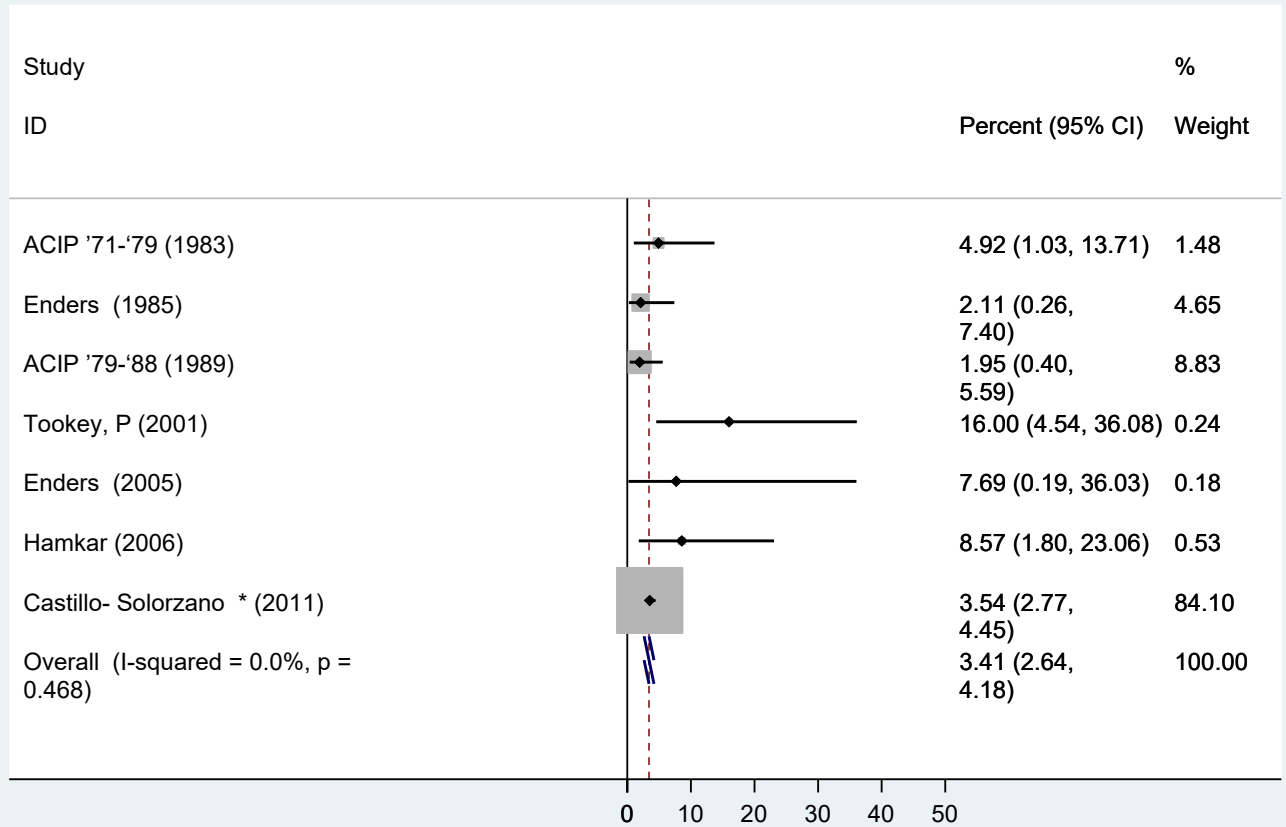
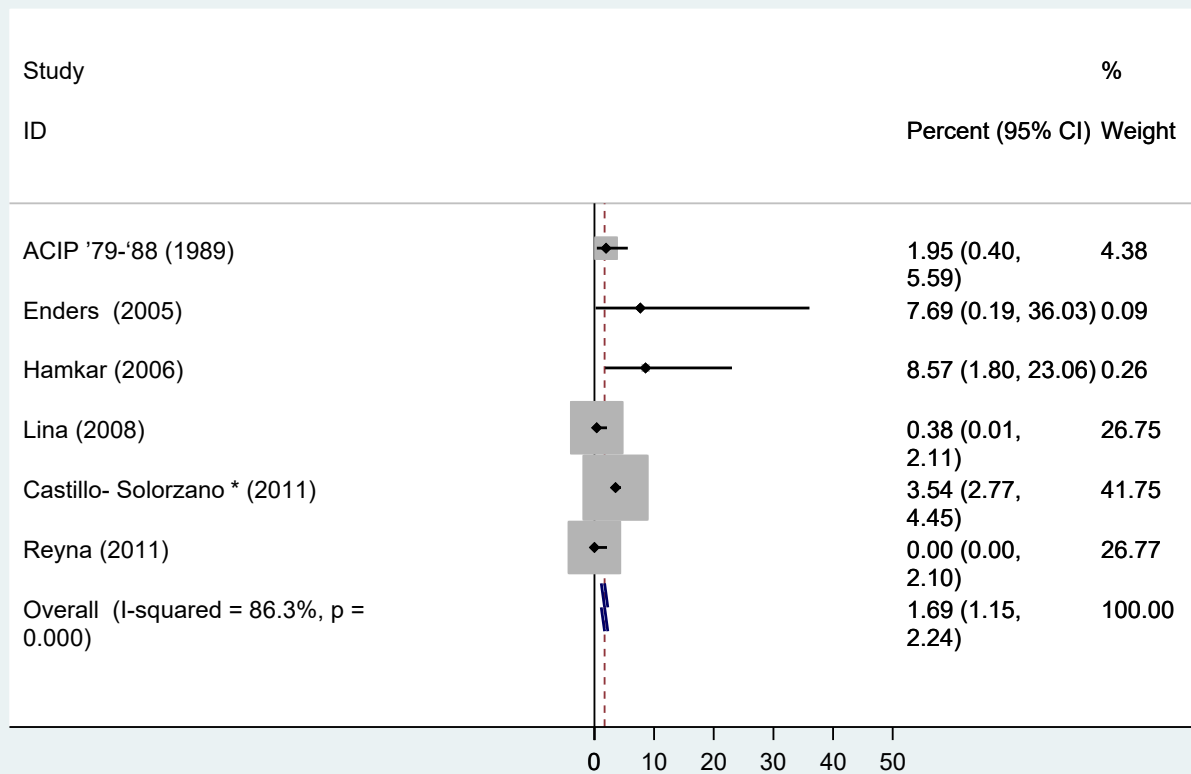


Appendix figure 1: Individual study risk of CRI and overall meta-analysis (fixed-effects) of the risk of CRI in pregnancy cohorts of women susceptible to rubella and vaccinated against rubella peri-conceptually: data from Reyna[1] and Lina[2] omitted.



Appendix figure 2: Individual study risk of CRI and overall meta-analysis (fixed-effects) of the risk of CRI in pregnancy cohorts of women susceptible to rubella and vaccinated against rubella peri-conceptually: data from Reyna[1] and Lina[2] omitted.

studies where other vaccines apart from RA27/3 was used (ie ACIP 1971-1979, Enders 1985 and Tookey 2001 omitted)



Appendix Table 1. Baseline characteristics of vaccinated pregnancy cohorts without information on outcomes stratified by susceptibility of mother to rubella

Author, year	Location, recruitment year(s), publication type	Vaccine R/MR/MMR, rubella strain, type of programme and target age range	N(%) susceptible, N(%) immune, N(%) unknown	N (%) vaccinated by timing of vaccination	N followed up for (CRS/CRI) By timing of vaccination	Type and N outcomes	Virological test for vaccine strain done?	Comments
Chin, J., 1971[3]	Los Angeles area USA, 1969- 1970, brief report	NK	5(29.4%) susceptible	Time of immunization ranged from 84 days before to 88 days after the last menstrual period 5 women vaccinated 12-4 weeks before LMP 4 women vaccinated < 4 weeks before LMP 8 women vaccinated up to 13 weeks after LMP	4 apparently normal infants by time of writing	6 therapeutic (concern about possible teratogenic effect of the vaccine) abortions, 1 spontaneous abortion, 4 apparently normal infants, 6 continuing pregnancies at the time of writing	Virological studies on the products of conception from some of the therapeutic abortions and from the apparently normal infants in progress, but results not available at the time of writing	
Larson, H. E., 1971 [4]	Laboratory of viral immunology, Maryland, USA, journal article	HPV-77(5) (3 women) HPV-77 (12) (1 woman) Unknown (5 women) Inadvertent vaccination during pregnancy: 7 women vaccinated by private physician, 2 by public vaccination program directed at prepubertal children	9 women- immune status unknown	8 women were vaccinated 7-81 days after LMP 1 woman was vaccinated on the day of her LMP	8 induced abortion	Virus was recovered from the products of conception of 2 women (out of 5 who received HPV-77). 1 had serologic confirmation of infection	yes 'Unmodified and the attenuated HPV-77 viruses were included in each test. Isolates from the vaccines produced prompt, marked cytopathic effects in "roller-tube "cultures of RK ₁₃ monolayers, and induced interferon. These findings were also characteristic of the HPV-77 attenuated virus and were in contrast to the behaviour of unmodified virus in the same test systems.' P. 872	1 pregnancy proceeded to term and despite prematurity, the baby had developed normally and was negative for virus.
Mair, H. J., 1972[5]	UK, 1970-1972, journal article	Inadvertent vaccination during pregnancy	6 women 2 mothers susceptible, 4 unknown	3 (50%) <1 month before LMP, 3 (50%) <1 month after LMP	Until termination of pregnancy (maximum follow up 19 weeks)	5 therapeutic abortions, 1 spontaneous abortion. Each product of conception examined virologically, with negative results.	NA	
Allan, B. C., 1973[6]	Australia, 1973, journal article	Cendehill , Inadvertent vaccination in pregnancy,	7 (11.1%) susceptible, 58 (89.2%) unknown	8: > 6 weeks before conception 53: 6 weeks before to 6 weeks after conception, 4: > 6 weeks of gestation	19 live infants by time of paper	0 CRS 1 virus isolation from placenta material, but no duplication possible, probably cross-contamination in the lab (susceptible mother) Examination of infants by	Unclear	

						paediatricians/specialists not explicitly mentioned		
Fleet, W. F., 1974 [7]	Tennessee, USA, journal article	NK	1 (5.3%) susceptible, 1 (5.3%) immune, 17 (89.5%) unknown	Weeks before conception: 9-12 = 3(18.7%) 5-8= 3(18.7%) 0-4=4(25%) After 0-4=3(18.7%) 5-8=1 (6.2%) 9-12= 1(6.2%) 13-24= 1(6.2%)	10 liveborn children, 9 throat swabs available + 5 urine: no virus isolated in specimen: FU studies on 9 children for 3 to 11 months physical exam normal	9 abortions, 6 were in those vaccinated within 4 weeks of conception. 1 was a spontaneous abortion and 7 were induced abortions. No virus was isolated in these 8 abortions. In the 9 th , in a mother vaccinated 7 weeks after conception, an “interfering” agent was isolated from the eye suspension of the fetus after an induced abortion. Congenital rubella infection of eye, from a virus-positive vaccine strain was concluded by the authors.	Yes in eye, based on in-vitro growth characteristics of all cultures	Results of physical examinations, Denver Development Screening Tests and hearing tests were normal in all infants
Marks, J. S., 1981 [8]	Hawaii, journal article	Inadvertent vaccination during pregnancy: 23 vaccinated at public clinics 17 vaccinated by private physicians	40 women	NK	NK	28 (70%) elective abortion 1 spontaneous miscarriage 11 delivered normal children	No information	
Nasiri, R., 2009 [9]	Islamic Republic of Iran, 2003, journal article	MR vaccine, RA27/3 strain, Nationwide campaign, 5-25year olds	60 pregnant women	All women vaccinated in 1-4 week periconceptional period	60 newborns had physical examined at birth and at 1 month of age by paediatrician. All neonates had umbilical cord blood tested	0/60 CRS (None of the neonates had evidence of IUGR, cardiovascular, ophthalmic, CNS, or other organ system anomalies) 0/60 CRI 4/60 preterm	NA (no CRI)	35 (65%) pregnant women had no history of rubella infection, 7 (11.7%) had previous history of rubella, 14 (23.3%) had no clear history
Ergenoglu A. M., 2012 [10] ,	Turkey Began in 2009, journal article	R-VAC, Keymen Pharmaceuticals* National campaign targeted at women aged 18-35, *Most likely RA27/3	62 women Immune status not known. Serological evaluation of mothers prior to vaccination was not possible, but all mothers IgG(+).	During first trimester of pregnancy or within one month before last menstrual period (application during first trimester)	17 followed up until end of pregnancy, normal deliveries	IgM and IgG antibodies assessed by rubella ELISA IgM and IgG kits at first visit after vaccination of women and from fetal cord blood at birth . Only 1 woman IgM weakly positive. Rubella-specific IgM antibodies testing of cord blood negative in all cases. None of the neonates had clinical evidence of intrauterine growth restriction or cardiovascular, ophthalmologic, central nervous system or other system abnormalities. All auditory screenings were normal.	NA	

Appendix table 2. Baseline table with study characteristics (Case series, Case studies)

First Author, year	Location	Time period of study	Vaccine R/MR/MMR, rubella strain	N (%) vaccinated by timing of vaccination	Length and type of follow up	Type and N outcomes	Virological test for vaccine strain done?	Comments
Phillips, C. A, 1970 [11]	US	1970	NK	1, 3 weeks of gestation in a susceptible woman	Pregnancy terminated at 8 weeks. Tissue specimens obtained at surgery.	1 case of vaccine induced intrauterine rubella infection	Yes	Interferon assays carried out in cultures were higher than that seen with wild type virus and lower inhibition of in vitro lymphocyte transformation activity than wild type virus both consistent with vaccine virus
Ebbin, A. J. 1972 [12]	US	1972	Cendehill	55 days after LMP	Pregnancy terminated 98 days after vaccination	HI titre 1970 <1/8. After vaccination (82 days) HI titre 1/512. No evidence of congenital abnormality. Virus cultured only from femoral bone marrow, not from other organ systems or placenta	No	Viral culture according to "standard methods" in "RK-13 and BSC-1 cell lines"
Giles P. F. H., 1973 [13]	Australia	1970-1972	Cendehill	1, 3 days after LMP	NK	Baby delivered at 39 weeks gestation with no evidence of congenital abnormality. No CRI at testing 6 weeks after delivery	No	Viral culture done placental material negative
Colombo, M. L., 1976 [14]	Italy	1973	NK	1, >3 months after conception	13.5 months, active follow up	Baby born with malformations and low birth weight. At 10 months was diagnosed with some clinical features compatible with CRS; deafness, mental retardation as well as thorax and tibial deformity with no movement lower legs.	No	No laboratory tests mentioned
Banatvala, J. E., 1981 [15]	UK	1980	RA27/3	1, 2 weeks after conception	Pregnancy terminated at week 12	Products of conception cultured in RK 13 rubella virus antigen detected by indirect immunofluorescence, no rubella specific IgM detected	No	Not stated, presumed not done
Higaki, Y 1989 [16]	Japan	1987	Takahashi (The Kitasato Institute)	Within 6 weeks of conception	Followup to 9 months of age	Baby delivered at 41 weeks by caesarean section with no clinical evidence of CRS. HI rubella antibody test <1:8 and IgG and IgM Elisa antibodies negative at 9 months	No	No information of bloods done at birth or if rubella IgM in cord blood tested.
Hofmann, J., 2000 [17]	Germany	1998-2000	RA27/3	1 case of intrauterine infection, mother vaccinated 3 weeks after conception.	Virus isolation in week 16. 6 further ultrasounds (until week 36). Physical examinations of infant until 14 months	At week 16 virus isolation and PCR in amniotic fluid and fetal blood was positive for vaccine strain rubella virus. Healthy baby delivered at 40 weeks, no anomalies in physical examinations,	Yes including PCR tests	5 other women with live births and no CRI or CRS. No mention of how identified or if reported elsewhere

				5 other susceptible women mentioned		hearing tests inconspicuous. Regular development until 14 months of age.		
Sukumaran, L. , 2015[18]	US,	2003-2013	MMR	131, immune status not known 82 [62.6%] in the first trimester		Data mining of spontaneous reports VAERS database. No unusual patterns with regard to pregnancy and pregnancy outcomes	NA	Spontaneous reports from clinicians and public to VAERS database. MMR receipt and its associated reported outcome was examined and data mining based on a Bayesian method of comparison to spontaneous reports for all other vaccines.
Korkmaz, H.A., 2015[19]	Turkey	2013	Unclear	10 gestational weeks	Active follow-up up to six months of age regarding serology	At 1 months of age positive IgG, at 2 months of age positive IgM, at 6 months of age negative IgM, no virus PCR performed, no further anomalies, but raised liver enzymes, returning to normal at 2 months of age	No	

Appendix table 3. Studies of pregnant women scheduled for induced abortion who agreed to be vaccinated beforehand

Author, year	Location, recruitment year(s), publication type	Vaccine R/MR/MMR, rubella strain	N(%) susceptible, N(%) immune, N(%) unknown	N (%) vaccinated by timing of vaccination	N followed up for (CRS/CRI) By timing of vaccination	Type and N outcomes	Virological test for vaccine strain done?	Comments
Furukawa, T., 1969 [20]	Nagoya, Japan	RA 27/3 (10 women) Cendehill (5 women)	7 susceptible 8 borderline	All women were vaccinated in 1 st trimester	NK	1 spontaneous abortion, no other adverse effects. Examination of the products of conception for presence of rubella virus and for histologic abnormalities was negative in all cases. The results of all tests, including those of suspensions, of deciduas, chorion, and fetus, were negative.		volunteers from an abortion clinic with low or absent HI antibody vaccinated.
Vaheri, A., 1972 [21]	Finland, 1969, journal article	HPV-77(12) (29 women), HPV-77(5) (6 susceptible women) Women scheduled for abortion	35 women vaccinated	35 (100%) post conception, 14 weeks maximum gestational age at vaccination	35 followed up with interview at 6 weeks after vaccination. Serum specimens were obtained from each subject before vaccination, 2-3 weeks later and again 6-9 weeks after vaccination. Throat and nasopharyngeal swabs for virus isolation were collected from 8-36 days after vaccination.	13/35 virus isolated from throat or nasopharyngeal swab 13/22 virus isolated from cervical swab 5/24 viremia in those who seroconverted 2/15 virus in appendix 6/35 placental tissue virus isolations	Not reported	A single virus isolation was made from fetal tissue. This was obtained from the kidney homogenate of a fetus obtained from a 22-year-old woman vaccinated on the 55 th day of gestation who had an induced abortion 24 days later. Virus was recovered in two laboratories and not in the third.' p.1072 Preliminary findings presented in Vaheri, A. 1969[22]
Bernstein, D.I., 1973 [23]	US	RA27/3	Several women, all susceptible (no details of numbers given)	'Several' women were vaccinated 6-10 weeks after conception (and 10-14 weeks before scheduled abortions)	Products of conception examined via 'techniques of tissue culture infection, immunofluorescence and hemagglutination-inhibition'.	No rubella antibody activity or infectious virus was recovered in the fetal or placental tissues.	NA	Immunofluorescence revealed the presence of rubella antigen in one placenta. However, no antigen was found in fetal tissues
Bolognese, R. J., 1974 [24]	US, 1972-1973, Journal article	Cendehill, HPV77, HPV77.DE5, Merck As above HPV77DK12, Philips-Roxane Cendehill Smith-Kline RA27/3, Merck	16 (38.1%) susceptible 24 (57.1%) immune 2 (4.76%) borderline (prevaccination titre 8)	42 (100%) 1st trimester	40 women were followed up until the abortion. 2 women gave birth and pregnancy outcome recorded	CRS:0 Immune: 0/2, 2 normal term infants have been delivered to immune mothers Intrauterine infection: Susceptible: 3/16 (18.75%) (virus isolated in abortus material)	Yes, by injecting isolated virus and Freedman 'wild' strain rubella virus into ear vein of 4 New Zealand white rabbits	When virus isolated in 2 cases was injected into rabbits, titres identical with those of the "wild" strain were recorded.
Appendix table 4 Types of rubella vaccines (supplementary information)								
		Strain derivation	Attenuation	Licensure				
		Army recruits with rubella (1961) vaccinated	AGMK (77)*, monkey cell strain AGMK (77); duck embryo (5)	1969				
		As above	AGMK (77); dog kidney (12)	1969				
		Urine of a case of post natally acquired rubella (1963)	AGMK (3); primary rabbit kidney (51)	1969				
		Kidney of rubella-infected fetus (1964)	Human embryonic kidney(4); WI-38 fibroblast (17-25)	1979				

*Figures in parentheses indicate number of passages.

Appendix table 5. Risk of bias for cohort studies of rubella vaccinated susceptible pregnant women

Author	Risk of bias due to losses to followup?	Risk of reporting bias
ACIP '71-'79[25]	Moderate	low
ACIP '79-'88[26]	Moderate	low
Badilla, X.[27]	serious	moderate
Da Silva e Sa, G.R.[28]	Serious	moderate
Emadi, H.[29]	moderate	moderate
Hamkar, R.[30]	Serious	moderate
Lina[2]	Serious	moderate
Pardon, F.[31]	moderate	moderate
Reyna[1]	Moderate	moderate
Sato, H. K.[32]	moderate	moderate
Sheppard, S[33].	moderate	low
Castillo-Solorzano, C., 2011 ⁴³	See individual studies	See individual studies

Appendix 2 example of a full search in Medline_

Search included: Ovid MEDLINE(R), Ovid MEDLINE(R) In-Process & Other Non-Indexed Citations, Ovid MEDLINE(R) Daily and Ovid OLDMEDLINE(R) 1946 to Present

1. Pregnant wom?n.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
2. Maternal.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
3. Mother*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
4. Antenatal.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
5. Wom?n.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
6. Gestation*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
7. Gravid*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
8. Female adolescent*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
9. Prenatal care.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
10. Preconception*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
11. Perinatal.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
12. Maternal health.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
13. Pregnancy.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
14. mother/
15. pregnant woman/
16. exp maternal welfare/
17. exp pregnancy/

18. 1 or 2 or 3 or 4 or 5 or 6 or 7 or 8 or 9 or 10 or 11 or 12 or 13 or 14 or 15 or 16 or 17
19. Vaccin*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
20. Immuni*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
21. Inoculat*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
22. Trivalen*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
23. Combin*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
24. Simultan*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
25. Tripl*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
26. Trebl*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
27. exp Vaccines, Attenuated/
28. exp Vaccines, Combined/
29. 19 or 20 or 21 or 22 or 23 or 24 or 25 or 26 or 27 or 28
30. rubella.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
31. German measles.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
32. MMR*.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
33. Triviraten Berna.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
34. Priorix.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
35. Trimovax.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
36. Morupar.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]
37. MERUVAX.mp. [mp=title, abstract, original title, name of substance word, subject heading word, keyword heading word, protocol supplementary concept, rare disease supplementary concept, unique identifier]

38. exp rubella/
 39. 30 or 31 or 32 or 33 or 34 or 35 or 36 or 37 or 38
 40. 29 and 39
 41. 18 and 40

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