



University of Thessaly

“Critical appraisal of the literature reporting association between Measles-Mumps-Rubella Vaccine and autism.”

«Κριτική αξιολόγηση της βιβλιογραφικής αναφοράς για τη σύνδεση μεταξύ του εμβολίου της ιλαράς – παρωτίτιδας - ερυθράς και του αυτισμού»

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Critical appraisal of the literature reporting association between Measles-Mumps-Rubella Vaccine and autism.

Abstract

Background: It has been suggested that the measles, mumps, and rubella vaccine (MMR) is a cause of regressive autism. Mumps, measles and rubella are serious diseases that can lead to potentially fatal illness, disability and death. However, public debate over the safety of the trivalent MMR vaccine and the resultant drop in vaccination coverage in several countries persists, despite its almost universal use and accepted effectiveness. The cause of autism is unclear, vaccines have been incriminated. The aim of this study was the critical appraisal of the literature reporting association between Measles-Mumps-Rubella Vaccine (MMR) and autism.

Methods: PubMed was searched for systematic reviews (SRs), meta-analysis, observation studies, of association between MMR and autism published from January 2006 through February 2016. In the present study we focused to assess the scientific validity and quality of published article. Guidelines and assessment tools we used to provide a structured approach to the process of critical appraisal follow the check compliance PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) statement for meta-analysis or systematic review and STROBE (Strengthening the Reporting of Observational Studies in Epidemiology) Explanation and Elaboration document for observational studies.

Results: The search identified 18 eligible article included in two SRs, two meta-analysis, ten OS. Eleven items /sub-items (PRISMA) were reported by more than 75% of SRs, meta-analysis and 14 items/sub-items (STROBE) were reported by more than 70% of OS. Some essential methodological aspects of SRs, meta-analysis and OS (such as risk of bias, effect estimates, absolute risks, missing data and flow diagram) were underreported.

Conclusion: The total of the meta-analysis, the systematic review and the observational studies have found no evidence for the link between vaccination and the subsequent risk of developing autism or autistic spectrum disorder. The quality of reporting in meta-analysis systematic review and observational studies in MMR and autism was considered satisfactory, although certain items were underreported.

Keywords MMR, Autism, ASD, epidemiology, developmental disorders, vaccine/immunization

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Introduction

Autism is a developmental disorder characterized by impaired social interaction, difficulty with verbal and nonverbal communication, and limited activities and interests (NINDS 2006). Although autism was first described by American psychiatrist Leo Kanner¹⁻³ in 1943 the cause and treatment of this brain disorder still remains poorly understood. The Diagnostic and Statistical Manual of Mental Disorders (DSM –IV) classifies autism as one disease in a class of developmental disorders referred to as autism spectrum disorders (ASDs) or pervasive developmental disorders (PDDs) (Strock 2007, NINDS 2006). Asperger syndrome, Rett syndrome, childhood disintegrative disorder, and pervasive developmental disorder not otherwise specified along with autism make up the ASDs. Autism is the most common of the ASDs. The etiology of ASD is unknown, although both genetic and environmental factors play a causative role⁴⁻⁶. Now, autism is no longer regarded as a simple developmental disorder but rather a biological disorder of complex etiology and heterogeneity⁷⁻¹⁰, with evidence of developmental delay within the first 3 years of life. Typically, autism is characterized by “qualitative deficits” in 4 major categories: 1) deficits of developmental rates and/or profiles, 2) deficits of responses to sensory stimuli, 3) deficits of speech, language and communication capabilities, and 4) deficits of social interactions and/or manners of relating to other people. Although the diagnosis of autism is made during early childhood, the disorder continues to persist well into adulthood, eventually becoming a lifelong neuro disability.

Vaccines are considered one of the greatest public health achievements of the 20th century. Vaccinations have significantly reduced or abolished numerous communicable diseases that used to harm or kill many people. Yet, those infectious diseases can still occur in people who are not protected by the immunizations, so the reason for widespread use of vaccinations is clearly evident. Measles, mumps and rubella (MMR) are serious diseases that can lead to potentially fatal illnesses, disabilities and death. MMR are particularly prevalent in low-income countries where vaccination programs are inconsistent and the mortality rate from disease is high. However, in high-income countries MMR are now rare, due to large-scale vaccination programs¹¹⁻¹⁴. The single component live attenuated vaccines of MMR have been licensed in the USA since the 1960s COHRANE. These single vaccines have been shown to be highly effective at reducing the morbidity and mortality rates associated with these childhood illnesses. No national health policy recommends that the MMR vaccine be given as three separate vaccines. Combined live attenuated MMR vaccine was introduced in the USA in the 1970s. MMR is included in the World Health Organization’s Expanded Program on Immunization and it is used in over 50 European countries, the USA, Canada, Australia and New Zealand; in total, over 90 countries around the world use the MMR vaccine. Accepted recommendations are that the first dose should be administered on or after the first birthday and the second dose of MMR at least 28 days later. In many European countries the second dose is administered at four to 10 years of age¹⁵⁻¹⁸.

Over the past several years much concern has been raised regarding the potential links of childhood vaccinations with the development of autism and autism spectrum disorders (ASD). The vaccinations that have received the most attention are the measles, mumps, rubella (MMR) vaccine and thimerosal-containing vaccines such as the diphtheria, tetanus, pertussis (DPT or DT) vaccine. Wakefield et al¹⁸ were the first to propose that administration of the measles, mumps, and rubella (MMR) vaccine may be causally related to the development of autism. A rising awareness of autism incidence, prevalence, and the postulated causation of childhood vaccinations has led to both an increased distrust in the trade-off between vaccine benefit out-weighting potential risks and an opportunity for disease resurgence. Vaccine-preventable diseases clearly still hold a presence in modern day society and the decision to opt out of MMR or other childhood vaccination schedules because of concerns regarding the development of autism should be properly evaluated with available evidence. MMR vaccination is a requirement for entry into schools, so any increase in adverse events associated with the vaccine carries widespread public health importance.

The objective of this study was the critical appraisal of the literature reporting association between Measles-Mumps-Rubella Vaccine (MMR) and autism. This is a systematic process that was used to identify the strengths and weaknesses of research articles in order to assess the usefulness and validity of the research findings. Although the methodological criteria by which the validity of a study is assessed vary according to its design, some general principles underpin the evaluation of any research study.

Methods

Data Sources, Search Strategies and Studies Selection

Literature for this review was systematically identified by searching PubMed for papers published between January 2006 and February 2016. The search was limited to the following criteria: "English" language, "Humans" species, and as a search criterion the following term: "measles-mumps-rubella" And "autism (i.e., to appear in the title – checking by MESH). The search strategy included Clinical Trials (CTs): "meta-analyses", "systematic reviews" (SRs), "randomized controlled trials" (RCTs), "observational studies" (OS i.e., cohort, case control, and cross-sectional). An SR or meta-analysis was considered eligible when it were published in a peer-reviewed journal and provided the complete list of references of all articles included in the SR or meta-analysis. These articles were eligible if they had been published as full papers or short reports in a regular issue or supplement of peer-reviewed journals indexed in PubMed. Articles published as editorials, letters, conferences or meeting abstracts were excluded. In the study, were include article which refer to the link between MMR and autism. The articles excluded because studied other factors of correlation.

Data Extraction and Reporting Assessment Tool

In the present study we focused to assess the scientific validity and quality of published Clinical Trials (CTs). The first question to ask in any research article is whether its topic is relevant to field of study and if the article add new ideas and knowledge the scientific research endeavor. The fundamental task of critical appraisal is to identify the specific research question that an article addresses, as this process will determine the optimal study design. Furthermore, the questions to be answered when evaluating are if the study design is appropriate for the research question and if the study methods address the key potential sources of bias (Table 1). This process enables to assess the study's usefulness and whether its findings are trustworthy. The criteria used to assess the validity and relevance of scientific literature, vary according to its design of research study.

Guidelines and assessment tools we used to provide a structured approach to the process of critical appraisal follow the check compliance PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) statement for meta-analysis or systematic review and STROBE (Strengthening the Reporting of Observational Studies in Epidemiology) Explanation and Elaboration document for observational studies. The checklist in the PRISMA includes 27 items pertaining to the content of a systematic review and meta-analysis, which include the title, abstract, methods (twelve items), results (seven items), discussion (three items) and funding. Respectively, the checklist in the STROBE includes 22 items pertaining to the content of Observational Studies, which include the title, abstract, methods (nine items), results (five items), discussion (four items) and funding. All items were investigated in terms of whether they were reported, not whether they were actually carried out during the study. Items were to be scored as "yes" if they were reported in enough detail to allow the reader to judge that the definition had been met. Especially in the case of matching criteria, the item was coded as "yes" only when the matching procedure was explicitly described (i.e., the number of controls per case was specified and the matching variables were clearly stated). Alternative responses (apart from "yes" or "no") and unclear responses to each question were coded as negative responses.

In order to clarify whether an article will be included or not from the study we used the flow diagram. It depicts the flow of information and maps out information about the number of records identified in the literature searches, the number of studies included and excluded, and the reasons for exclusions. Then, we focused of recording of the findings. In order to clarify whether an article is supporting or not the correlation of MMR to autism.

Selected Abbreviations and Acronyms

STROBE = *Strengthening the Reporting of Observational Studies in Epidemiology*

PRISMA = *Preferred Reporting Items for Systematic Reviews and Meta-Analyses*

SRs = *Systematic Reviews*

OS = *Observational Studies*

Methodological Evaluation

The evaluation of articles both in systematic reviews, meta-analysis and in observations studies were restricted to items concerning the methods and results sections.

The PRISMA statement: methodological items refer to the reporting of protocol and registration, study characteristics (e.g. PICOS, length of follow-up, years considered, language, publication status) and report characteristics (e.g. years considered, language, publication status), the electronic search strategy, the list and define all variables for which data were sought, the describe of methods used for assessing risk of bias of individual studies (e.g. risk ratio, difference in means), data collection process and data items (e.g. databases with dates of coverage, contact with study authors to identify additional studies), any efforts to address potential sources of bias, risk of bias in individual studies and risk of bias across studies, the principal summary measures, synthesis of results and additional analyses (e.g. sensitivity or subgroup analyses, meta-regression). Furthermore, the items in the results section refer to the reporting of study selection and study characteristics (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 and for each study, present characteristics for which data were extracted, results of individual studies (results of individual studies and effect estimates and confidence intervals, ideally with a forest plot), synthesis of results (present results of each meta-analysis done, including confidence intervals and measures of consistency), additional analysis (results of additional analyses).

The STROBE statement: methodological items refer to the reporting of study design, setting of the study, participants' information (eligibility criteria, sources and methods of selection, or matching criteria if relevant), definition of all variables used, data sources and methods of measurement, any efforts to address potential sources of bias, study size, handling of quantitative variables in the study and performed statistical methods (i.e. methods used to control for confounding and to examine subgroups and interactions, methods of handling missing data or how loss to follow-up was addressed, methods of matching of cases and controls, analytical methods taking account of sampling strategy and any description of sensitivity analysis). Furthermore, the items in the results section refer to the reporting of participants' information (numbers of individuals at each stage of the study, reasons for nonparticipation at each stage, use of flow diagram), descriptive data (characteristics of study participants, numbers of participants with missing data, summary of follow-up time), outcome data (numbers of outcome events or summary measures), main results (unadjusted or confounder-adjusted estimates and their precision, presentation of 95% confidence intervals, category boundaries when continuous variables were categorized and translation of estimates of relative risk to absolute risk for a meaningful time period), and other analyses done (e.g. analyses of subgroups and interactions, and sensitivity analyses).

Table 1. Basic Questions for Ask when Critically Appraising a Research Article

Does the study add anything new?

What type of research question is being asked?

Was the study design appropriate for the research question?

Did the study methods address the most important potential sources of bias?

Was the study performed according to the original protocol?

Does the study test a stated hypothesis?

Were the statistical analyses performed correctly?

Do the data justify the conclusions?

Are there any conflicts of interest?

Table 2. Frequency of reporting of the items in the PRISMA statement, overall and in each systematic review and meta-analysis.

Section / topic	#	Checklist item	n (%)
TITLE			
Title	1	Identify the report as a systematic review, meta-analysis, or both.	4(100%)
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.	4(100%)
INTRODUCTION			
Rationale	3	Describe the rationale for the review in the context of what is already known.	4(100%)
Objectives	4	Provide an explicit statement of questions being addressed with reference to participants, interventions, comparisons, outcomes, and study design (PICOS).	4(100%)
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.	4(100%)
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(100%)
Information sources	7	Describe all information sources in the search and date last searched.	4(100%)
Search	8	Present full electronic search strategy for at least one database, including any limits used, such that it could be repeated.	3(75%)
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).	3(75%)
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.	3(75%)
Data items	11	List and define all variables for which data were sought (e.g., PICOS, funding sources) and any assumptions and simplifications made.	3(75%)
Risk of bias in individual studies	12	Describe methods used for assessing risk of bias of individual studies and how this information is to be used in any data synthesis.	2(50%)
Summary measures	13	State the principal summary measures (e.g., risk ratio, difference in means).	3(75%)
Synthesis of results	14	Describe the methods of handling data and combining results of studies, if done, including measures of consistency for each meta-analysis.	3(75%)
Risk of bias across studies	15	Specify any assessment of risk of bias that may affect the cumulative evidence.	2(50%)
Additional analyses	16	Describe methods of additional analyses, if done, indicating which were pre-specified.	2(50%)
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.	1(25%)
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.	4(100%)
Risk of bias within studies	19	Present data on risk of bias of each study and, if available, any outcome level assessment.	2(50%)
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.	2(50%)
Synthesis of results	21	Present results of each meta-analysis done, including confidence intervals and measures of consistency.	2(50%)
Risk of bias across studies	22	Present results of any assessment of risk of bias across studies	2(50%)
Additional analysis	23	Give results of additional analyses, if done (e.g., sensitivity or subgroup analyses, meta-regression)	3(75%)
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).	4(100%)
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).	2(50%)
Conclusions	26	Provide a general interpretation of the results in the context of other evidence, and implications for future research.	3(75%)
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.	3(75%)

Table 3. Frequency of reporting of the items in the STROBE statement, observational studies

Section / topic	#	Checklist item	n (%)
TITLE			
	1	a. Indicate the study's design with a commonly used term in the title or the abstract	9 (90%)
		b. Provide in the abstract an informative and balanced summary of what was done and what was found	10 (100%)
INTRODUCTION			
Rationale	2	Explain the scientific background and rationale for the investigation being reported	10 (100%)
Objectives	3	State specific objectives, including any prespecified hypotheses	8 (80%)
METHODS			
Study design	4	Present key elements of study design early in the paper	8 (80%)
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	8 (80%)
Participants	6	a. <i>Cohort study</i> —Give the eligibility criteria, and the sources and methods of selection of participants. <i>Case-control study</i> —Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. <i>Cross-sectional study</i> —Give the eligibility criteria, and the sources and methods of selection of participants b. <i>Cohort study</i> —For matched studies, give matching criteria and number of exposed and unexposed <i>Case-control study</i> —For matched studies, give matching criteria and the number of controls per case	6 (60%)
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers.	7 (70%)
Data sources	8	For each variable of interest, give sources of data and details of methods of assessment.	5 (50%)
Bias	9	Describe any efforts to address potential sources of bias	3 (30%)
Study size	10	Explain how the study size was arrived at	6 (60%)
Quantitative variables	11	Explain how quantitative variables were handled in the analyses.	2 (20%)
Statistical methods	12	a. Describe all statistical methods, including those used to control for confounding b. Describe any methods used to examine subgroups and interactions c. Explain how missing data were addressed d. <i>Cohort study</i> —If applicable, explain how loss to follow-up was addressed <i>Case-control study</i> —If applicable, explain how matching of cases and controls was addressed <i>Cross-sectional study</i> —If applicable, describe analytical methods taking account of sampling strategy e. Describe any sensitivity analyses	8 (80%)
RESULTS			
Participants	13	a. Report numbers of individuals at each stage of study—e.g. numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analyzed b. Give reasons for non-participation at each stage c. Consider use of a flow diagram	4 (40%)
Descriptive data	14	a. Give characteristics of study participants (e.g. demographic, clinical, social) and information on exposures and potential confounders b. Indicate number of participants with missing data for each variable of interest c. <i>Cohort study</i> —Summarize follow-up time (e.g., average and total amount)	8 (80%)
Outcome data	15	<i>Cohort study</i> —Report numbers of outcome events or summary measures over time <i>Case-control study</i> —Report numbers in each exposure category, or summary measures of exposure <i>Cross-sectional study</i> —Report numbers of outcome events or summary measures	8 (80%)
Main results	16	a. Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (e.g., 95% confidence interval). b. Report category boundaries when continuous variables were categorized c. If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	5 (50%)
Other analyses	17	Report other analyses done—e.g. analyses of subgroups and interactions, and sensitivity analyses	6 (60%)
DISCUSSION			
Key results	18	Summarize key results with reference to study objectives	10 (100%)
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision.	9 (90%)
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	7 (70%)
General is ability	21	Discuss the general is ability (external validity) of the study results	5 (50%)
FUNDING			
	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	7 (70%)

Results

Eligible Studies

Our search strategy identified 289 potentially eligible studies involving the association between measles-mumps-rubella vaccine and autism, of which fourteen met the inclusion criteria (Fig. 1). The fourteen articles were published during the period from January 2006 through February 2016. A full list of the reports that were retrieved as full-text and included in the final analysis is found in the end of paper (Table 4). Were excluded immediately on inspection of the abstracts and title as they clearly did not meet inclusion criteria 36 papers (twenty article comment, letter and editorial, five historical article, four article news-paper, twelve review) (Table 5), leaving 53 papers whose methods sections were analyzed in more detail to determine suitability. Further, 39 full-text articles excluded because studied other factors of correlation, leaving fourteen article with usable information were included in this study.

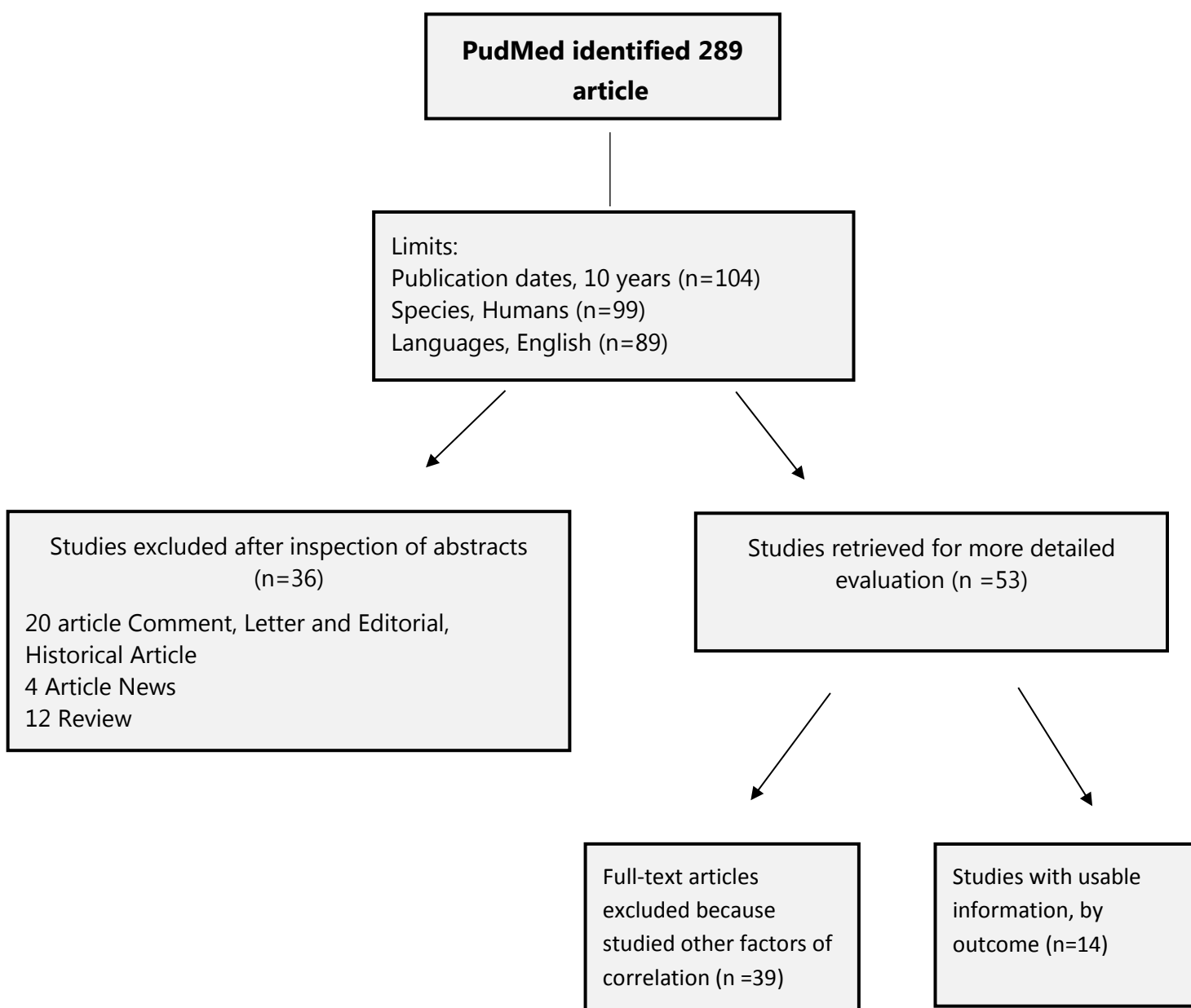


Figure 1. Flow diagram of citations through the retrieval and screening process

The fourteen eligible studies including: two meta-analysis (eight cohort studies, eight case-control studies, two time-series studies, two self-controlled case), two systematic review (six ecological studies, four retrospective observational studies, five prospective observational studies, four population studies), ten observation studies (eight case control studies, two cohort study). (Fig. 2)

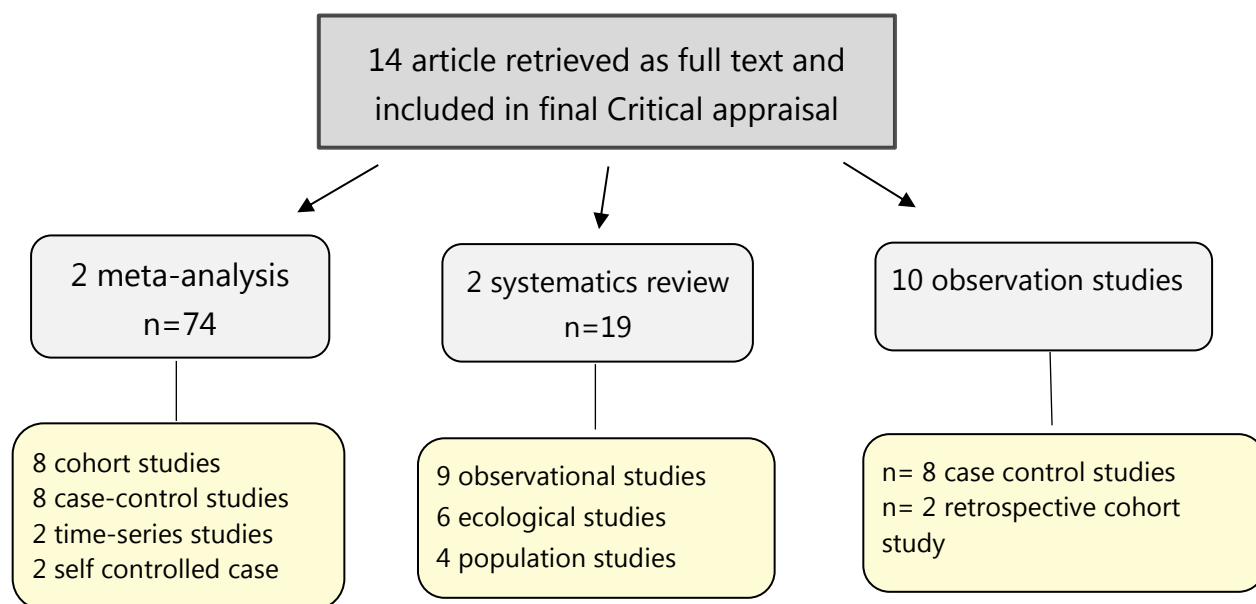


Figure 2. Flow diagram of citations through the retrieval and screening process

Main Results

This critical appraisal of two meta-analysis, two systematic review and ten observation studies data has found no evidence for the link between vaccination and the subsequent risk of developing autism or autistic spectrum disorder.

PRISMA was used for critical appraisal purposes of systematic review and meta-analysis. The checklist included 27 items pertaining to the content of a systematic review and meta-analysis, which include the title, abstract, methods, results, discussion and funding. These key questions were used to assess the validity and relevance of a research article. Also, assisted to identify the most relevant, high-quality studies that are available to guide their clinical practice. Overall, four items /sub-items (three and one items/subitems in methods and results sections, respectively) were reported by 100% of the studies (see Table 2). In methods, the items include the presentation of: 1) the key elements of protocol design and registration, 2) the eligibility criteria for participants (PICOS, length of follow-up, years considered, language, publication status) and 3) the information sources (e.g. databases with dates of coverage, contact with study). In results, the item include the presentation of the presentation of the study selection. Furthermore, seven items/sub-items (including the four items already mentioned above) were reported by 75% (three of four) of the studies. The six additional items were 1) the electronic search strategy, 2) study selection (i.e. screening, eligibility, included in systematic review, and, if applicable, included in the meta-analysis), 3) the method of data extraction from reports, 4) list and define all variables for which data were sought, 5) the principal summary measures (e.g. risk ratio, difference in means), 6) the synthesis of results and 7) the reporting of risk of bias across studies. In results, (including the four items already mentioned above) the item include the presentation of the results of additional analyses. In contrast, some items were reported only by a small fraction of articles. For example, only two of four of articles provided the results of any assessment of risk of bias across studies or the results of each meta-analysis done, including confidence intervals and measures of consistency, data on risk of bias of each study and, if available, any outcome level assessment. Also, only two of four of articles provided all outcomes considered present, for each study: a) simple summary data for each intervention group, b) effect estimates and confidence intervals. The presentation

of a flow diagram and reporting of absolute risk for a meaningful time period were very uncommon, refers to only one out of four of articles.

Table 2 shows the results sections of the STROBE statement. Overall, five items/sub-items (three and two items/subitems in methods and results sections, respectively) were reported in eight of ten of the studies (Table 3). In methods, the items include the presentation of: 1) the key elements of study design, 2) the setting, locations, relevant dates, including periods of recruitment, exposure, follow-up, and data collection and 3) the description of all statistical methods. In results, the items include: 1) descriptive data and 2) the details of outcome data. Furthermore, 11 items/sub-items (including the nine items already mentioned above) were reported in five or more of the studies. The six (four in methods and two results sections) additional items were, 1) the eligibility criteria, and the sources and methods of selection of participants, 2) the clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers, 3) the sources of data and the details of methods of assessment and 4) explain how the study size was arrived at, 5) the main results, 6) the report other analyses done - e.g. analyses of subgroups and interactions, and sensitivity analyses. In contrast, some items were reported only by a small fraction of articles. For example, only two of ten of articles provided quantitative variables and only three of ten describe any efforts to address potential sources of bias. The presentation of a flow diagram and reporting of absolute risk for a meaningful time period were very uncommon, only four out of ten articles present it.

Discussion

After the publication of Andrew Wakefield's research in 1998, which caused a great deal of confusion and debate, a lot of clinical researches were reported. In this paper, the majority of the researches come to a negative conclusion regarding the correlation between MMR and autism. Our analysis focused on the reporting of methodological items (items in method and results sections). In total, 14 articles published from January 2006 through February 2016 were evaluated. This critical appraisal of two meta-analysis, two systematic review, eight case-control and two cohort studies has found no evidence for the link between vaccination and the subsequent risk of developing autism or autistic spectrum disorder (Table 3).

Table 3. Studies that fail to support an association between MMR vaccine and autism.

negative results / title of articles
The combined measles, mumps, and rubella vaccines and the total number of vaccines are not associated with development of autism spectrum disorder: the first case-control study in Asia.
Measles vaccination and antibody response in autism spectrum disorders.
Early exposure to the combined measles-mumps-rubella vaccine and thimerosal-containing vaccines and risk of autism spectrum disorder.
Response to measles-mumps-rubella vaccine in children with autism spectrum disorders.
Lack of association between measles-mumps-rubella vaccination and autism in children: a case-control study.
Acetaminophen (paracetamol) use, measles-mumps-rubella vaccination, and autistic disorder: the results of a parent survey.
Vaccines for measles, mumps and rubella in children.
Vaccines are not associated with autism: an evidence-based meta-analysis of case-control and cohort studies.
Congenital rubella syndrome and autism spectrum disorder prevented by rubella vaccination--United States, 2001-2010
MMR-vaccine and regression in autism spectrum disorders: negative results presented from Japan.
Vaccines and the changing epidemiology of autism
positive results/ title of articles
Autism occurrence by MMR vaccine status among US children with older siblings with and without autism.
Autism occurrence by MMR vaccine status among US children with older siblings with and without autism.

The quality of reporting in meta-analysis systematic review and observation studies in MMR and autism was considered satisfactory, although certain items were underreported. The present study investigated the scientific validity and quality of published reporting of MMR and autism, according to the STROBE and PRISMA statement. We concluded that most of these researches used the appropriate study for the research question and were performed according to the original protocol. However, the study methods didn't address the most important potential of bias and any conflicts of interest. Still, the statistical analyses were performed correctly both in systematic statistical analyses and in OS.

Although the overall reporting quality was relatively good (18 items/sub-items were reported by three of four the meta-analysis, systematic reviews and 14 items/sub-items were reported by 70% or more of the studies), there are some essential methodological aspects of meta-analysis and systematic reviews (such as risk of bias in individual studies, risk of bias across studies, flow diagram, synthesis of results) that are seldom reported, making it difficult for the reader to assess explicitly the validity of an OS, SRs and meta-analysis. Also, the observation studies there were some essential methodological aspects which were referred in fewer research (such as sources of bias and quantitative variables).

There are several limitations to our study. We searched only in PubMed, which is the most common used medical database, for the eligible article and did not extent to the Cochrane Collaboration database to combine our results with one more sensitive search strategy. However, a more comprehensive literature search would be costly and time-consuming. In addition, trials which are difficultly retrieved tend to be of lower methodological quality and thus, bias could be introduced¹⁹. We considered only articles published in English, which could lead to language bias, since authors tend to publish article in English-language journals if the results are statistically.

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10. Singh VK. Rehabilitation of autism by immune modulation therapy. *Journal of Special Education and Rehabilitation*. 2004;3-4:161-178.
11. Rebuilding health systems to improve health and promote statebuilding in post-conflict countries: A theoretical framework and research agenda Margaret E. Kruk, Lynn P. Freedman, Grace A. Anglin, Ronald J. Waldman.
12. Latin America and the Caribbean: Assessment of the Advances in Public Health for the Achievement of the Millennium Development Goals. Amal K. Mitra and Gisela Rodriguez-Fernandez.
13. Hepatitis B Virus Infection: Epidemiology and Vaccination Colin W. Shepard, Edgar P. Simard, Lyn Finelli, Anthony E. Fiore and Beth P. Bell.
14. Vaccines for measles, mumps and rubella in children Vittorio Demicheli, Alessandro Rivetti,, Maria Grazia Debalini,, Carlo Di Pietrantonj
15. How to optimise the coverage rate of infant and adult immunisations in Europe Heinz-J Schmitt, Robert Booy, Robert Aston, Pierre Van Damme, R Fabian Schumacher, Magda Campins, Carlos Rodrigo, Terho Heikkinen, Catherine Weil-Olivier, Adam Finn, Per Olcén, David Fedson and Heikki Peltola.
16. Economics of Vaccine Development and Implementation: Changes Over the Past 20 Years Julie Milstien, Ph.D., and Brenda Candries, Ph.D.
17. Introduction of human papillomavirus vaccination in Nordic countries Bente Braad Sander, Matejka Rebolj, Palle Valentiner-Branth, Elsebeth Lynge
18. Wakefield A, Murch S, Anthony A, et al. Ileal-lymphoid-nodular hyperplasia, non-specific colitis, and pervasive developmental disorder in children. *Lancet*. 1998;351:637– 641.
19. Egger M, Jóni P, Bartlett C, Holenstein F and Sterne J: How important are comprehensive literature searches and the assessment of trail quality in systematic.

Table 4. Article included

PMID	TITLE	JOURNAL	YEAR	AUTHOR	
22521285	The combined measles, mumps, and rubella vaccines and the total number of vaccines are not associated with development of autism spectrum disorder: the first case-control study in Asia.	Vaccine	2012 Jun 13	Uno Y, Uchiyama T , Kurosawa M, Aleksic B, Ozaki N	case-control study
18252754	Measles vaccination and antibody response in autism spectrum disorders.	Arch Dis Child	2008 Oct	Baird G1, Pickles A, Simonoff E, Charman T, Sullivan P, Chandler S, Loucas T, Meldrum D, Afzal M, Thomas B, Jin L, Brown D.	case-control study
25562790	Early exposure to the combined measles-mumps-rubella vaccine and thimerosal-containing vaccines and risk of autism spectrum disorder.	Vaccine	2015 May 15	Uno Y, Uchiyama T, Kurosawa M, Aleksic B, Ozaki N	case-control study
23606694	Response to measles-mumps-rubella vaccine in children with autism spectrum disorders.	In Vivo	2013 May-Jun	Gentile I1, Bravaccio C, Bonavolta R, Zappulo E, Scarica S, Riccio MP, Settini A, Portella G, Pascotto A, Borgia G	case-control study
19952979	Lack of association between measles-mumps-rubella vaccination and autism in children: a case-control study	Pediatr Infect Dis J.	2010 May	Mrozek-Budzyn D1, Kiełtyka A, Majewska R	case-control study
18445737	Acetaminophen (paracetamol) use, measles-mumps-rubella vaccination, and autistic disorder: the results of a parent survey.	Autism.	2008 May	Schultz ST, Klonoff-Cohen HS, Wingard DL, Akshoomoff NA, Macera CA, Ji M	case-control, study
22336803	Vaccines for measles, mumps and rubella in children.	Cochrane Database Syst Rev	2012 Feb 15	Demicheli V, Rivetti A, Debalini MG, Di Pietrantonj C	metanalysis
17015560	No evidence of persisting measles virus in peripheral blood mononuclear cells from children with autism spectrum disorder.	Pediatrics	2006 Oct	D'Souza Y, Fombonne E, Ward BJ	Systematic Review
21592401	Congenital rubella syndrome and autism spectrum disorder prevented by rubella vaccination--United States, 2001-2010	BMC Public Health.	2011 May 19	Berger BE, Navar-Boggan AM, Omer SB	mathematici model
24814559	Vaccines are not associated with autism: an evidence-based meta-analysis of case-control and cohort studies.	Vaccine.	2014 Jun 17	Taylor LE 1 , Swerdfeger AL 1 , Eslick GD	meta-analysis
25898051	Autism occurrence by MMR vaccine status among US children with older siblings with and without autism.	JAMA	2015 Apr 21	Jain A, Marshall J, Buikema A, Bancroft T, Kelly JP, Newschaffer CJ	retrospective cohort study
16865547	MMR-vaccine and regression in autism spectrum disorders: negative results presented from Japan.	J Autism Dev Disord.	2007 Feb	Uchiyama T1, Kurosawa M, Inaba Y	Observational studies
19614825	Autism and vaccination-the current evidence.	J Spec Pediatr Nurs	2009 Jul	Miller L, Reynolds J	Systematic Review
19128068	Vaccines and autism: a tale of shifting hypotheses.	Clin Infect Dis.	2009 Feb 15	Gerber JS, Offit PA	Systematic Review
17928818	Vaccines and autism: evidence does not support a causal association.	Clin Pharmacol Ther.	2007 Dec	DeStefano F1	Systematic Review
16919130	Vaccines and the changing epidemiology of autism	Child Care Health Dev.	2006 Sep	Taylor B	Systematic Review
25086160	Safety of vaccines used for routine immunization of U.S. children: a systematic review.	Pediatrics	2014 Aug	Maglione MA , Das L , Raaen L , Smith A , Chari R , Newberry S	Systematic Review
26103708	Epidemiologic and Molecular Relationship Between Vaccine Manufacture and Autism Spectrum Disorder Prevalence.	Issues Law Med	2015	Deisher TA , Doan NV , Koyama K , Bwabye S	Systematic Review
19758536	Phenotypic expression of autoimmune autistic disorder (AAD): a major subset of autism.	Ann Clin Psychiatry	2009 Jul-Sep	Singh VK	Observational studies
21071320	Closer look at autism and the measles-mumps-rubella vaccine	Hensley E1, Briars L.	2010 Nov-Dec	J Am Pharm Assoc	Systematic Review

Table 5. Article excluded

PMID	TITLE	REASON
20142376	Lancet retracts 12-year-old article linking autism to MMR vaccines.	Biography, Historical Article, News
17075042	A surprising METamorphosis: autism genetics finds a common functional variant.	Comment
26192352	Answers regarding the link between vaccines and the development of autism: A question of appropriate study design, ethics, and bias.	comment letter
25898047	Promising forecast for autism spectrum disorders.	Comment, Editorial
21209060	Wakefield's article linking MMR vaccine and autism was fraudulent.	Comment, Editorial
17168157	Autism and MMR vaccination or thimerosal exposure: an urban legend?	Comment, Editorial
19176580	A response to the article on the association between paracetamol/ acetaminophen :use and autism by Stephen T. Schultz.	Comment, Letter
18762548	Immunization uptake in siblings of children with autism.	Comment, Letter
26757474	Correction of Description of MMR Vaccine Receipt Coding and Minor Errors in MMR Vaccine and Autism Study.	Comment, Letter
20432106	Setting the record straight: vaccines, autism, and the Lancet.	Editorial
10967744	MMR vaccine and autism.	Editorial
21985898	Epidemiological designs for vaccine safety assessment: methods and pitfalls.	Evaluation Studies, Review
25947030	Immunizing against influenza is tricky (Dummheit.)	Historical Article
21465869	Fallout of the enterocolitis, autism, MMR vaccine paper.	Historical Article
22930976	Lancet retracts study linking autism to MMR vaccine.	Historical Article
20222272	Debunked. A pivotal paper linking vaccines and autism is retracted. Will the antivaccine movement go on?	News
18451989	Attention focuses on autism.	News
17923648	Vaccine autism link discounted, but effect of "study" is unknown	News
19200293	The rise in autism and the mercury myth.	Research Support
22848999	Credibility battles in the autism litigation.	Research Support
23324619	Vaccine administration and the development of immune thrombocytopenic purpura in children.	Research Support
25612664	Spotlight on measles in Italy: why outbreaks of a vaccine-preventable infection continue in the 21st century.	Other studies/reviews
23449385	On alert for autism spectrum disorders.	Other studies/reviews
22108039	Acceptance on the move: public reaction to shifting vaccination realities.	Other studies/reviews
20653261	MMR vaccine and autism: is there a link	Other studies/reviews
20299908	Speak the language of autism.	Other studies/reviews
20030462	Did acetaminophen provoke the autism epidemic	Other studies/reviews
19015994	Autism overflows: increasing prevalence and proliferating theories.	Other studies/reviews
18771165	Update on autism and childhood vaccines	Other studies/reviews
19968949	Vaccines and autism: an update	Other studies/reviews
17894204	Autism and environmental influences: review and commentary.	Other studies/reviews
17181438	Postlicensure epidemiology of childhood vaccination: the Danish experience.	Other studies/reviews
17168158	Immunizations and autism: a review of the literature.	Comment
25523970	Measles outbreak in Greater Manchester, England, October 2012 to September 2013: epidemiology and control.	Other studies/reviews
22428439	I've heard some things that scare me". Responding with empathy to parents' fears of vaccinations.	Historical Article
21560548	Dilemmas of a vitalizing vaccine market: lessons from the MMR vaccine/autism debate	Article studying other factors of correlation
21387868	Autism and vaccines: search for cause amidst controversy.	Article studying other factors of correlation
21343697	MMR vaccination and autism: learnings and implications.	Article studying other factors of correlation
20944043	The autism-vaccine story: fiction and deception	Review

	Increasing immunization coverage.	Comment
19266897	Autism and vaccinations: is there a correlation	Comment
19213289	Autism spectrum disorders: prevalence and vaccines.	Comment
18726760	National vaccine injury compensation program: the potential impact of Cedillo for vaccine-related autism cases	Article studying other factors of correlation
18323140	Autism: part I. Deficits, prevalence, symptoms, and environmental factors.	Article studying other factors of correlation
17898095	Cases in vaccine court--legal battles over vaccines and autism.	Article studying other factors of correlation
17867721	A case study of a graphical misrepresentation: drawing the wrong conclusions about the measles, mumps and rubella virus vaccine.	Comment
22879643	Advertising watchdog orders website to remove claims linking MMR vaccine with autism.	News
21878599	MMR vaccine and autism: vaccine nihilism and postmodern science.	Comment
25724821	Autism and vaccination: The value of the evidence base of a recent meta-analysis.	meta-analysis
23229992	Dispelling vaccine myths: MMR and considerations for practicing pharmacists.	Systematic Review
18019187	Update on autism and vaccines.	Comment
19006807	Another study on the safety of measles vaccine and risks of autism.	Comment
17595690	Child development. An unexpected effect of the autism-vaccine controversy.	Comment
24590751		Article studying other factors of correlation
25002000	Childhood vaccine beliefs reported by Somali and non-Somali parents.	Article studying other factors of correlation
23045216	Immunization uptake in younger siblings of children with autism spectrum disorder.	Article studying other factors of correlation
22230590	U.K. parents' decision-making about measles-mumps-rubella (MMR) vaccine 10 years after the MMR autism controversy: a qualitative analysis	Article studying other factors of correlation
17540488	Children's health and the social theory of risk: insights from the British measles, mumps and rubella (MMR) controversy.	Article studying other factors of correlation
18019187	Current controversies in the USA regarding vaccine safety.	Article studying other factors of correlation
26596077	Addressing MMR Vaccine Resistance in Minnesota's Somali Community.	Article studying other factors of correlation
17395344	Tracking mothers' attitudes to MMR immunisation 1996-2006.	Article studying other factors of correlation
19813430	Parental vaccine concerns in Kentucky.	Article studying other factors of correlation
17376937	MMR: marginalised, misrepresented and rejected? Autism: a focus group study.	Article studying other factors of correlation
18381512	Media coverage of the measles-mumps-rubella vaccine and autism controversy and its relationship to MMR immunization rates in the United State	Article studying other factors of correlation
22496631	Evolutionary game theory and social learning can determine how vaccine scares unfold.	Article studying other factors of correlation
22063388	Lessons from an online debate about measles-mumps-rubella (MMR) immunization.	Article studying other factors of correlation
22236220	The blame frame: media attribution of culpability about the MMR-autism vaccination scare.	Article studying other factors of correlation
24857555	Science, pseudoscience, and the frontline practitioner: the vaccination/autism debate.	Article studying other factors of correlation