Appendix: Review Protocol

Safety of cotrimoxazole in pregnancy: a systematic review and meta-analysis

BACKGROUND

Daily prophylaxis with cotrimoxazole (CTX) significantly reduces the risk of morbidity and mortality among people living with HIV. Data from animal studies and small, retrospective cohorts have led the US Food and Drugs Administration to class cotrimoxazole as a Class D drug, and guidelines from the United States and Europe recommend against using cotrimoxazole in the first trimester of pregnancy. However, data derived from such studies are subject to well know biases and the overall evidence base has not been systematically reviewed since 2005; moreover, the benefits of administering cotrimoxazole in terms of reduced risk of mortality and morbidity are substantial.

The proposed systematic review aims to update the available evidence on adverse maternal and infant outcomes associated with exposure to cotrimoxazole during pregnancy in order to inform recommendations for the updated World Health Organization guidelines for the use of cotrimoxazole prophylaxis in HIV positve individuals, including during pregnancy.

SEARCH STRATEGY

Search terms

1. pregnan* (all fields)
2. pregnancy (mesh)
3. 1 or 2
4. Biseptol or Septrin or Cotrim or Bactrimel or Cotrimoxazole or Co-trimoxazole or Trimethoprim-Sulfamethoxazole or TMP-SMZ or Trimethoprim or Bactrim or Septra or SXT or TMP-SMX or TMP-SMZ or TMP-sulfa or Sulfatrim (all fields)
5. 3 AND 4

Databases

- MEDLINE via PubMed
- EMBASE

- Current Controlled Trials (www.controlled-trials.com)
- International AIDS Society

Restrictions

No date or language restriction will be applied.

INCLUSION CRITERIA

Types of studies

- Randomized and non-randomized trials
- Prospective and retrospective cohorts
- Case control studies
- Unsystematic observations (case series or case reports) will be excluded from all analyses

Types of participants

Inclusions:

- Women exposed to CTX during pregnancy

Types of interventions

- CTX during pregnancy. Women exposed to Trimethoprim will also be eligible for inclusion

Types of comparitors

- Pregnant women not exposed to CTX during pregnancy

Types of outcomes

Primary

- Birth defect of any kind
    - This outcome will be stratified by trimester of exposure

Secondary

- Neural tube defects
- Spontaneous abortions
- Termination of pregnancy
- Stillbirths
- Preterm delivery
- Severe AEs
- Mortality due to AEs

ASSESSMENT OF METHODOLOGICAL QUALITY

The following data will be extracted as potentially influencing the methodological quality of studies:

- Direct ascertainment of CTX use
- Adjustment for confounders
- Prospective study design
- Outcomes reported by trimester
- Outcomes reported by folate supplement use
- Confounding by indication

DATA ANALYSIS

Prevalence estimates

Point estimates and 95% confidence intervals (95% CI) will be calculated for the proportion of birth defects reported among live births for each study. Spontaneous and induced abortions and stillbirths will be excluded from the denominator of birth defects, consistent with reporting norms. The variance of the raw proportions will be stabilised using a Freeman-Tukey type arcsine square-root transformation and estimates pooled using a DerSimonian-Laird random effects model. Prevalence and 95%CIs will be calculated for all secondary outcomes. Because the background prevalence rates of these outcomes varies considerably across study settings, these data will not be pooled, but where rates are reported for women exposed to both efavirenz- and non-efavirenz-based regimens, pooled relative risks will be calculated.

Meta-analysis

For case-control studies reporting on birth outcomes of infants exposed to CTX during the first trimester vs. infants not exposed to any drug, odds ratios (ORs) and 95% CIs will be calculated and data pooled using the DerSimonian-Laird random effects method. In the case of zero outcome events in one arm, the Haldane method will be applied, adding 0.5 to each arm.

Heterogeneity

The τ^2 statistic will be calculated to assess the proportion of overall variation attributable to between-study heterogeneity as this is less affected by the number of studies than the more commonly used I^2 statistic. Subgroup analyses will be conducted to assess the potential effect on the pooled estimates of study design, study location, duration of efavirenz exposure, and status of publication. A p-value less than 0.05 will be considered to be significant.

Statistical software

Analyses will be conducted using Stata (version 12, www.stata.com).

PRISMA 2009 Checklist

Section/topic	#	Checklist item	Reported on page #
TITLE			
Title	1	Identify the report as a systematic review, meta-analysis, or both.	1
ABSTRACT			
Structured summary	2	Provide a structured summary including, as applicable: background; objectives; data sources; study eligibility criteria, participants, and interventions; study appraisal and synthesis methods; results; limitations; conclusions and implications of key findings; systematic review registration number.	2
INTRODUCTION			
Rationale	3	Describe the rationale for the review in the context of what is already known.	3
Objectives	4	Provide an explicit statement of questions being addressed with reference to participants, interventions, comparisons, outcomes, and study design (PICOS).	3
METHODS			
Protocol and registration	5	Indicate if a review protocol exists, if and where it can be accessed (e.g., Web address), and, if available, provide registration information including registration number.	Protocol
Eligibility criteria	6	Specify study characteristics (e.g., PICOS, length of follow-up) and report characteristics (e.g., years considered, language, publication status) used as criteria for eligibility, giving rationale.	4, Protocol
Information sources	7	Describe all information sources (e.g., databases with dates of coverage, contact with study authors to identify additional studies) in the search and date last searched.	4, Protocol
Search	8	Present full electronic search strategy for at least one database, including any limits used, such that it could be repeated.	Protocol
Study selection	9	State the process for selecting studies (i.e., screening, eligibility, included in systematic review, and, if applicable, included in the meta-analysis).	4, Protocol
Data collection process	10	Describe method of data extraction from reports (e.g., piloted forms, independently, in duplicate) and any processes for obtaining and confirming data from investigators.	4, Protocol
Data items	11	List and define all variables for which data were sought (e.g., PICOS, funding sources) and any assumptions and simplifications made.	4, Protocol
Risk of bias in individual studies	12	Describe methods used for assessing risk of bias of individual studies (including specification of whether this was done at the study or outcome level), and how this information is to be used in any data synthesis.	5, Protocol
Summary measures	13	State the principal summary measures (e.g., risk ratio, difference in means).	5, Protocol

PRISMA 2009 Checklist

Synthesis of results	14	Describe the methods of handling data and combining results of studies, if done, including measures of consistency (e.g., I^2) for each meta-analysis.	5, Protocol

Section/topic	#	Checklist item	Reported on page #
Risk of bias across studies	15	Specify any assessment of risk of bias that may affect the cumulative evidence (e.g., publication bias, selective reporting within studies).	5, Protocol
Additional analyses	16	Describe methods of additional analyses (e.g., sensitivity or subgroup analyses, meta-regression), if done, indicating which were pre-specified.	5, Protocol
RESULTS			
Study selection	17	Give numbers of studies screened, assessed for eligibility, and included in the review, with reasons for exclusions at each stage, ideally with a flow diagram.	6
Study characteristics	18	For each study, present characteristics for which data were extracted (e.g., study size, PICOS, follow-up period) and provide the citations.	6
Risk of bias within studies	19	Present data on risk of bias of each study and, if available, any outcome level assessment (see item 12).	8
Results of individual studies	20	For all outcomes considered (benefits or harms), present, for each study: (a) simple summary data for each intervention group (b) effect estimates and confidence intervals, ideally with a forest plot.	6
Synthesis of results	21	Present results of each meta-analysis done, including confidence intervals and measures of consistency.	6, Figure 2
Risk of bias across studies	22	Present results of any assessment of risk of bias across studies (see Item 15).	8, Supplementary tables
Additional analysis	23	Give results of additional analyses, if done (e.g., sensitivity or subgroup analyses, meta-regression [see Item 16]).	6, Figure 2
DISCUSSION			
Summary of evidence	24	Summarize the main findings including the strength of evidence for each main outcome; consider their relevance to key groups (e.g., healthcare providers, users, and policy makers).	8-11
Limitations	25	Discuss limitations at study and outcome level (e.g., risk of bias), and at review-level (e.g., incomplete retrieval of identified research, reporting bias).	8-11
Conclusions	26	Provide a general interpretation of the results in the context of other evidence, and implications for future research.	8-11
FUNDING			
Funding	27	Describe sources of funding for the systematic review and other support (e.g., supply of data); role of funders for the systematic review.	11

PRISMA 2009 Checklist

From: Moher D, Liberati A, Tetzlaff J, Altman DG, The PRISMA Group (2009). Preferred Reporting Items for Systematic Reviews and Meta-Analyses: The PRISMA Statement. PLoS Med 6(6): e1000097. doi:10.1371/journal.pmed1000097

For more information, visit: **www.prisma-statement.org**.

Supplementary Table 1: leave-one-out meta-analysis

Study	Pooled proportion if corresponding study left out
Anderson et al, 2013	3.3 (1.5-5.0)
Angelakis et al, 2013 (T1)	3.5 (1.8-5.1)
Angelakis et al, 2013 (T2/3)	3.5 (1.8-5.1)
Bailey et al, 1983	3.7 (2.0-5.5)
Brumfitt et al 1973 (T1)	3.7 (2.0-5.4)
Brumfitt et al 1973 T2/3)	3.4 (1.7-5.2)
Brumfitt et al 1973 (T1)	3.5 (1.8-5.2)
Carcopino et al, 2007	3.4 (1.7-5.1)
Colley et al, 1982	3.4 (1.7-5.2)
Denoeud-Ndam et al, 2014	3.7 (1.8-5.5)
Jungmann et al, 2001	3.4 (1.7-5.1)
Khan et al, 2001	3.7 (2.0-5.4)
MMSS, 2003	3.3 (1.6-5.0)
Klement, 2014	3.7 (1.9-5.4)
Matok et al, 2009	2.6 (1.2-4.0)
Roushan er al, 2009	3.5 (1.8-5.2)
Valentini et al, 2009	3.7 (2.0-5.5)
Walter et al, 2006	3.6 (1.8-5.3)
Yaris et al, 2004	3.5 (1.8-5.2)

Supplementary Table 2: Secondary outcomes

Study	Co-infection	Spontaneous abortion	Stillbirth/ IUFD	Small for gestational age	Pre-term birth	Comments
Anderson *et al*, 2013	UTI	31/265	--	--	--	Risk with first trimester exposure
Angelakis et al, 2013	Brucellosis cohort	--	--	1/13 (CTX live births) versus 0/1	--	Compared to those without exposure to any drug; note that here exposure to CTX is any duration (>2weeks) but study refers to long-term treatment as >5weeks
Bailey et al, 1983	Covert UTI	--	1/44	--	--	
Colley et al, 2005	Unclear	--	2/211 CTX 7/127 SMZ	12/211 CTX 13/127 SMZ	13/211 CTX 7/127 SMZ	Small for gestational age = low birth weight =<2500g + Intrauterine growth retardation; stillbirth/Intrauterine growth retardation= stillborn + perinatal death (death within 30 days of birth)
Denoeud-Ndam et al, 2014	HIV	8/364	12/364	--	--	
Dow et al, 2013	HIV	--	--	58/762	88/373	Small for gestational age = low birth weight =<2500g
Klement et al, 2014	HIV	0/126	4/126	23/117	18/117	Small for gestational age = low birth weight =<2500g
Matok et al, 2009	UTI	--	6/571 (perinatal mortality)	--	--	All folate antagonists (n=571)
Roushan, et al 2011	Brucellosis	5/14	--	--	0/14	
Santos et al, 2011	Unclear		--	49/8192 vs 165/55146 AOR 1.61 (1.16–2.23)	--	
Valentini et al, 2009	Toxoplasmosis	0/76	0/76	--	--	Likely no small for gestational age cases but unclear with relation to those without congenital infection at birth
Walter et al, 2006	HIV			10/65	12/67	Low birth weight=small for gestational age (<2500grams)
Wen et al, 2008	UTI (mainly)	--	--	AOR 1.05 (0.99–1.13)	--	Low birth weight= intrauterine growth retardation; (<10th percentile)
Yaris et al, 2004	UTI	0/11	--	--	0/11	Includes 2 cases where gentamicin was also

						given

AOR, Adjusted odds ratio; CTX, cotrimoxazole; IUFD, Intrauterine fetal death; IUGR, Intrauterine growth retardation; LBW, low birth weight; SGA, small for gestational age; SMZ, sulfamethoxazole; UTI, Urinary tract infection

Supplementary Table 3: Maternal toxicity

Study	Number of adverse drug events	Description
Brumfitt et al 1973	22/126	Vaginitis, rash, nausea, vomiting, diarrhoea
Carcopino et al, 2007	2/22	Hepatitis*, cutaneous rash (2 stopped)
Denoeud-Ndam et al, 2014	2/364	Cutaneous rash (1 stopped)
Klement et al, 2014	4*/132	Cutaneous rash (1 stopped)
Valentini et al, 2009	1/76**	Rash

**2 additional cases of rash were reported but these were unrelated to cotrimoxazole use

Supplementary Table 4: Risk of bias

Study	Direct ascertainment of CTX use	Adjustment for confounders	Prospective study design	Outcomes reported by trimester	Outcomes reported by folate supplement use	Confounding by indication**	Overall risk of bias
Anderson *et al*, 2013	Yes	Yes	No	Yes	No	Yes	Moderate
Anderson et al, 2013	Yes	Yes	No	Yes	Yes*	No	Moderate
Angelakis et al, 2013	No	No	No	Yes	No	Yes	High
Bailey et al, 1983	Yes	No	Yes	No	No	Yes	High
Brumfitt & Pursell, 1973	Yes	Yes	Yes	Yes	No	No	Low
Carcopino et al, 2007	Yes	No	Yes	Yes	No	No	Low
Colley et al, 1982	Yes	No	No	Yes	No	Unclear	High
Czeizel et al, 2001	No	Yes	No	Yes	Yes	No	Low
Denoeud-Ndam et al, 2014	Yes	No	Yes	No	Yes	No	Low
Dow et al, 2013	Yes	Yes	Yes	No	Yes	No	Low
Jungman et al, 2001	Yes	No	No	Yes	No	No	Moderate
Hernández-Díaz, 2001	No	No	No	No	Yes	No	High
Hernández-Díaz et al, 2000	No	No	No	Yes	Yes	No	Moderate
Hill et al, 1988	No	No	No	Yes	No	Unclear	High
Khan et al, 2001	Yes	No	No	Yes	No	Yes	High
Klement et al, 2014	Yes	Yes	Yes	No	Yes	No	Low
Matok et al, 2009	Yes	Yes	No	Yes	No	Unclear	Moderate/high
Meijer et al, 2005	Yes	No	No	No	No	Unclear	High
MMSS 2003	Unclear	No	Yes	No	No	Unclear	High
Roushan, et al 2011	Unclear	No	Unclear	Yes	No	Yes	High
Santos et al, 2011	Yes	Yes	Yes	No	No	Unclear	Moderate
Valentini et al, 2009	Yes	No	No	Yes	Yes	Yes	Moderate
Walter et al, 2006	No	No	Yes	Yes	Yes	No	Low
Wen et al, 2008	Yes	Yes	No	No	No	Unclear	High
Yaris et al, 2004	Yes	No	Yes	Yes	No	Yes	High

*Controlled for in regression model

**co-infection related to outcome reported

Supplementary Table 5: Prevalence of congenital anomalies associated with exposure to cotrimoxazole during pregnancy

Quality assessment							No of patients	Pooled prevalence (95% CI)	Quality	Importance
No of studies	Design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations				
Prevalence of congenital anomalies										
16	4 RCTs and 12 observational studies	Serious[1]	Serious[2]	Serious[3]	Serious[4]	Reporting bias[5]	232/4196	3.5% (95% CI 1.8-5.1%)	VERY LOW	CRITICAL

[1] Risk of bias rated as serious. This is due to unmeasured confounding (protective factors and risk factors) and information bias (retrospective analyses); [2] Point estimates vary widely across individual studies; high statistical heterogeneity; [3] Most evidence derived from HIV negative populations and high-income settings; [4] Small sample size and low number of events considering background prevalence of outcome; [5] High risk of publication bias towards the publication of adverse birth outcomes

Supplementary Table 6: Odds of being exposed to cotrimoxazole among patients with congenital anomalies compared to patients not exposed to cotrimoxazole

Quality assessment							No of patients	Effect	Quality	Importance
No of studies	Design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations	Control	Relative (95% CI)		
Any congenital anomaly										
2	Case control studies	Serious[1]	Serious[2]	Serious[3]	Serious[4]	Reporting bias[2]	1247/39537	0.64 (0.12-3.36)	VERY LOW	CRITICAL
Neural tube defects										
1	Case control study	Serious[1]	No serious inconsistency	Serious[3]	Serious[4]	Reporting bias[2]	8/6660	3.36 (1.10-10.29)	VERY LOW	CRITICAL
Cardiovascular defects										
1	Case control studies	Serious[1]	Not applicable[6]	Serious[3]	Serious[4]	Reporting bias[2]	17/8387	2.94 (1.57-5.52)	VERY LOW	CRITICAL
Oral clefts										
2	Case control studies	Serious[1]	No serious inconsistency	Serious[3]	Serious[2]	Reporting bias[2]	32/9063	2.04 (1.23-3.36)	VERY LOW	CRITICAL
Urinary tract defects										
1	Case control studies	Serious[1]	Not applicable[6]	Serious[3]	Serious[2]	Reporting bias[2]	18786/592899	0.90 (0.21-3.89)	VERY LOW	CRITICAL

[1] Risk of bias: Rated as Serious. This is due to unmeasured confounding (protective factors and risk factors) and information bias (retrospective analyses); [2] Inconsistency: Rated as Serious. There is unexplained heterogeneity with an I^2 of 84%. The two studies have odds ratios either side of the 'null' [3] Indirectness: Rated as Serious. The studies were done in HIV-negative individuals; [4] Imprecision: Rated as Serious. Wide confidence intervals; [5] Publication bias favouring the publication of negative birth outcomes; [6] Inconsistency: The results are from a single study and inconsistency is therefore not applicable