hedges LV, Higgins JPT, Rothstein HR. Introduction to Meta-Analysis. John Wiley & Sons, Ltd, Chichester, UK 2009.

13) Etwel F, Djokanovic N, Moretti ME, Boskovic R, Martinovic J, Koren G. The fetal safety of cetirizine: an observational cohort study and meta-analysis. J Obstet Gynaecol. 2014 Jul;34(5):392-9.

Appendices

Appendix 1: Copyright approval for previously published work.

1) Copyright policies for Annual Reviews.

Copyright Clearance Center RightsLink®

Home | Create Account | Help | Live Chat

ANNUAL REVIEWS

Title: Fetal and Perinatal Exposure to Drugs and Chemicals: Novel Biomarkers of Risk

Author: Fatma Etwel, Janine R. Hutson, Parvaz Madadi, et al

Publication: Annual Review of Pharmacology and Toxicology

Publisher: Annual Reviews

Date: Jan 6, 2014

Copyright © 2014, Annual Reviews

LOGIN

If you're a copyright.com user, you can login to RightsLink using your copyright.com credentials. Already **a RightsLink user** or want to learn more?

Permission Not Required

Material may be republished in a thesis / dissertation without obtaining additional permission from Annual Reviews, providing that the author and the original source of publication are fully acknowledged.

BACK | CLOSE WINDOW

Copyright © 2016 Copyright Clearance Center, Inc. All Rights Reserved. Privacy statement. Terms and Conditions.
Comments? We would like to hear from you. E-mail us at customercare@copyright.com

2) Permission for re-publishing from the journal of Obstetrics and Gynaecology Canada.

Your manuscript JOGC_2016_146_R1 has been accepted

Editor

Mon 9/12, 1:55 PMFatma Etwel

Dear Ms Etwel:

Your manuscript is scheduled for publication in the December issue of JOGC and may be considered "in press". You should receive edited versions for approval in the next 10 days.

The manuscript may be cited as in press with JOGC. Once you have approved the pdf version that the publisher will send you, the manuscript will have an assigned doi for citation.

With kind regards.

Yours sincerely,

Timothy Rowe
Editor-in-Chief, Journal of Obstetrics and Gynaecology Canada
www.jogc.com

3) Permission for re-publishing from the journal of Obstetrics and Gynaecology.

Our Ref: DE/IJOG/P6663

04 March 2016

Dear Fatma Etwel,

Thank you for your correspondence requesting permission to reproduce the following article published in our journal in your printed thesis and to be posted in your university's repository.

F. Etwel, N. Djokanovic, M. E. Moretti, R. Boskovic, J. Martinovic & G. Koren (2014) The fetal safety of cetirizine: An observational cohort study and meta-analysis, Journal of Obstetrics and Gynaecology, 34:5, 392-399

We will be pleased to grant permission on the sole condition that you acknowledge the original source of publication and insert a reference to the article on the Journals website:

This is the authors accepted manuscript of an article published as the version of record in Journal of Obstetrics and Gynaecology 28 Mar 2014 http://www.tandfonline.com/ http://tandfonline.com/doi/full/10.3109/01443615.2014.896887

Please note that this license does not allow you to post our content on any third party websites or repositories.

Thank you for your interest in our Journal.

Yours sincerely

Debbie East– Permissions & Licence Administrator - Journals. Routledge, Taylor & Francis Group.

4) Permission for re-publishing from the journal Drug Safety.

RE: DRSA-D-16-00222R1 -

Joshi, Nitin, Springer

Thu 11/3/2016 7:01 PM

To: Fatma Etwel

Hi Fatma

If your thesis won't be available on a public domain, you can use the accepted version of your manuscript, but not the final publisher's version, in your thesis. The final version is the one that we will publish on our Website. You can use the word document that was finally accepted.

Please let me know if you need more information.

Best Regards

Nitin

5) Permission for re-publishing from the journal Clinical and Investigative Medicine.

March 3, 2016

To Whom it may concern

Fatma Etwel, PhD Candidate in Physiology and Pharmacology at Western University, has asked for permission to cite the following paper in her PhD thesis dissertation:

Etwel F, Koren G. When positive studies of novel therapies are subsequently nullified: cumulative meta-analyses in preeclampsia. Clin Invest Med. 2015 Oct 7;38(5):E274-83.

As Editor in Chief of Clinical Investigative Medicine and owner of the copyright, I grant permission for Fatma Etwel to cite this paper as part of her PhD thesis dissertation.

Sincerely,

Robert Bortolussi

Professor Emeritus Dalhousie University

Editor in Chief, Clinical and Investigative Medicine

"Opportunity is missed by most people, because it is dressed in overalls and looks like work."
Thomas Edison

Appendix 2: Detailed characteristics of the included examples for Chapter 3.

1) Benzodiazepines and risk of major malformations:

In 1998 a meta-analysis of benzodiazepine and major congenital malformation concluded that benzodiazepine were not associated with risk of major malformation (odds ratio 0.90; 95% confidence interval 0.61 to 1.35). This meta-analysis combined 7 small cohort studies (n < 335 in each study). The last published study was in 1997 [1].

In 2007 a Swedish group published a large cohort study from Swedish Medical Birth Register. The total number of group exposed to benzodiazepine was 1929. The final conclusion was the same as the meta-analysis (odds ratio 1.12; 95% confidence interval 0.91 to 1.36) [2]. An updated meta-analysis published in 2011 combined the old meta-analysis with two other cohort studies including the large one and the final conclusion did not change (odds ratio 1.07; 95% confidence interval 0.91 to 1.25) [3].

2) Untreated epilepsy and major malformations:

In 2004 a meta-analysis of the association between untreated epilepsy and major malformation was published. The conclusion was that the risk for congenital malformation in the offspring of women with epilepsy who had not used antiepileptic was not higher than among nonepileptic controls (odds ratio 1.92; 95% confidence interval 0.92 to 4.00). This meta-analysis combined 10 small cohort studies (n < 99 in each study). The last published study was in 2001 [4].

In 2009 a group from Norway published a large cohort study from The Medical Birth Register of Norway. The total number of children of women with untreated epilepsy was 1900. The final conclusion was similar to that of meta-analysis (odds ratio 1.0; 95% confidence interval 0.8 to 1.4) [5].

3) Proton Pump Inhibitors and major malformation:

In 2002 a meta-analysis of proton pump Inhibitors and major malformation was published with the final conclusion that proton pump Inhibitors are not associated with increased risk of major malformation (Relative Risk 1.18; 95% confidence interval 0.72 to 1.94). This meta-analysis combined 5 small cohort studies (n< 276 in each study). The last published study was in 2001 [6]. An updated meta-analysis published in 2009 combined the old meta-analysis with two more recent cohort studies (one study in 2005 and the other one in 2008), the new

meta-analysis gave the same final conclusion (odds ratio 1.12; 95% confidence interval 0.86 to 1.45) [7].

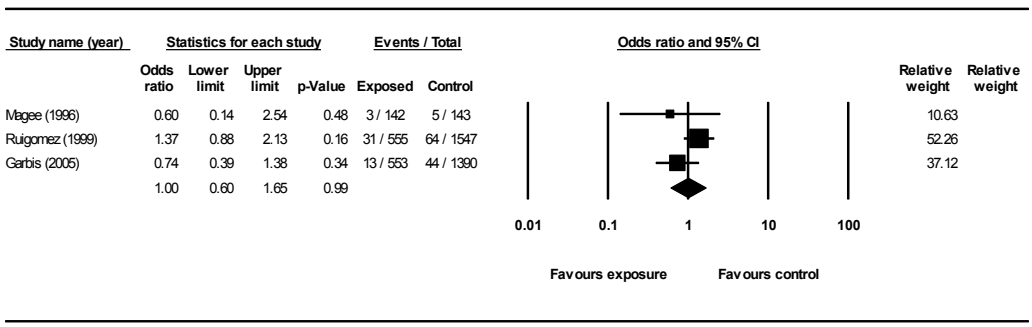
In 2010 a group from Denmark published a large cohort study using data from The Medical Birth Register, the Prescription Drug Register, the National Patient Register, the Center Person Register, and Statistics Denmark. The total number in the group exposed to proton pump Inhibitors was 3651. The final conclusion was comparable to the meta-analysis (odds ratio 1.10; 95% confidence interval 0.91 to 1.34) [8].

4) Histamine 2 (H2) blocker and major malformations:

In 2009 a meta-analysis of H2 blocker and major malformation was published [9]. This meta-analysis combined four cohort studies, three of them small, and the fourth, which was the most recent one consisted of a large cohort study that at that time was published as an abstract in 2008 [10] and the full study published in 2010 [11]. The big study was removed from the meta-analysis by us and the remaining three small cohort studies were recombined using comprehensive meta-analysis version 2 software. The meta-analysis, after removing the large cohort study showed no association with the malformation (odds ratio 1.00; 95% confidence interval 0.60 to 1.65) and the most recent study in the reconstructed meta-analysis was in 2005 (Figure 3).

In 2010 a group from Israel published a large cohort study; using data from Clalit Health Services in Israel, [11]. The total number of exposed group to H2 blocker was 1148. The final conclusion was the same as the reconstructed meta-analysis (odds ratio 1.14; 95% confidence interval 0.89 to 1.45).

Figure 3. Forest plot of the incidence of congenital malformations after in utero exposure to H2 blockers (without the large cohort study).


Study name (year)
Statistics for each study
Events / Total
Odds ratio and 95% CI
Odds ratio
Lower limit
Upper limit
p-Value
Exposed
Control
Relative weight
Relative weight
Magee (1996) 0.60 0.14 2.54 0.48 3 / 142 5 / 143 10.63
Ruigomez (1999) 1.37 0.88 2.13 0.16 31 / 555 64 / 1547 52.26
Garbis (2005) 0.74 0.39 1.38 0.34 13 / 553 44 / 1390 37.12
1.00 0.60 1.65 0.99
0.01
0.1
1
10
100
Favours exposure
Favours control

5) Angiotensin converting enzyme inhibitors (ACEI) and major malformations:

In August 2011 a meta-analysis of ACEI and major malformation was published, concluding that ACEI exposure was not associated with major malformation compared to "other" antihypertensive exposed controls (odds ratio 1.41; 95% confidence interval 0.66 to 3.04). This meta-analysis combined 4 small cohort studies (n < 210 in each study). The last published study was in 2011. Moreover, there were another two meta-analyses in the same publication, one examining the association between the ACEI exposed group versus healthy controls (5 studies) and the other looked for the association between "other" antihypertensive exposed group versus healthy control (4 studies). The two meta-analyses showed an association between congenital malformation and the use of ACEI (odds ratio 1.78; 95% confidence interval 1.07 to 2.94), or the use of "other" antihypertensive (odds ratio 1.45; 95% confidence interval 1.15 to 1.83) was significant [12].

In September 2011 an American group published a large cohort study from the California birth certificate data and the Kaiser Permanente Northern California (RER). The total number of exposed group to ACEI was 704 (more than the total number of the combined studies in the meta-analysis). The association between exposure to ACEI and "other" antihypertensive was calculated by us. This was done by creating 2x2 table and by using the risk in the ACEI exposed group (58/704) and the risk of "other" antihypertensive exposed as a control (327/4390). The odd ratio was calculated (odds ratio 1.12; 95% confidence interval 0.83 to 1.49) and the conclusion was ACEI exposed group was not associated with major malformation compared to "other" antihypertensive exposed controls and this concurred with the meta-analysis conclusion. Additionally, the large cohort study measured a positive association between ACEI exposed group versus healthy control (odds ratio 1.58; 95% confidence interval 1.21 to 2.06) and between "other" antihypertensive exposed group versus healthy control (odds ratio 1.41; 95% confidence interval 1.26 to 1.58). In conclusion, this large study agreed with the meta-analysis of small studies that ACEI and "other" antihypertensive had the same risk estimate [13].

6) Valproic acid and major malformations:

In 2006 a meta-analysis of valproic acid and major congenital malformations was published, concluding that valproic acid was associated with risk of major malformation (Relative risk 3.77; 95% confidence interval 2.18 to 6.52). This

meta-analysis combined 3 small cohort studies (n < 159 in each study). The last published study was in 1999 [14].

In 2010 the EUROCAT antiepileptic group published a large observational study. The total number of babies exposed to valproic acid was 180. The conclusion was similar to that of the meta-analysis, where the use of valproic acid monotherapy was associated with significantly increased risks for 6 of the 14 malformations under consideration example: spina bifida relative risk = 12.7, 95% CI (7.7-20.7) [15].

7) Valproic acid and intellectual development:

In 2010 a meta-analysis of valproic acid exposure in pregnancy and intellectual development was published, showing that valproic acid was associated with reduced intelligence (full-scale IQ was significantly lower (P = 0.001) in the valproic acid group when compared to the control group). This meta-analysis combined 3 small cohort studies (n < 42 in each study). The last published study was in 2005 [16].

In 2010 a group published a prospective observational study by using data from 25 epilepsy centers in the United State and the United Kingdom. The total number of exposed group to valproic acid was 92 (more than the total number of the combined studies in the meta-analysis). The results showed that children exposed to valproic acid had an IQ score 9 points lower than the score of those exposed to lamotrigine (P= 0.009). In this study, the lamotrigine exposed group was considered as a control group because in this study there was no control group of unexposed children included. The final conclusion indicates that valproic acid was associated with reduced intelligence [17].

8) Carbamazepine and intellectual development:

In 2010 a meta-analysis of carbamazepine and intellectual development was published, concluding that carbamazepine was not associated with reduced overall intelligence (full-scale IQ was not significantly lower (P= 0.39) in the carbamazepine group compared with the control group). This meta-analysis combined 3 small cohort studies (n < 87 in each study). The last published study was in 2005 [16].

In 2010 a group from the US and UK published a prospective observational study by using data from 25 epilepsy center in the United State and the United Kingdom. The total number of the exposed group to carbamazepine was 98 (more than the biggest study in the meta-analysis). The final conclusion was that children

exposed to carbamazepine had an IQ score 3 points lower than the score of those exposed to lamotrigine (P= 0. 0.20), where the P value was calculated by us from the existing data. In this model the lamotrigine-exposed group was considered as a control group because in this study there was no control groups of unexposed children included. The final conclusion agreed with the meta-analysis, that carbamazepine was not associated with reduced intelligence [17].

9) Maternal varicella-zoster infection and major malformations:

In March 1994 a meta-analysis of fetal risk of congenital varicella syndrome (CVS) after maternal varicella –zoster virus infection in pregnancy, combined 5 small observation studies (n < than 50 in each study). They found the weighted average risk for CVS associated with maternal varicella infection was 2.2 % (95% confidence interval, 0 to 4.6 %; range, 0 to 9.1 %). The last published study included in was in 1994 [18].

In June 1994 a joint large prospective study in Germany and United Kingdom was published. Between 1980 and 1993, 1739 pregnant women who had varicella during the first 36 weeks of gestation were followed up. The calculated risk of congenital varicella syndrome during the first trimester was 2/472 (0.4%, 95% confidence interval, 0.05 to 1.5 %). The denominator (472) was all pregnancy infected during 0-12 weeks and continuing past 20 weeks gestation, this number was more than the total number of the combined studies in the meta-analysis The conclusion from both meta-analysis study and large prospective study was similar, where the risk of embryopathy associated with maternal infection with varicella-zoster virus after first trimester was < 5% [19].

Quality assessment:

All the selected studies were subjected to methodological quality assessment using the AMASTR method for meta-analytical studies [20], and the MINORS tool for the large cohort studies [21]. The 'assessment of multiple systematic reviews' (AMSTAR) is a tool consisting of an 11 item questionnaire, and each item has four possible answers. There is no overall scoring system in this method. The AMSTAR quality assessment tool falls into three ranges, High (9-11), Medium (5-8), and Low (0-4) [22]. MINORS is a validated methodological index to assess the quality of non-randomized, observational studies. Two points are given for each question if the article reports and provides an adequate answer; one point is given if the article reports on the issue but the answer is inadequate; and no points are given if the article does not report the answer. The global ideal score is out of 24 for comparative studies with a higher score representing greater quality.

References:

1. Dolovich LR, Addis A, Vaillancourt JM, Power JD, Koren G, Einarson TR. Benzodiazepine use in pregnancy and major malformations or oral cleft: meta-analysis of cohort and case-control studies. BMJ. 1998; 317:839–43.

2. Wikner BN, Stiller CO, Bergman U, Asker C, Källén B. Use of benzodiazepines and benzodiazepine receptor agonists during pregnancy: neonatal outcome and congenital malformations. Pharmacoepidemiol. Drug Saf. 2007; 16:1203–10.

3. Enato E, Moretti M, Koren G. The fetal safety of benzodiazepines: an updated meta-analysis. J Obstet Gynaecol Can. 2011 Jan;33(1):46-8. Erratum in: J Obstet Gynaecol Can. 2011 Apr;33(4):319.

4. Fried S, Kozer E, Nulman I, Einarson TR, Koren G. Malformation rates in children of women with untreated epilepsy: a meta-analysis. Drug Saf. 2004; 27:197–202.

5. Veiby G, Daltveit AK, Engelsen BA, Gilhus NE. Pregnancy, delivery, and outcome for the child in maternal epilepsy. Epilepsia. 2009; 50:2130–39.

6. Nikfar S, Abdollahi M, Moretti ME, Magee LA, Koren G. Use of proton pump inhibitors during pregnancy and rates of major malformations: a meta-analysis. Dig. Dis. Sci. 2002; 47:1526–29.

7. Gill SK, O'Brien L, Einarson TR, Koren G. The safety of proton pump inhibitors (PPIs) in pregnancy: a meta-analysis. Am J Gastroenterol. 2009 ;104(6):1541-5.

8. Pasternak B, Hviid A. Use of proton-pump inhibitors in early pregnancy and the risk of birth defects. N. Engl. J. Med. 2010; 363:2114–23.

9. Gill SK, O'Brien L, Koren G. The safety of histamine 2 (H2) blockers in pregnancy: a meta-analysis. Dig. Dis. Sci. 2009; 54:1835–38.

10. Matok I, Gorodischer R, Koren G, Levy A. The safety of intrauterine exposure to H2-blockers: a study by linking computerized databases. Proceedings of the 11th Congress of the European Society for Developmental Pharmacology. Rotterdam, The Netherlands, June 2008, Abstract 45

11. Matok I, Gorodischer R, Koren G, Sheiner E, Wiznitzer A, et al. The safety of H2-blockers use during pregnancy. J. Clin. Pharmacol. 2010; 50:81–87.

12. Walfisch A, Al-maawali A, Moretti ME, Nickel C, Koren G. Teratogenicity of angiotensin converting enzyme inhibitors or receptor blockers. J. Obstet. Gynaecol. 2011; 31:465–72.

13. Li DK, Yang C, Andrade S, Tavares V, Ferber JR. Maternal exposure to angiotensin converting enzyme inhibitors in the first trimester and risk of malformations in offspring: a retrospective cohort study. BMJ. 2011; 343:d5931.

14. Koren G, Nava-Ocampo AA, Moretti ME, Sussman R, Nulman I. Major malformations with valproic acid. Can. Fam. Physician. 2006; 52:441–42.

15. Jentink J, Loane MA, Dolk H, Barisic I, Garne E, et al. Valproic acid monotherapy in pregnancy and major congenital malformations. N. Engl. J. Med. 2010; 362:2185–93.

16. Banach R, Boskovic R, Einarson T, Koren G. Long-term developmental outcome of children of women with epilepsy, unexposed or exposed prenatally to antiepileptic drugs: a meta-analysis of cohort studies. Drug Saf. 2010; 33:73–79.

17. Meador KJ, Baker GA, Browning N, Clayton-Smith J, Combs-Cantrell DT, et al. Cognitive function at 3 years of age after fetal exposure to antiepileptic drugs. N. Engl. J. Med. 2009; 360:1597–605.

18. Pastuszak AL, Levy M, Schick B, Zuber C, Feldkamp M, et al. Outcome after maternal varicella infection in the first 20 weeks of pregnancy. N. Engl. J. Med. 1994; 330:901–5.

19. Enders G, Miller E, Cradock-Watson J, Bolley I, Ridehalgh M. Consequences of varicella and herpes zoster in pregnancy: prospective study of 1739 cases. Lancet. 1994; 343:1548–51.

20. Shea BJ, Grimshaw JM, Wells GA, Boers M, Andersson N, Hamel C, Porter AC, Tugwell P, Moher D, Bouter LM. Development of AMSTAR: BMC Med Res Methodol. 2007 Feb 15;7:10.

21. Slim K, Nini E, Forestier D, Kwiatkowski F, Panis Y, Chipponi J. Methodological index for non-randomized studies (minors): development and validation of a new instrument. ANZ J Surg. 2003 Sep;73(9):712-6.

22. Sequeira-Byron P, Fedorowicz Z, Jagannath VA, Sharif MO. An AMSTAR assessment of the methodological quality of systematic reviews of oral healthcare interventions published in the Journal of Applied Oral Science (JAOS). J Appl Oral Sci. 2011 Oct;19(5):440-7.

Appendix 3: Characterization of studies included in the analysis of Chapter 4.

1) Quetiapine:

Data were from the Seroquel pregnancy registry up to July 2010. In the prospective report, there were six live births with congenital anomaly. The report had 167 live births, 29 cases of elective termination, 19 cases of miscarriage, and 3 cases of fetal death, all the cases without congenital anomaly. In the retrospective report, there were 18 live births with congenital anomaly and two cases of elective termination with congenital anomaly. In the report there were 168 live births, 26 cases of elective termination, 33 cases of miscarriage, and six cases of fetal death, all the cases without congenital anomaly.

2) Quadrivalent human papillomavirus vaccine:

Data were from the Merck pregnancy registry covering the period from first approval (June 1, 2006) to May 31, 2009. In the prospective report, there were 23 cases of infants/fetuses with major birth defects (21 live births, one fetal death and one elective abortion) and one miscarriage with a major malformation. The total number of live births was 974, the total number of fetal death was 10, the total number of elective abortions was 65 and the total number of miscarriages was 64. In the retrospective report, there were 11 cases of infants/fetuses with major birth defects (8 live births, two fetal deaths, and one elective abortion) and one miscarriage with major malformation. The total number of live births was 190, the total number of fetal deaths was six, the total number of elective abortions was 21, and the total number of miscarriages was 44.

3) Montelukast sodium:

Data were from the Merck pregnancy registry covering the period from first approval (February 20, 1998) to July31, 2009. In the prospective report, there were eight cases of infants/fetuses with major birth defects (7 live births, and one elective abortion). The total number of live births was 245, the total number of elective abortions was two, and the total number of miscarriages was three, with no reports of fetal death. In the retrospective report, there were nine cases of infants/fetuses with major birth defects (9 live births) and two miscarriages with major malformations. The total number of live births was 56, there were 10 miscarriages, and there were no fetal deaths or elective abortions.

4) Itraconazole:

Data were from pregnancies reported to the international pharmacovigilance department of the manufacturer (Janssen Pharmaceuticals) [1]. In the prospective

report, there were five cases of infants/fetuses with major birth defects (5 live births). The total number of live births was 156, the total number of fetal deaths was three, the total number of elective abortions was 15, and the total number of miscarriages was 25. In the retrospective report, there were 17 cases of infants/fetuses with major birth defects (14 live births and three elective abortions). The total number of live births was 108, the total number of fetal deaths was one, the total number of elective abortions was 26, and the total number of miscarriages was 31.

5) Fluoxetine:

Data included fluoxetine-exposed pregnancies reported to Lilly and its affiliates before April 1996 [2]. There were 658 first-trimester fluoxetine-exposed pregnancies with outcome other than miscarriage identified prospectively from spontaneous reports. There were 23 cases of major malformations reported in the 658 pregnancies. A total of 426 pregnancies were reported retrospectively, with 89 cases of malformations associated with fluoxetine exposure in the first trimester.

6) Acyclovir:

Data were from a published article based on the acyclovir in pregnancy registry, managed by Wellcome Co., between June 1, 1984 and June 30, 1990 with reports only from the United States [3]. In the prospective report, the number of infants with congenital abnormalities was five and the number of infants without congenital abnormalities was 96. In the retrospective report there were seven infants with congenital abnormalities and 24 infants without congenital abnormalities.

7) Statins:

Data were from a published article with reports from Merck and Co. to 31 December 2002 [4]. In the prospective cases, there were six children with malformations and the denominator was the total live births (154) and total fetal deaths.4 In the retrospective cases, there were 13 offspring with congenital abnormalities (8 live births, 4 elective abortions, and one miscarriage). The denominator was 91, which included the total number of pregnancy outcomes.

8) Mefloquine:

Data were from a published article using the Hoffmann–La Roche global drug safety database for the time frame 31 January 1986 to 26 October 2010 [5]; only cases with first trimester exposure were included. Among prospective cases there were 38 infants and fetuses with birth defects. The denominator was 717,

which included all cases with birth defects (n = 38), normal infants (n = 635), normal fetuses (n = 4), and other disorders (not birth defects) (n = 40). Among the retrospective cases the number of infants/fetuses with birth defects after first trimester mefloquine exposure was 29. The denominator was 115, which included all birth defect cases (n = 29), normal infants (n = 70), normal fetuses (n = 4) and other disorders (not birth defects) (n = 12).

References:

1. Bar-Oz B, Moretti ME, Mareels G, Van Tittelboom T, Koren G. Reporting bias in retrospective ascertainment of drug-induced embryopathy. Lancet 1999;354:1700–1.

2. Goldstein DJ, Corbin LA, Sundell KL. Effects of first-trimester fluoxetine exposure on the newborn. Obstet Gynecol 1997;89:713–8.

3. Andrews EB, Yankaskas BC, Cordero JF, Schoeffler K, Hampp S. Acyclovir in pregnancy registry: six years; experience. The Acyclovir in Pregnancy Registry Advisory Committee. Obstet Gynecol 1992;79:7–13.

4. Pollack PS, Shields KE, Burnett DM, Osborne MJ, Cunningham ML, Stepanavage ME. Pregnancy outcomes after maternal exposure to simvastatin and lovastatin. Birth Defects Res A Clin Mol Teratol 2005;73:888–96.

5. Schlagenhauf P, Blumentals WA, Suter P, Regep L, Vital-Durand G, Schaerer MT, Boutros MS, et al. Pregnancy and fetal outcomes after exposure to mefloquine in the pre- and periconception period and during pregnancy. Clin Infect Dis 2012;54:e124-31.

Appendix 4: Research Ethic Board approval.

Western Research

Research Ethics

Use of Human Participants - Ethics Approval Notice

Principal Investigator: Dr. Doreen Matsui
File Number:6186
Review Level: Delegated
Approved Local Adult Participants:400
Approved Local Minor Participants:0
Protocol Title:FRAME (Foetal Risk Assessment from Maternal Exposure) Clinic Database - 16111E
Department & Institution: Schulich School of Medicine and Dentistry\Paediatrics,Children's Hospital of Western Ontario
Sponsor:
Ethics Approval Date: June 03, 2013 **Expiry Date:**May 31, 2015
Documents Reviewed & Approved & Documents Received for Information:

Document Name	Comments	Version Date
Revised Study End Date	The study end date has been extended to May 31 2015 to allow for continuation of the study.	

This is to notify you that The University of Western Ontario Research Ethics Board for Health Sciences Research Involving Human Subjects (HSREB) which is organized and operates according to the Tri-Council Policy Statement: Ethical Conduct of Research Involving Humans and the Health Canada/ICH Good Clinical Practice Practices: Consolidated Guidelines; and the applicable laws and regulations of Ontario has reviewed and granted approval to the above referenced revision(s) or amendment(s) on the approval date noted above. The membership of this REB also complies with the membership requirements for REB's as defined in Division 5 of the Food and Drug Regulations.

The ethics approval for this study shall remain valid until the expiry date noted above assuming timely and acceptable responses to the HSREB's periodic requests for surveillance and monitoring information. If you require an updated approval notice prior to that time you must request it using the University of Western Ontario Updated Approval Request Form.

Members of the HSREB who are named as investigators in research studies, or declare a conflict of interest, do not participate in discussion related to, nor vote on, such studies when they are presented to the HSREB.

The Chair of the HSREB is Dr. Joseph Gilbert. The HSREB is registered with the U.S. Department of Health & Human Services under the IRB registration number IRB 00000940.

Appendix 5: Search strategies for Chapter 6.

Set	History
1	H1 Antagonists, Histamine OR Antagonists, Histamine H1 Receptor OR Antihistaminics, H1 OR H1 Antihistaminics OR Receptor Blockaders, H1 OR H1 Receptor Blockaders OR Histamine H1 Blockers OR Histamine H1 Receptor Antagonists OR Histamine H1 Receptor Blockaders OR Antagonists, Histamine H1 OR Blockaders, Histamine H1 Receptor OR Antihistamines, Classical OR Classical Antihistamines OR Antihistaminics, Classical OR Classical Antihistaminics OR First Generation H1 Antagonists OR Antihistamines, Sedating OR Sedating Antihistamines OR Histamine H1 Antagonists, Non Sedating OR Second Generation H1 Antagonists OR H1 Antihistamines, Non-Sedating OR H1 Antihistamines, Non Sedating OR Non-Sedating H1 Antihistamines OR Second Generation Antihistamines OR Antihistamines, Second Generation OR Third Generation H1 Antagonists OR Brompheniramine OR Promethazine OR Hydroxyzine OR Tripelennamne OR Dexchlorpheniramine OR Cyproheptadine OR Chlorpheniramine OR Diphenhydramine OR Dimenhydinate OR Doxylamine OR Dimetinidene OR Cyclazine OR Meclizine OR Triprolidine OR Dexchlorpheniramine OR Chloropyramine OR Piperazine Derivatives OR Clemastino OR Pheniramire OR Mizolastine OR Triflupromazine OR Chlorphenoxamine OR Buclzine OR Prochorperazine OR Dicyclomine OR Phenothiazine OR Pheniramine OR Methapyrilene OR Thonzylamine OR Pyrilamine OR Tripelennamine OR Phenyltoloxamine OR Buclizine Or Chlorothen OR Cetirizine OR Fexofenadine OR Loratidine OR Levocetrizine OR Desloratidine OR Terfenadine OR Astemizole
2	limit 1 to humans
3	Malformation OR Abnormality, Congenital OR Congenital Abnormality OR Deformities OR Deformity OR Congenital Defects OR Congenital Defect OR Defect, Congenital OR Defects, Congenital OR Abnormalities, Congenital OR Birth Defects OR Birth Defect OR Defect, Birth OR Defects, Birth OR embryopathy OR Pregnancy Outcomes OR Outcome, Pregnancy OR Pregnancy Complication OR Live Birth OR Stillbirth OR Premature Birth OR Prematurity OR Spontaneous Abortion OR Induced Abortion OR Therapeutic Abortion OR Miscarriage OR Low Birth Weight
4	limit 3 to humans
5	2 AND 4

Appendix 6: Data extraction form for Chapter 6.

Study Title: Drug Name: First Author: Year: Journal:

Design: ❒ retrospective ❒ prospective❒ cohort❒ case control❒ randomized Reference #:

Study Location (year): First Trimester: Quality Score: Type of Control: Publication Type:

	# of pregnancy	# of live birth	Comments
Exposed			
Control			
	# of major malformation	Total	
Exposed			
Control			
	# of preterm infants	Total	
Exposed			
Control			
	# of spontaneous abortions	Total	
Exposed			
Control			
	# of stillbirths	Total	
Exposed			
Control			
	# of low birth weight infants	Total	
Exposed			
Control			

Appendix 7: Characteristics of the included studies for Chapter117 6.

Table 6. Characteristics of the included studies.

First author (publication year)	Publication type (study design)	H1-AHs drug names	Type of control	Pregnancy outcomes measured	Comments
GPRG (1963) [23]	Peer-review journal (cohort study)	Collection of H1-AHs	Not exposed to any H1-AHs	MM, S, St	For NVP meta-analysis, doxylamine data was only used
Mellin (1963) [24]	Peer-review journal (cohort study)	Meclizine, dimenhydrinate, cyclizine	Exposed to H1-AHs under study before or after pregnancy	MM	Used for NVP meta-analysis
Bunde (1963) [25]	Peer-review journal (cohort study)	Doxylamine	Healthy matched control	MM	Used for NVP meta-analysis
Yerushalmy (1965) [26]	Peer-review journal (cohort study)	Meclizine, cylizine	Not exposed to any H1-AHs	MM	Used for NVP meta-analysis
Erez (1971) [27]	Peer-review journal (cohort study)	Hydroxyzine	NVP group that took placebo	MM, S	Used for NVP meta-analysis
Milkovich (1976) [28]	Peer-review journal (cohort study)	Collection of H1-AHs	NVP group that did took treatment and NVP free group	MM	Used for NVP meta-analysis
Kullander (1976) [29]	Peer-review journal (cohort study)	Collection of H1-AHs	Not exposed to any H1-AHs	MM, P	Used for NVP meta-analysis
Heinonen (1977) [30]	Book (cohort study)	Collection of H1-AHs	Not exposed to H1-AHs under study	MM	Excluded in the sensitivity analysis
Newman (1977) [31]	Peer-review journal (cohort study)	Collection of H1-AHs	Not exposed to any H1-AHs	MM	Used for NVP meta-analysis
Shapiro (1978) [32]	Peer-review journal (cohort study)	Meclizine	Not exposed to H1-AHs under study	MM	Excluded in the sensitivity analysis
Smithells (1978) [33]	Peer-review journal (cohort study)	Doxylamine	Exposed to doxylamine in the non-first trimester of pregnancy	MM	Used for NVP meta-analysis

H1-AHs: H1 Antihistamines, MM: Major Malformation, P: Prematurity, S: Spontaneous, St: Stillbirth, L: Low birth weight

Table 6. Characteristics of the included studies (continue).

First author (publication year)	Publication type (study design)	H1-AHs drug names	Type of control	Pregnancy outcomes measured	Comments
Jick (1981) [34]	Peer-review journal (cohort study)	Doxylamine	Not exposed to any H1-AHs	MM	Used for NVP meta-analysis
Fleming (1981) [35]	Peer-review journal (cohort study)	Doxylamine	Not exposed to any H1-AHs	MM	Used for NVP meta-analysis
Gibson (1981) [36]	Peer-review journal (cohort study)	Doxylamine, dicyclome	Not exposed to any H1-AHs	MM	Used for NVP meta-analysis
Michaelis 1 (1983) [37]	Peer-review journal (cohort study)	Doxylamine	Healthy matched control	MM	Used for NVP meta-analysis
Michaelis 2 (1983) [37]	Peer-review journal (cohort study)	Meclizine	Healthy matched control	MM	Used for NVP meta-analysis
Seto (1993) [38]	Peer-review journal (cohort study)	Brompheniramine	Healthy matched control	MM	-
Pastuzak (1996) [39]	Peer-review journal (cohort study)	Astemizole	Healthy matched control	MM, S, St	-
Einarson (1997) [40]	Peer-review journal (cohort study)	Cetirizine, hydroxyzine	Healthy matched control	MM, S, St	-
Schatz (Michigan Medical study) (1997) [41]	Guest editorial (cohort study)	Collection of H1-AHs	Not exposed to H1-AHs under study	MM	Excluded in the sensitivity analysis
Wilton (1998) [42]	Peer-review journal (cohort study)	Cetirizine, loratidine	Not exposed to any H1-AHs	MM, P, S	-
Loebstein (1999) [43]	Peer-review journal (cohort study)	Terfenadine	Healthy matched control	MM, P, St, L	-
Kallen (2002) [44]	Peer-review journal (cohort study)	Collection of H1-AHs that is used for allergy and Collection of H1-AHs that is used for NVP	General population that is not exposed to any H1-AHs	MM, P	For the all H1-AHs meta-analysis, data for allergy was used and for NVP meta-analysis, data for NVP was used

H1-AHs: H1 Antihistamines, MM: Major Malformation, P: Prematurity, S: Spontaneous, St: Stillbirth, L: Low birth weight

Table 6. Characteristics of the included studies (continue).

First author (publication year)	Publication type (study design)	H1-AHs drug names	Type of control	Pregnancy outcomes measured	Comments
Kallen & Mottet (2003) [45]	Peer-review journal (cohort study)	Meclizine	General population that is not exposed to Meclizine	MM	Excluded in the sensitivity analysis
Bsat (2003) [46]	Peer-review journal (cohort study)	Promethazine	NVP group that took non H1-AHs treatment	MM	Used for NVP meta-analysis
Diav-Citrin (2003) [47]	Peer-review journal (cohort study)	Collection of H1-AHs	Healthy control	MM, P, S	-
Moretti (2003) [48]	Peer-review journal (cohort study)	Loratidine	Healthy matched control	MM, S, St	-
Paulus (2004) [49]	Abstract (cohort study)	Cetirizine, levocetirizine	Healthy control	MM, S	-
Boskovic (2004) [50]	Peer-review journal (cohort study)	Doxylamine	Healthy matched control (with no NVP)	MM, S, St, L	Used for NVP meta-analysis
Weber (2008) [51]	Peer-review journal (cohort study)	Cetirizine	Healthy control	MM, P, S	-
Ashkenazi (2013) [52]	Peer-review journal (cohort study)	Doxylamine	NVP group that took non H1-AHs treatment	MM, P, S, L	Used for NVP meta-analysis
Etwel (2014) [16]	Peer-review journal (cohort study)	Cetirizine	Healthy matched control	MM, P, S, St	For P we used that data without confounders
Aldridge (2014) [7]	Peer-review journal (cohort study)	Collection of H1-AHs	Healthy matched control	P, S, St	H2-AHs data was excluded
Eskenazi (1982) [53]	Peer-review journal (case control study)	Doxylamine	Not exposed to Doxylamine	MM	Minor malformation was excluded
Czeizel (2005) [54]	Peer-review journal (case control study)	Dimenhydrinate	Not exposed to Dimenhydrinate	MM	-
Gilboa (2009) [55]	Peer-review journal (case control study)	Collection of H1-AHs	Not exposed to any H1-AHs	MM	-
Li (2013) [3]	Peer-review journal (case control study)	Collection of H1-AHs	Not exposed to any H1-AHs	MM	-

H1-AHs: H1 Antihistamines, MM: Major Malformation, P: Prematurity, S: Spontaneous, St: Stillbirth, L: Low birth weigh

Curriculum vitae

Name: Fatma Etwel

Place and Year of Birth: Tripoli, Libya. 1976

Post-Secondary Education and Degrees:

- Tripoli University Tripoli, Libya 1994-1999 B.Sc. Pharmacy
- University of Western Ontario, Department of Physiology and Pharmacology 2005-2007 Masters of Science
- University of Western Ontario, Department of Physiology and Pharmacology 2011-2016 Ph.D. Candidate

Honors and Awards:

- Tripoli University Tripoli, Libya. Dean's Honors Top Student of Faculty of Pharmacy 1999
- Ministry of High Education in Libya Graduate Scholarship 2004-2007
- Ministry of High Education in Libya Graduate Scholarship 2011-2017

Related Work Experience:

- Demonstrator and Teaching Assistant. Tripoli University Tripoli, Libya. Department of Pharmacology and Clinical Pharmacy 2000-2003
- Lecturer and Teaching Assistant. University of Western Ontario, Department of Physiology and Pharmacology 2006
- Assistant Lecturer. Tripoli University Tripoli, Libya. Department of Pharmacology and Clinical Pharmacy 2007-2010
- Lecturer and Teaching Assistant. University of Western Ontario, Department of Physiology and Pharmacology 2012-2016

Publications:

- **Etwel F**, ElGadi M (2001) Epidemiology of poisoning: A retrospective study at Tripoli Medical Center. 5th Aljam Conf fpr Med Sci.

- **Etwel** F, Rieder MJ, Bend JR, Koren G (2008) A surveillance method for the early identification of idiosyncratic adverse drug reactions. Drug Saf. 2008;31(2):169-80.

- Koren G, Elzagallaai A, **Etwel F** (2011) Safety assessment in pediatric studies. Handb Exp Pharmacol. 2011;205:169-80.

- **Etwel F**, Hutson JR, Madadi P, Gareri J, Koren G. Fetal and perinatal exposure to drugs and chemicals: novel biomarkers of risk. Annu Rev Pharmacol Toxicol. 2014;54:295-315.

- Fujii H, Goel A, Bernard N, Pistelli A, Yates LM, Stephens S, Han JY, Matsui D, **Etwell F**, Einarson TR, Koren G, Einarson A. Pregnancy outcomes following gabapentin use: results of a prospective comparative cohort study. Neurology. 2013 Apr 23;80(17):1565-70.

- **Etwel F**, Djokanovic N, Moretti ME, Boskovic R, Martinovic J, Koren G. The fetal safety of cetirizine: an observational cohort study and meta-analysis. J Obstet Gynaecol. 2014 Jul;34(5):392-9.

- **Etwel F**, Russell E, Rieder MJ, Van Uum SH, Koren G. Hair cortisol as a biomarker of stress in the 2011 Libyan war. Clin Invest Med. 2014 Dec 1;37(6).

- Marchenko A, **Etwel F**, Olutunfese O, Nickel C, Koren G, Nulman I. Pregnancy outcome following prenatal exposure to triptan medications: a meta-analysis. Headache. 2015 Apr;55(4):490-501.

- Kaplan YC, Ozsarfati J, **Etwel F**, Nickel C, Nulman I, Koren G. Pregnancy outcomes following first-trimester exposure to topical retinoids: a systematic review and meta-analysis. Br J Dermatol. 2015 Nov;173(5):1132-41.

- Terrana N, Koren G, Pivovarov J, **Etwel F**, Nulman I. Pregnancy Outcomes Following In Utero Exposure to Second-Generation Antipsychotics: A Systematic Review and Meta-Analysis. J Clin Psychopharmacol. 2015 Oct;35(5):559-65.

- **Etwel F**, Koren G. When positive studies of novel therapies are subsequently nullified: cumulative meta-analyses in preeclampsia. Clin Invest Med. 2015 Oct 7;38(5):E274-83.

- **Etwel F**, Koren G. Bias against the Null Hypothesis in Retrospective Registries of Gestational Drug Exposure. J Obstet Gynaecol Can. 2016 (in press).

- **Etwel F**, Faught LH, Rieder MJ, Koren G. The risk of adverse pregnancy outcome after first trimester exposure to H1 antihistamines: a systematic review and meta-analysis. Drug Safety 2016 (in press).