Technical report

Volume 3 – Additional literature

Prepared for
National Blood Authority

Project
Update of the 2003 Guideline on the Prophylactic Use of Rh D Immunoglobulin (Anti-D) in Obstetrics

Technical report prepared by
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Note

This volume ('the 2021 udpate') presents additional literature published and identified after a systematic literature review on use of Rh D Immunoglobulin (Anti-D) in RhD negative pregnant women. Volume 1 presents the main body of evidence. Volume 2 present the appendixes (Appendix A to Appendix F) that document the evidence synthesis (published in 2018). Together the three volumes cover all research questions and evidence reviewed for this topic.

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Findings of the systematic review

Results of the literature search

The medical literature was searched on 27-28 September 2021 to identify relevant studies and systematic reviews published between 2018 to the literature search date. Searches were conducted of the databases and sources described previously (**Section 3.2, Technical report, Volume 1**). Manual searches of the reference lists of relevant articles were also performed.

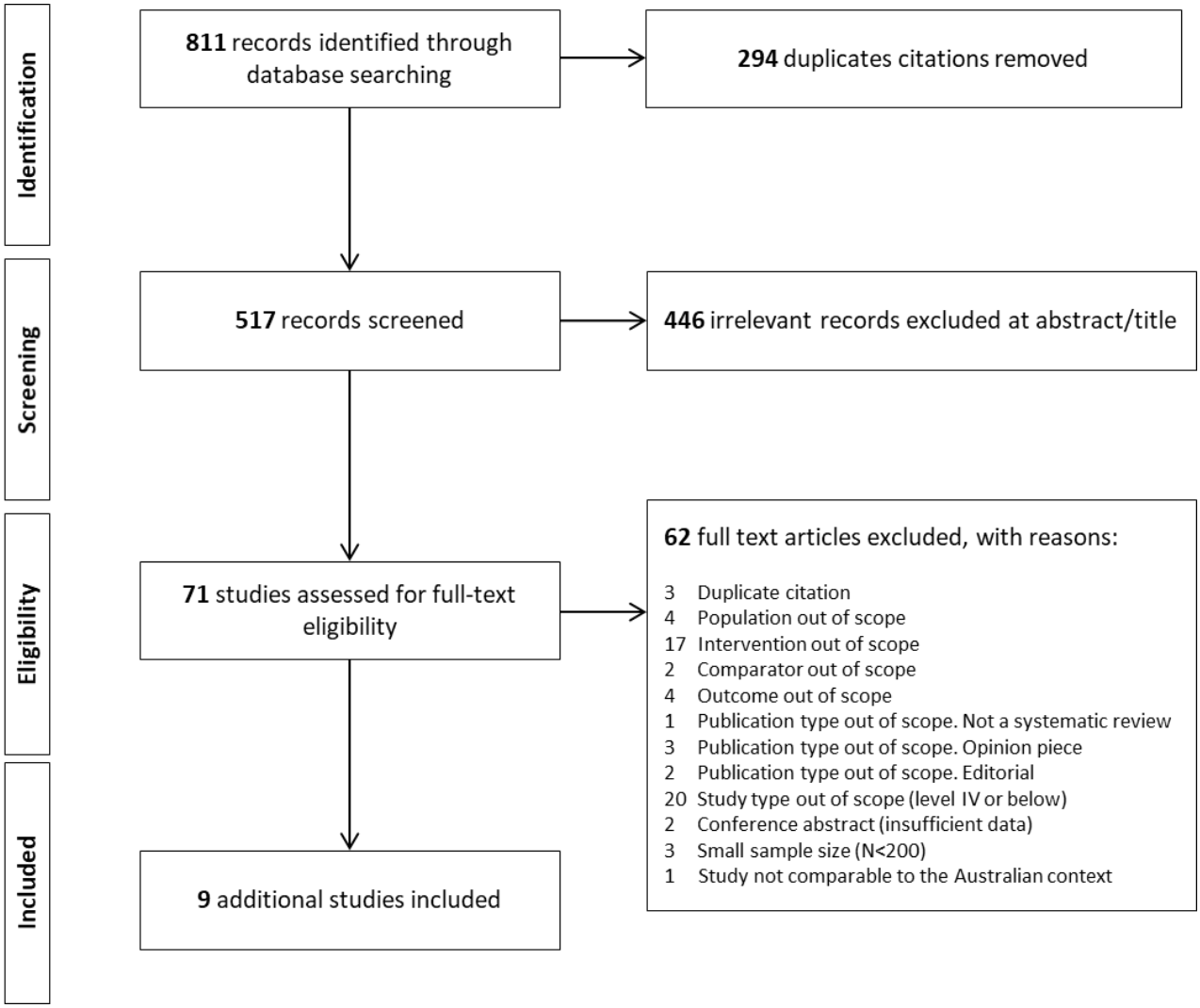
Search terms are as described in **Appendix A**, with methodological filters applied to identify specific study types. Studies were excluded based on hierarchical, prespecified exclusion criteria as described previously (see **Technical report, volume 1**), with all citations returned by the literature searches reviewed based on information in the publication title and, where available, the abstract. Relevant publications were retrieved and reviewed in full text before a final decision was made on their inclusion or exclusion for the review. The expert group was consulted in cases where further judgement was required.

The results of the screening process and the application of the study selection criteria is provided in **Appendix B**. A PRISMA flow summarising the screening results is provided in **Figure 1** (all questions) and **Figure 2** (subquestion 3, diagnostic accuracy).

A total of 12 new studies were identified and included in the review (Alshehri, 2021, Jernman, 2021, Legler, 2021, Ontario Health, 2020, Parchure, 2021, Pazourkova, 2021, Runkel, 2020, Schmidt-Hansen, 2020, White, 2019, Wikman, 2021, Xie, 2020, Yang, 2019).

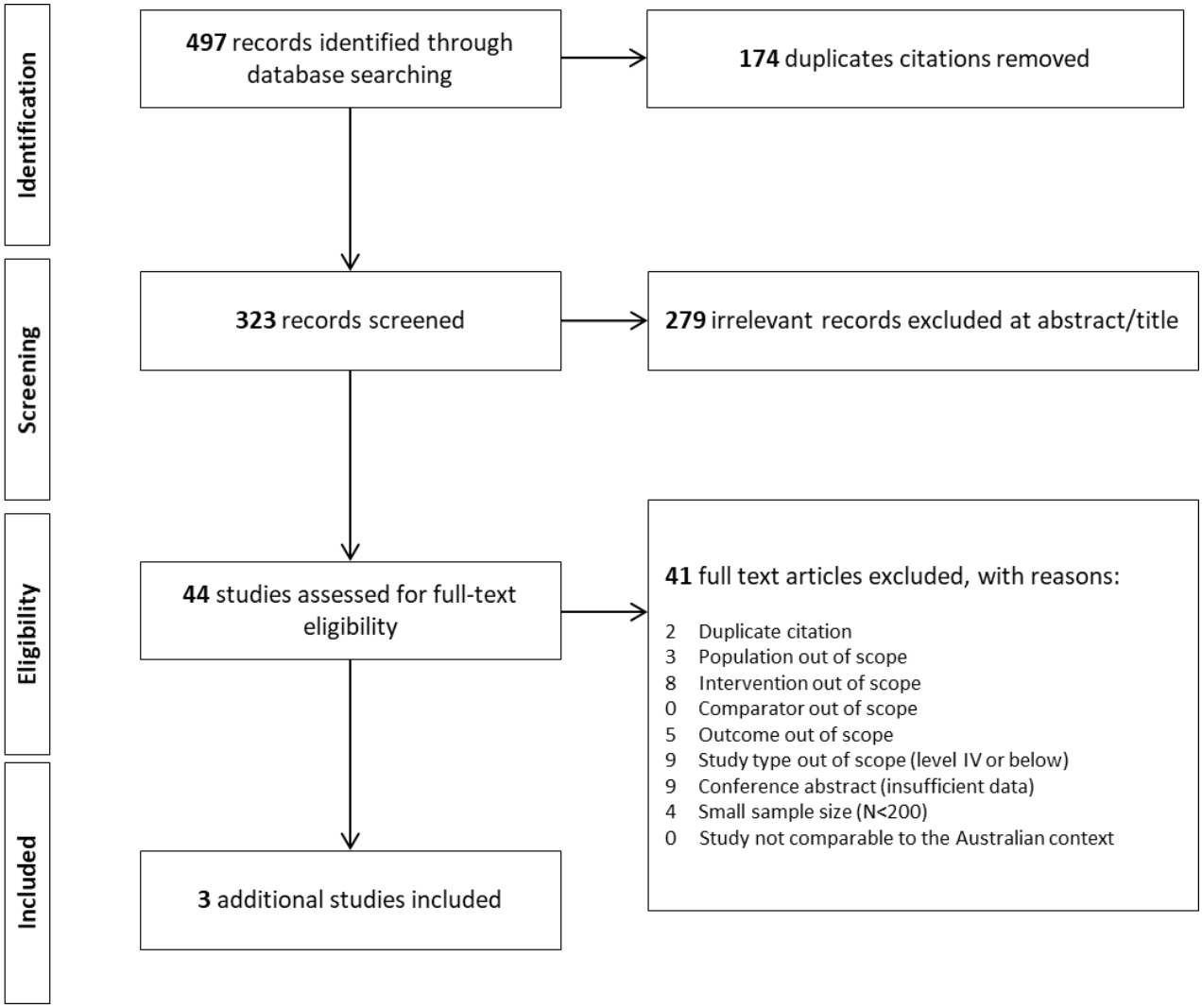
Studies that technically met the inclusion criteria (or potentially) but were later excluded (e.g., contained insufficient or inadequate data for inclusion, were considered incompatible with the Australian context) are listed in **Appendix C**.

Figure 1 Literature screening results. Questions 1 to 4.



Search conducted 27-28 Sept 2021, including Embase, MEDLINE, PubMed, Cochrane and CINAHL

Figure 2 Literature screening results. Questions 3.



Search conducted 27-28 Sept 2021, including Embase, MEDLINE, PubMed, Cochrane and CINAHL

Question 1 - *Routine* antenatal Rh D immunoprophylaxis

Question 1 – (Intervention)

In Rh D negative pregnant women with no preformed anti-D, does universal *routine* antenatal prophylaxis with Rh D immunoglobulin (1 or 2 doses) prevent Rh D alloimmunisation?

Subquestion 1 – (Intervention)

In Rh D negative pregnant women with no preformed anti-D, is universal *routine* antenatal prophylaxis with one dose of Rh D immunoglobulin as effective at preventing Rh D alloimmunisation as universal *routine* prophylaxis with two doses of Rh D immunoglobulin?

Background

The 2018 review identified four systematic reviews (Chilcott, 2003, McBain, 2015, Pilgrim, 2009, Turner, 2012) and one Level III study (Koelewijn, 2008) that evaluated the effectiveness of RAADP in Rh D negative women. The reviews identified two Level II studies (Huchet, 1987, Lee, 1995) and nine Level III studies (Bowman, 1978, Bowman, 1978, 1987, Hermann, 1984, MacKenzie, 1999, Mayne, 1997, Parsons, 1998, Tovey, 1983, Trolle, 1989) meeting their search criteria.

Summary of evidence

The 2021 update found one additional systematic review (Xie, 2020) that evaluated the effectiveness of RAADP in Rh D negative women. One Level II study (White, 2019) was also included that reported on serum anti-D antibody levels in Rh D negative women who had received one or two doses of RAADP.

Xie 2020 was a network meta-analysis that examined varying doses of Rh D immunoglobulin compared to no treatment in Rh D negative women. The authors searched multiple databases (including a Chinese database) up to 7 July 2019 and included studies that examined both antenatal and postnatal administration Rh D immunoglobulin that were published between 1958 and 2004. Doses of Rh D immunoglobulin administered varied between a single dose (250 µg) at 28 weeks through to two doses (300 µg) at 28 and 34 gestational weeks, with or without administration of 100 to 300 µg up to 72 hours postnatally. No new studies were found. Treatments were ranked using surface area under the curve analysis of cumulative probability of preventing Rh D alloimmunisation.

White 2019 is the published report of the Australian trial previously included in the 2018 review (see Pennell 2017 conference abstract). White 2019 compared two doses of Rh(D) immunoglobin-VF 625 (IU) administered at 28 and 32 weeks' gestation with a single dose of 1500 IU given at 28 weeks' gestation. Recruitment occurred through randomising Rh D pregnant women who intended to give birth at a tertiary obstetric referral hospital in Perth between May 2013 and November 2015. The main outcome assessed was the presence of Rh(D) immunoglobin antibodies in maternal blood at the time of delivery.

Results

Incidence of Rh D alloimmunisation

As reported by previous SRs, the network meta-analysis by Xie 2020 also showed an effect favouring RAADP compared to no treatment in preventing Rh D alloimmunisation. The analyses included different doses and timing of Rh D immunoglobin but favoured RAADP in all cases (odds ratio ranging from 0.00 to 0.15).

Based on analysis of the surface area under the cumulative ranking curve (SUCRA), Xie 2020 suggested that two dose of 1500 IU of Rh D immunoglobulin given at 28 and 34 gestational weeks' is better than other dosing regimens (SUCRA = 96.8%), with the second alternative being a single dose (1500 IU) given at 28

gestational weeks (SUCRA = 89.2%), followed by two doses (500 IU) given between 28 and 34 gestational weeks (SUCRA = 75.1%).

Serum anti-D antibody levels

White 2019 reported similar numbers to that reported in the 2018 review, noting that the number of women with anti-D antibody present at birth was higher in those women who received the two-dose regime compared to the one-dose regimen (86% v 56%; OR 4.91; CI 2.67, 9.02; $p < 0.001$). Concerns about the effect estimate exist, relating to missing antibody screening data (8%) and that twelve women in the single dose group (9%) received an incorrect dose (625 IU) at 28–30 weeks and were therefore given a second dose at 34–36 weeks to avoid potential late antenatal sensitisation.

As previously noted, the relationship between a lack of detectable circulating anti-D antibody following Rh D immunoprophylaxis and risk of alloimmunisation detected in a subsequent pregnancy is not known.

Discussion

If and how the 2021 search has impacted on evidence base?

The 2021 search provided two additional studies relevant to Question 1 (Xie 2020 and White 2019). Both studies provided solidified the existing evidence in favour of issuing universal routine antenatal Rh D immunoglobin to prevent Rh D alloimmunisation.

If and how the 2021 search has created changes in the evidence?

The studies found did not conflict or contradict any of the existing evidence, therefore no changes should be made to the 2018 recommendations. Questions regarding the effectiveness of a single dose of Rh D immunoglobulin compared to two doses remain unanswered.

Question 2 - Universal sensitising event prophylaxis in the first trimester

Question 2 – (Intervention)

In Rh D negative women with no preformed anti-D who have experienced one of the following first trimester sensitising events – abdominal trauma, molar pregnancy, ectopic pregnancy, spontaneous miscarriage, threatened miscarriage or medical termination of pregnancy (with/without a curette) – does universal first trimester sensitising event prophylaxis with Rh D immunoglobulin prevent Rh D alloimmunisation?

Background

The 2018 review identified two systematic reviews (Karanth, 2013, NCCWCH, 2012) that evaluated the effectiveness of prophylactic Rh D immunoglobulin in response to a first trimester sensitising event. The reviews included one Level II study (Visscher, 1972) and two Level III studies meeting the PICO criteria (Gavin, 1972, Simonovits, 1974).

No additional studies evaluating the use of prophylactic Rh D immunoglobulin in women with first trimester ectopic pregnancy, threatened miscarriage, or molar pregnancy were found.

Summary of evidence

The 2021 update found one additional systematic review (Schmidt-Hansen, 2020) that searched for evidence relating to sensitising events in women undergoing either medical abortion with mifepristone and misoprostol or surgical abortion using vacuum aspiration of a pregnancy up to 13^{+6} weeks' gestation. The review was used to inform the 2019 NICE guidelines on abortion care (NICE, 2019).

Results

In the absence of evidence, the following expert consensus guide was developed:

- Offer anti-D prophylaxis to women who are rhesus D negative and are having an abortion after 10^{+0} weeks' gestation.
- Do not offer anti-D prophylaxis to women who are having a medical abortion up to and including 10^{+0} weeks' gestation.
- Consider anti-D prophylaxis for women who are rhesus D negative and are having a surgical abortion up to and including 10^{+0} weeks' gestation.
- Providers should ensure that:
    - rhesus status testing and anti-D prophylaxis supply does not cause any delays to women having an abortion
    - anti-D prophylaxis is available at the time of the abortion.

Discussion

If and how the 2021 search has impacted on evidence base?

The 2021 search provided one additional systematic review relevant to Question 2 (Schmidt-Hansen 2020), which found no new evidence relating to administration of antenatal Rh D immunoglobin to prevent Rh D alloimmunisation in women undergoing medical or surgical abortion.

If and how the 2021 search has created changes in the evidence?

No new studies were found therefore no changes should be made to the 2018 recommendations. In the absence of evidence, the precise benefits and risks of anti-D prophylaxis relating to medical termination of pregnancy before 10 weeks of gestation remain unclear.

Question 3 - *Targeted* routine antenatal or sensitising event prophylaxis

Question 3 – (Screening intervention)

In Rh D negative pregnant women with no preformed anti-D, does *targeted* routine antenatal or sensitising event prophylaxis to women with a Rh D positive fetus increase the incidence of Rh D alloimmunisation compared with *universal* routine antenatal or sensitising event prophylaxis?

Subquestion 3 – (diagnostic accuracy)

In Rh D negative pregnant women with no preformed anti-D, what is the diagnostic accuracy of noninvasive prenatal screening to identify fetal Rh D status?

Background

The 2018 review identified one systematic review (Saramago, 2018) that searched for evidence regarding the comparative effectiveness of targeted antenatal Rh D immunoprophylaxis against universal routine Rh D immunoprophylaxis. The report did not identify any head-to-head studies of targeted versus routine antenatal prophylaxis regimes that met the criteria for the review.

There were four systematic reviews that examined the diagnostic accuracy of NIPT to identify fetal Rh D status (Geifman-Holtzman, 2006, Mackie, 2017, Saramago, 2018, Zhu, 2014). The reviews included over 90 studies meeting their search criteria. Five additional Level II studies (Haimila, 2017, Macher, 2012, Manfroi, 2018, Moise, 2016, Picchiassi, 2015) and six additional Level III study (Hyland, 2017, Jakobsen, 2018, Orzińska, 2015, Papasavva, 2016, Ryan, 2017, Sorensen, 2018) were identified and subsequently included in the evidence review. Studies that were of small sample size (N<200), conference abstracts that did not provide sufficient data, and those in which the NIPT was not conducted in the context considered similar to Australia were excluded (see **Technical report, volume 1**).

Summary of evidence

The 2021 update found four systematic review that searched for evidence regarding the comparative effectiveness of targeted RAADP against universal RAADP and/or examined the diagnostic accuracy of NIPT to identify fetal Rh D status (Alshehri, 2021, Ontario Health, 2020, Runkel, 2020, Yang, 2019). Three of the reviews were published reports of health technology assessments used to inform the Canadian Agency for Drugs and Technologies in Health (Ontario Health, 2020) the German Institute for Quality and Efficiency in Health Care (Runkel, 2020) and the NHS (Yang, 2019). Alsheri 2021 was a systematic review focused on the diagnostic accuracy of NIPT to identify fetal Rh D status. The authors identified 16 studies, 11 of which were included in a meta-analysis.

One additional Level III study was identified that examined the effectiveness of targeted antenatal Rh D immunoprophylaxis against no routine prophylaxis (Jernman, 2021). Jernman 2021 reported the results of a nationwide cohort study conducted in all pregnant women with anti-D antibodies detected in the Finnish Red Cross (FRC) Blood Service between January 1, 2014 and December 31, 2017.

Two Level II studies (Parchure, 2021, Pazourkova, 2021) and one Level III study (Legler, 2021) that examined the diagnostic accuracy of NIPT to identify fetal Rh D status were also identified and included in the evidence review. Studies that were of small sample size (N<200), conference abstracts that did not provide sufficient data, and those in which the NIPT was not conducted in the context considered similar to Australia were excluded (see **Appendix C**).

Results

Incidence of Rh D alloimmunisation

Similar to the evidence found previously, the data reported in the SRs and that reported by Jernman 2021 suggests that the risk of Rh D alloimmunisation is lower in the cohort that received targeted RAADP compared with the historic reference cohort that received postnatal and antenatal Rh D immunoglobulin prophylaxis following any potentially sensitising events.

Utilisation of Rh D immunoglobulin

The Ontario health report noted that across studies, 25.3% to 39% of all Rh D negative pregnancies avoided unnecessary Rh D immunoglobulin after noninvasive fetal RhD blood group genotyping. Among the Rh D negative mothers carrying an Rh D negative fetus, over 90% avoided unnecessary Rh D immunoglobulin.

Diagnostic performance

Similar to the evidence reported in 2018, the data reported in the SRs and newly included studies suggests that the diagnostic performance of NIPT to identify fetal Rh D status is good, with the bivariate analysis reported by Runkel 2020 (12 studies, 60 011 participants) estimating high sensitivity 99.9% (95% CI 99.5, 100) and high specificity 99.2% (95% CI 98.5, 99.5).

Discussion

If and how the 2021 search has impacted on evidence base?

The 2021 search provided four additional systematic review (Alshehri 2021, Ontario Health 2020, Runkel 2020, Yang 2019), one cohort study (Jernman 2021) and three diagnostic accuracy studies (Parchure 2021, Pazourkova 2021, Legler 2021) relating to the effectiveness of non-invasive diagnostic testing of fetal Rh D status. The 2021 update has provided studies that impact on the evidence base through consolidating non-invasive techniques with high sensitivity, specificity and diagnostic accuracy.

If and how the 2021 search has created changes in the evidence?

The 2021 update does not change any of the findings from the evidence base. The additional studies have outcomes and findings similar to that of the previous search. Questions remain regarding the true effectiveness of NIPT on patient-relevant outcomes (i.e. the incidence of Rh D sensitisations or HDFN.

Question 4 - Risk of failure of Rh D immunoprophylaxis due to increased BMI

Question 4 – (Prognostic)

In Rh D negative pregnant or postpartum women with no preformed anti-D, does increasing BMI increase the risk of failure of Rh D immunoglobulin administration?

Background

The 2018 review identified two Level II studies (MacKenzie, 2004, Woelfer, 2004) and two Level III studies (Bichler J., 2003, Koelewijn, 2009) that provided some evidence relating maternal body weight to Rh D immunoglobulin administration.

Summary of evidence

The 2021 update found one additional Level III study (Wikman, 2021) that retrospectively examined the proportion of women with undetectable levels of prophylactic Rh D immunoglobulin at the time of delivery after RAADP (single dose of 1500 IU at28-29 gestational weeks'). It was noted that 16.5% had BMI > 30 and 4.4% had BMI > 35.

Results

During the retrospective study period (Oct 2010 to Oct 2012), Wikman 2021 found there were 876 (20.5%) cases among 4280 Rh D negative women carrying an RHD positive fetus in which the antibody screen result was negative (i.e., not detectable at delivery). In the prospective cohort, 7/39 (18%) women did not have detectable levels of anti-D at screening (38 gestational weeks), and in 10/39 (26%), the anti-D levels were below the lower limit of quantification.

After administration of the second dose at 38 gestational weeks', the mean increase in anti-D concentration (IU/mL) was 0.066 (SD 0.045) and showed a significant correlation with body mass index (p = 0.0118). The authors noted a large interindividual variation of anti-D concentration at delivery, which is suggested to depend on individual IgG clearance from plasma and consumption of anti-D, giving a variability in residual anti-D levels and in half-life. Uptake from muscular compartments and fat tissue may vary as well.

The incidence of FMH was analysed after delivery and the results were negative in all 25 of 39 (64%) patients tested (i.e., test result was below the limit of detection being 1 ml fetal blood in maternal circulation). Data were missing for 14/39 (36%) patients.

Discussion

If and how the 2021 search has impacted on evidence base?

The 2021 search provided one additional cohort study that show a correlation between anti-D levels and BMI. It enhances the evidence relating to the proportion of RhD negative pregnant women at risk of Rh D sensitisation with no detectable anti-D at delivery, despite RAADP

If and how the 2021 search has created changes in the evidence?

The 2021 update does not change any of the findings from the evidence base.

Appendix A Literature search results

This appendix documents the literature search strategy for a systematic review on the prophylactic use of Rh D Immunoglobulin (Anti-D) in pregnant women.

A search strategy to address all questions was developed via Ovid for both Embase and MEDLINE. An additional search for studies reporting diagnostic accuracy specific to subquestion 3 was also conducted. Both search strategies were then translated for PubMed (limited to in-process citations and citations not indexed in MEDLINE) and CINAHL.

A1 Questions 1 to 4

Embase

Table A.1 Search results Questions 1 to 4: Embase (via Ovid) for Level I, Level II and Level III studies

#	Searches	Results [a] 19 July 2018	Results [b] 27 Sept 2021
1	exp "obstetrics"/ or exp "obstetric care"/ or exp "pregnancy"/ or exp "pregnancy disorder"/ or exp "prenatal disorder"/	1138534	1224786
2	(obstetric or obstetrics or pregnancy or maternal).ti,ab,kw.	688903	798197
3	(prepartum or pre partum or pre-partum or intrapartum or intra partum or intra-partum or perinatal or peri natal or peri-natal).ti,ab,kw.	98059	115994
4	(antenatal or ante natal or ante-natal or prenatal or pre natal or pre-natal).ti,ab,kw.	151232	178341
5	(postnatal or post natal or post-natal or postpartum or post partum or post-partum).ti,ab,kw.	194295	231147
6	1 or 2 or 3 or 4 or 5	1434130	1566408
7	exp "fetus"/	189819	199261
8	(fetu* or fetal* or f?etu* or f?etal*).ti,ab,kw.	375459	416482
9	7 or 8	428990	466603
10	exp alloimmunization/	4373	5319
11	exp Rh Isoimmunization/	1604	1500
12	(Rh* Isoimmuni?ation or Rh* D Isoimmuni?ation).ti,ab.	719	588
13	(Rh* alloimmuni?ation or Rh* D alloimmuni?ation).ti,ab.	381	456
14	(Rh* incompatibility or Rh* D incompatibility or blood group incompatibility).ti,ab.	1102	1022
15	(((Rh* adj3 incompatib*) or Rh* D) adj3 incompatibl*).ti,ab.	203	194
16	((Rh or RhD or rhesus) adj5 sensiti*).ti,ab.	1325	265
17	((fetomaternal or feto-maternal or foetomaternal or foeto-maternal) adj2 immuni?ation).ti,ab.	81	54
18	((rh or RhD or rhesus) adj2 (immuni?ation or autoimmuni?ation)).ti,ab.	862	541
19	10 or 11 or 12 or 13 or 14 or 15 or 16 or 17 or 18	9257	8692
20	exp rhesus D antigen/	1158	1532
21	rhesus D antigen.ti,ab.	55	57
22	rh* D antigen.ti,ab.	234	241
23	(RhD or rhesus D or Rh?D or Rh-?D or Rh D).ti,ab.	7552	9309
24	(Rh-negative or Rh-positive).ti,ab.	1312	1376
25	(Rhesus negative or Rhesus positive).ti,ab.	362	382
26	((rh or rhesus) adj2 (factor or factors or antigen$ or system or group)).ti,ab.	4806	5015
27	20 or 21 or 22 or 23 or 24 or 25 or 26	12535	14306
28	(Macaca mulatta or Simian Immunodeficiency Virus or zika or macaque?).ti,ab.	38080	48547
29	27 not 28	12360	14114
30	(isoimmuni?ation or alloimmuni?ation).ti,ab,kw.	5921	6513
31	(isoimmuni* or iso-immuni* or isoimmune or iso-immune).ti,ab,kw.	2122	1570
32	(alloimmuni* or allo-immuni* or alloimmune or allo-immune).ti,ab,kw.	11072	13031
33	(unsensiti?ed or un-sensiti?ed or non-sensiti?ed).ti,ab,kw.	2409	2617

34	(sensiti?ation* or sensiti?ed).ti,ab,kw.	119508	134779
35	30 or 31 or 32 or 33 or 34	132314	148922
36	exp Erythroblastosis, Fetal/	11405	9270
37	((erythroblastoses or erythroblastosis) adj2 (fetal* or f?etal*)).ti,ab,kw.	1103	266
38	(h?emolytic disease* or h?emolytic disorder*).ti,ab,kw.	5204	4940
39	(HDFN or HDN).ti,ab,kw.	1169	1533
40	36 or 37 or 38 or 39	14943	12898
41	6 or 9 or 19 or 29 or 35 or 40	1702755	1858675
42	exp Rh D immunoglobulin/	3931	4252
43	exp Rho D Immune Globulin/	3931	4252
44	exp "Rho(D) Immune Globulin"/	3931	4252
45	exp anti-D immunoglobulin/	3931	4252
46	Rh* D Immune Globulin.ti,ab.	93	88
47	(rh* immunoglobulin or rh* d immunoglobulin).ti,ab.	310	332
48	(rh* immuni?ation or rh* d immuni?ation).ti,ab.	574	425
49	42 or 43 or 44 or 45 or 46 or 47 or 48	4584	4753
50	exp rhesus D antibody/	3931	4252
51	rhesus D antibody.ti,ab.	11	11
52	(rh* D antibody or rh*D antibody).ti,ab.	108	107
53	(anti-D or anti D or anti?D).ti,ab.	4652	5158
54	50 or 51 or 52 or 53	6309	6974
55	exp rhogam/	3931	4252
56	rhogam.ti,ab.	47	46
57	exp winrho/	3931	4252
58	winrho.ti,ab.	64	65
59	exp rhophylac/	3931	4252
60	rhophylac.ti,ab.	21	21
61	exp MICRhoGam/	3931	4252
62	exp BayRHo-D/	3931	4252
63	exp rhesonativ/	3931	4252
64	'RhD immunoglobulin vf'.ti,ab.	0	0
65	55 or 56 or 57 or 58 or 59 or 60 or 61 or 62 or 63 or 64	3952	4267
66	49 or 54 or 65	6855	7363
67	41 and 66	4895	5152
68	exp meta analysis/ or meta analysis.mp. or exp systematic review/ or systematic review.mp. or pooled analysis.mp. or ((exp review/ or review.mp.) and (systemat* or pool*).mp.)	424029	643063
69	exp comparative study/ or comparative study.mp. or exp clinical trial/ or clinical trial.mp. or randomized controlled trial.mp. or randomi?ed controlled trial.mp. or exp randomized controlled trial/ or exp randomization/ or randomization.mp. or randomi?ation.mp. or exp single blind procedure/ or single blind procedure.mp. or exp double blind procedure/ or double blind procedure.mp. or exp triple blind procedure/ or triple blind procedure.mp. or exp crossover procedure/ or crossover procedure.mp. or exp placebo/ or placebo*.mp. or random*.mp. or rct.mp. or single blind.mp. or single blinded.mp. or double blind.mp. or double blinded.mp. or treble blind.mp. or triple blind.mp. or triple blinded.mp. or exp prospective study/ or prospective study.mp.	3989862	4922439
70	exp clinical study/ or exp case control study/ or exp family study/ or exp longitudinal study/ or exp retrospective study/ or exp cohort analysis/ or (cohort adj1 stud*).mp. or (case control adj1 stud*).mp. or (exp prospective study/ not randomi?ed controlled trials.mp.) or (follow up adj1 stud*).mp. or (observational adj1 stud*).mp. or (epidemiologic* adj1 stud*).mp. or (cross sectional adj1 stud*).mp.	9057811	11236854
71	"case report"/	2319441	2658800
72	(editorial or letter or comment or historical article).pt.	1600355	1895537
73	71 or 72	3716272	4329515
74	(animals/ or nonhuman/) not humans/	6126128	6443842
75	(67 and 68) not (72 or 74)	69	85
76	(67 and 69) not (73 or 74 or 75)	470	542
77	(67 and 70) not (73 or 74 or 75 or 76)	990	1222
78	limit 75 to yr="2018 -Current"	NA	**27**

79	limit 76 to yr="2018 -Current"	NA	**106**
80	limit 77 to yr="2018 -Current"	NA	**302**

NA, not applicable;

a. Embase <1974 to 18 July 2018>

b. Embase <1974 to 24 September 2021 >

MEDLINE

Table A.2 Search results Questions 1 to 4: Medline (via Ovid) for Level I, Level II and Level III studies

#	Searches	Results [a] 19 July 2018	Results [b] 27 Sept 2021
1	exp "obstetrics"/ or exp "obstetric care"/ or exp "pregnancy"/ or exp "pregnancy disorder"/ or exp "prenatal disorder"/	846826	945998
2	(obstetric or obstetrics or pregnancy or maternal).ti,ab,kw.	542671	643633
3	(prepartum or pre partum or pre-partum or intrapartum or intra partum or intra-partum or perinatal or peri natal or peri-natal).ti,ab,kw.	72542	87299
4	(antenatal or ante natal or ante-natal or prenatal or pre natal or pre-natal).ti,ab,kw.	113427	137293
5	(postnatal or post natal or post-natal or postpartum or post partum or post-partum).ti,ab,kw.	152703	181566
6	1 or 2 or 3 or 4 or 5	1127944	1280398
7	exp "fetus"/	151495	161305
8	(fetu* or fetal* or f?etu* or f?etal*).ti,ab,kw.	296734	333936
9	7 or 8	368759	409865
10	exp alloimmunization/	0	0
11	exp Rh Isoimmunization/	1672	1753
12	(Rh* Isoimmuni?ation or Rh* D Isoimmuni?ation).ti,ab.	602	614
13	(Rh* alloimmuni?ation or Rh* D alloimmuni?ation).ti,ab.	215	256
14	(Rh* incompatibility or Rh* D incompatibility or blood group incompatibility).ti,ab.	910	973
15	(((Rh* adj3 incompatib*) or Rh* D) adj3 incompatibl*).ti,ab.	154	166
16	((Rh or RhD or rhesus) adj5 sensiti*).ti,ab.	1197	1225
17	((fetomaternal or feto-maternal or foetomaternal or foeto-maternal) adj2 immuni?ation).ti,ab.	78	78
18	((rh or RhD or rhesus) adj2 (immuni?ation or autoimmuni?ation)).ti,ab.	755	780
19	10 or 11 or 12 or 13 or 14 or 15 or 16 or 17 or 18	4742	5020
20	exp rhesus D antigen/	0	0
21	rhesus D antigen.ti,ab.	37	38
22	rh* D antigen.ti,ab.	183	189
23	(RhD or rhesus D or Rh?D or Rh-?D or Rh D).ti,ab.	4497	5412
24	(Rh-negative or Rh-positive).ti,ab.	951	1026
25	(Rhesus negative or Rhesus positive).ti,ab.	241	258
26	((rh or rhesus) adj2 (factor or factors or antigen$ or system or group)).ti,ab.	3883	4358
27	20 or 21 or 22 or 23 or 24 or 25 or 26	8684	10069
28	(Macaca mulatta or Simian Immunodeficiency Virus or zika or macaque?).ti,ab.	32787	41471
29	27 not 28	8527	9889
30	(isoimmuni?ation or alloimmuni?ation).ti,ab,kw.	3791	4313
31	(isoimmuni* or iso-immuni* or isoimmune or iso-immune).ti,ab,kw.	2001	2064
32	(alloimmuni* or allo-immuni* or alloimmune or allo-immune).ti,ab,kw.	6618	7647
33	(unsensiti?ed or un-sensiti?ed or non-sensiti?ed).ti,ab,kw.	1631	1750
34	(sensiti?ation* or sensiti?ed).ti,ab,kw.	90235	103333
35	30 or 31 or 32 or 33 or 34	98707	112853
36	exp Erythroblastosis, Fetal/	11582	12015
37	((erythroblastoses or erythroblastosis) adj2 (fetal* or f?etal*)).ti,ab,kw.	858	908
38	(h?emolytic disease* or h?emolytic disorder*).ti,ab,kw.	4553	4970
39	(HDFN or HDN).ti,ab,kw.	552	722
40	36 or 37 or 38 or 39	13563	14346
41	6 or 9 or 19 or 29 or 35 or 40	1361580	1539131

#	Searches	Results [a] 19 July 2018	Results [b] 27 Sept 2021
42	exp Rh D immunoglobulin/	0	0
43	exp Rho D Immune Globulin/	1271	1388
44	exp "Rho(D) Immune Globulin"/	1271	1388
45	exp anti-D immunoglobulin/	1271	1388
46	Rh* D Immune Globulin.ti,ab.	68	73
47	(rh* immunoglobulin or rh* d immunoglobulin).ti,ab.	215	236
48	(rh* immuni?ation or rh* d immuni?ation).ti,ab.	486	504
49	42 or 43 or 44 or 45 or 46 or 47 or 48	1885	2028
50	exp rhesus D antibody/	0	0
51	rhesus D antibody.ti,ab.	10	10
52	(rh* D antibody or rh*D antibody).ti,ab.	86	89
53	(anti-D or anti D or anti?D).ti,ab.	2820	3050
54	50 or 51 or 52 or 53	2890	3123
55	exp rhogam/	1271	1408
56	rhogam.ti,ab.	32	32
57	exp winrho/	0	0
58	winrho.ti,ab.	41	42
59	exp rhophylac/	1271	1390
60	rhophylac.ti,ab.	8	8
61	exp MICRhoGam/	1271	1388
62	exp BayRHo-D/	0	0
63	exp rhesonativ/	0	0
64	'RhD immunoglobulin vf'.ti,ab.	0	0
65	55 or 56 or 57 or 58 or 59 or 60 or 61 or 62 or 63 or 64	1302	1421
66	49 or 54 or 65	3847	4144
67	41 and 66	2741	2972
68	exp meta analysis/ or meta analysis.mp. or exp systematic review/ or systematic review.mp. or pooled analysis.mp. or ((exp review/ or review.mp.) and (systemat* or pool*).mp.)	278527	432159
69	exp comparative study/ or comparative study.mp. or exp clinical trial/ or clinical trial.mp. or randomized controlled trial.mp. or randomi?ed controlled trial.mp. or exp randomized controlled trial/ or exp randomization/ or randomization.mp. or randomi?ation.mp. or exp single blind procedure/ or single blind procedure.mp. or exp double blind procedure/ or double blind procedure.mp. or exp triple blind procedure/ or triple blind procedure.mp. or exp crossover procedure/ or crossover procedure.mp. or exp placebo/ or placebo*.mp. or random*.mp. or rct.mp. or single blind.mp. or single blinded.mp. or double blind.mp. or double blinded.mp. or treble blind.mp. or triple blind.mp. or triple blinded.mp. or exp prospective study/ or prospective study.mp.	3476781	3994060
70	exp clinical study/ or exp case control study/ or exp family study/ or exp longitudinal study/ or exp retrospective study/ or exp cohort analysis/ or (cohort adj1 stud*).mp. or (case control adj1 stud*).mp. or (exp prospective study/ not randomi?ed controlled trials.mp.) or (follow up adj1 stud*).mp. or (observational adj1 stud*).mp. or (epidemiologic* adj1 stud*).mp. or (cross sectional adj1 stud*).mp.	2982544	3749193
71	"case report"/	1887103	2212725
72	(editorial or letter or comment or historical article).pt.	1969551	2342764
73	71 or 72	3656632	4331387
74	(animals/ or nonhuman/) not humans/	4443785	4856723
75	(67 and 68) not (72 or 74)	**29**	40
76	(67 and 69) not (73 or 74 or 75)	**316**	345
77	(67 and 70) not (73 or 74 or 75 or 76)	**190**	239
69	limit 66 to yr="2018 -Current"	NA	**12**
70	limit 67 to yr="2018 -Current"	NA	**37**
71	limit 68 to yr="2018 -Current"	NA	**53**

NA, not applicable

a. Ovid MEDLINE(R) Epub Ahead of Print, In-Process & Other Non-Indexed Citations, Ovid MEDLINE(R) Daily, Ovid MEDLINE and Versions(R) 1946 to July 18, 2018

b. Ovid MEDLINE(R) ALL <1946 to September 24, 2021>

Evidence-Based Medicine Reviews

Table A.3 Search results Questions 1 to 4: EBM Reviews (via Ovid)

#	Searches	Results 19 July 2018	Results 27 Sept 2021
1	exp "obstetrics"/ or exp "obstetric care"/ or exp "pregnancy"/ or exp "pregnancy disorder"/ or exp "prenatal disorder"/	19993	23474
2	(obstetric or obstetrics or pregnancy or maternal).ti,ab,kw.	37479	71083
3	(prepartum or pre partum or pre-partum or intrapartum or intra partum or intra-partum or perinatal or peri natal or peri-natal).ti,ab,kw.	4694	7790
4	(antenatal or ante natal or ante-natal or prenatal or pre natal or pre-natal).ti,ab,kw.	5834	10176
5	(postnatal or post natal or post-natal or postpartum or post partum or post-partum).ti,ab,kw.	8587	15414
6	1 or 2 or 3 or 4 or 5	50733	88340
7	exp "fetus"/	1614	1812
8	(fetu* or fetal* or f?etu* or f?etal*).ti,ab,kw.	8812	15176
9	7 or 8	9664	16152
10	exp alloimmunization/	0	0
11	exp Rh Isoimmunization/	30	32
12	(Rh* Isoimmuni?ation or Rh* D Isoimmuni?ation).ti,ab.	13	18
13	(Rh* alloimmuni?ation or Rh* D alloimmuni?ation).ti,ab.	6	14
14	(Rh* incompatibility or Rh* D incompatibility or blood group incompatibility).ti,ab.	25	52
15	(((Rh* adj3 incompatib*) or Rh* D) adj3 incompatibl*).ti,ab.	2	4
16	((Rh or RhD or rhesus) adj5 sensiti*).ti,ab.	23	28
17	((fetomaternal or feto-maternal or foetomaternal or foeto-maternal) adj2 immuni?ation).ti,ab.	0	0
18	((rh or RhD or rhesus) adj2 (immuni?ation or autoimmuni?ation)).ti,ab.	30	33
19	10 or 11 or 12 or 13 or 14 or 15 or 16 or 17 or 18	116	164
20	exp rhesus D antigen/	0	0
21	rhesus D antigen.ti,ab.	0	0
22	rh* D antigen.ti,ab.	0	0
23	(RhD or rhesus D or Rh?D or Rh-?D or Rh D).ti,ab.	139	217
24	(Rh-negative or Rh-positive).ti,ab.	23	46
25	(Rhesus negative or Rhesus positive).ti,ab.	17	22
26	((rh or rhesus) adj2 (factor or factors or antigen$ or system or group)).ti,ab.	117	162
27	20 or 21 or 22 or 23 or 24 or 25 or 26	283	418
28	(Macaca mulatta or Simian Immunodeficiency Virus or zika or macaque?).ti,ab.	151	281
29	27 not 28	283	418
30	(isoimmuni?ation or alloimmuni?ation).ti,ab,kw.	175	262
31	(isoimmuni* or iso-immuni* or isoimmune or iso-immune).ti,ab,kw.	46	65
32	(alloimmuni* or allo-immuni* or alloimmune or allo-immune).ti,ab,kw.	266	406
33	(unsensiti?ed or un-sensiti?ed or non-sensiti?ed).ti,ab,kw.	49	71
34	(sensiti?ation* or sensiti?ed).ti,ab,kw.	2899	4409
35	30 or 31 or 32 or 33 or 34	3200	4861
36	exp Erythroblastosis, Fetal/	70	76
37	((erythroblastoses or erythroblastosis) adj2 (fetal* or f?etal*)).ti,ab,kw.	14	10
38	(h?emolytic disease* or h?emolytic disorder*).ti,ab,kw.	106	157
39	(HDFN or HDN).ti,ab,kw.	21	32
40	36 or 37 or 38 or 39	150	210
41	6 or 9 or 19 or 29 or 35 or 40	55540	95795
42	exp Rh D immunoglobulin/	0	0
43	exp Rho D Immune Globulin/	169	240
44	exp "Rho(D) Immune Globulin"/	169	240
45	exp anti-D immunoglobulin/	169	240
46	Rh* D Immune Globulin.ti,ab.	9	11
47	(rh* immunoglobulin or rh* d immunoglobulin).ti,ab.	13	17

#	Searches	Results 19 July 2018	Results 27 Sept 2021
48	(rh* immuni?ation or rh* d immuni?ation).ti,ab.	28	32
49	42 or 43 or 44 or 45 or 46 or 47 or 48	208	288
50	exp rhesus D antibody/	0	0
51	rhesus D antibody.ti,ab.	0	0
52	(rh* D antibody or rh*D antibody).ti,ab.	5	9
53	(anti-D or anti D or anti?D).ti,ab.	145	216
54	50 or 51 or 52 or 53	150	223
55	exp rhogam/	169	240
56	rhogam.ti,ab.	2	4
57	exp winrho/	0	0
58	winrho.ti,ab.	5	7
59	exp rhophylac/	169	240
60	rhophylac.ti,ab.	5	7
61	exp MICRhoGam/	169	240
62	exp BayRHo-D/	0	0
63	exp rhesonativ/	0	0
64	'RhD immunoglobulin vf'.ti,ab.	0	0
65	55 or 56 or 57 or 58 or 59 or 60 or 61 or 62 or 63 or 64	176	252
66	49 or 54 or 65	310	457
67	41 and 66	102	149
68	limit 67 to yr="2018 -Current"	NA	**19**

NA, not applicable

a. EBM Reviews combines several resources into a single database and includes the following: ACP Journal Club <1991 to June 2018>; Cochrane Database of Systematic Reviews <2005 to July 18, 2018>; Database of Abstracts of Reviews of Effects <1st Quarter 2016>; Cochrane Clinical Answers <June 2018>; Cochrane Central Register of Controlled Trials <June 2018>; Cochrane Methodology Register <3rd Quarter 2012>; Health Technology Assessment <4th Quarter 2016>; NHS Economic Evaluation Database <1st Quarter 2016>.

b. EBM Reviews - Cochrane Database of Systematic Reviews <2005 to September 23, 2021>; EBM Reviews - ACP Journal Club <1991 to August 2021>; EBM Reviews - Database of Abstracts of Reviews of Effects <1st Quarter 2016>; EBM Reviews - Cochrane Clinical Answers <September 2021>; EBM Reviews - Cochrane Central Register of Controlled Trials <August 2021>; EBM Reviews - Cochrane Methodology Register <3rd Quarter 2012>; EBM Reviews - Health Technology Assessment <4th Quarter 2016>; EBM Reviews - NHS Economic Evaluation Database <1st Quarter 2016>

PubMed

The PubMed search is restricted to records that are not indexed for MEDLINE (i.e. in-process citations and citations from journals (or parts of journals) that are not currently MEDLINE-indexed) and to records added to PubMed since January 2006.

The search comprises free-text terms only and replicates the free-text sets in the Embase search (converted from the Ovid syntax).

Table A.4 Search results Questions 1 to 4: Pubmed (not MEDLINE)

#	Searches	Results 20 July 2018	Results 27 Sept 2021
#49	(#47 AND pubmednotmedline[sb]) from 2018 to 2021	NA	**108**
#48	(#47 AND pubmednotmedline[sb])	200	310
#47	(#32 AND #46)	4737	5,281
#46	(#36 OR #45)	8156	9,191
#45	(#38 OR #39 OR #40 OR #41 OR #42 OR #43 OR #44)	53	87
#44	RhD immunoglobulin vf[tiab]	0	0
#43	rhesonativ[tiab]	2	2
#42	BayRHo-D[tiab]	0	0
#41	MICRhoGam[tiab]	2	1
#40	RhD immunoglobulin-vf[tiab]	0	0

#	Searches	**Results 20 July 2018**	Results 27 Sept 2021
#39	rhophylac[tiab]	8	8
#38	winrho[tiab]	42	44
#37	rhogam[tiab]	33	36
#36	(#33 OR #34 OR #35)	8146	9,164
#35	(anti-d[tiab] OR anti d[tiab])	2811	3,059
#34	((rhesus[tiab] OR rh[tiab] OR rho[tiab]) AND antibody[tiab])	4989	5,692
#33	((rhesus[tiab] OR rh[tiab] OR rho[tiab] OR RHD[tiab]) AND (immunoglobulin[tiab] OR immune globulin[tiab]))	1576	1,781
#32	(#5 OR #6 OR #14 OR #22 OR #27 OR #31)	1018102	1,179,283
#31	(#28 OR #29 OR #30)	20920	23,548
#30	(hdfn[tiab] OR hdn[tiab])	553	750
#29	((hemolytic OR haemolytic) AND (disorder[tiab] OR disorders[tiab] OR disease[tiab] OR diseases[tiab]))	18492	21,044
#28	((erythroblastoses[tiab] OR erythroblastosis[tiab]) AND (fetal[tiab] OR foetal[tiab] OR fetalis[tiab] OR foetalis[tiab]))	3161	3,172
#27	(#23 OR #24 OR #25 OR #26)	98817	113,839
#26	(sensitisation*[tiab] OR sensitization*[tiab] OR sensitised[tiab] OR sensitized[tiab])	90526	104,466
#25	((rh[tiab] OR rho[tiab] OR rhesus[tiab]) AND (sensitising[tiab] OR sensitizing[tiab] OR sensitisation[tiab] OR sensitization[tiab] OR sensitised[tiab] OR sensitized[tiab]))	1599	1,735
#24	(alloimmuni*[tiab] OR allo-immuni*[tiab] OR alloimmune[tiab] OR allo-immune[tiab])	6637	7,693
#23	(isoimmuni*[tiab] OR iso-immuni*[tiab] OR isoimmune[tiab] OR iso-immune[tiab])	2006	2,084
#22	(#20 NOT #21)	23409	27,073
#21	(Macaca mulatta[tiab] OR Simian Immunodeficiency Virus[tiab] OR zika[tiab] OR macaque[tiab] OR macaques[tiab])	33079	42,075
#20	(#15 OR #16 OR #17 OR #18 OR #19)	27458	31,807
#19	(rh[tiab] OR rhesus[tiab]) AND (factor[tiab] OR factors[tiab] OR antigen*[tiab] OR antigens[tiab] OR system[tiab] OR group[tiab])	24249	27,870
#18	(rhesus negative[tiab] OR rhesus positive[tiab])	240	261
#17	(rh-negative[tiab] OR rh-positive[tiab] OR rh negative[tiab] OR rh positive[tiab])	949	1,038
#16	(RhD[tiab] OR rhesus d[tiab] OR Rh D[tiab])	3887	4,752
#15	((rh[tiab] OR rhd[tiab] OR rhesus[tiab]) AND antigen[tiab])	3429	3,815
#14	(#7 OR #8 OR #9 OR #10 OR #11 OR #12 OR #13)	11065	12,721
#13	((rh[tiab] OR RhD[tiab] OR rhesus[tiab]) AND (immunisation[tiab] OR immunization[tiab] OR autoimmunisation[tiab] OR autoimmunization[tiab]))	2055	2,251
#12	((fetomaternal[tiab] OR feto-maternal[tiab] OR foetomaternal[tiab] OR foeto-maternal[tiab]) AND (immunisation[tiab] OR immunization[tiab]))	166	174
#11	((Rh[tiab] OR RhD[tiab] OR rhesus[tiab]) AND (sensiti*[tiab]))	4933	5,734
#10	((rh[tiab] OR rhd[tiab] OR rhesus[tiab]) AND (incompatib*[tiab]))	1307	1,428
#9	((rh[tiab] OR rhd[tiab] OR rhesus[tiab]) AND (incompatibility[tiab]) OR (blood group incompatibility[tiab]))	1636	1,747
#8	((rh[tiab] OR rhd[tiab] rhesus[tiab]) AND (isoimmunization[tiab] OR isoimmunisation[tiab]))	63	82
#7	(alloimmunization[tiab] OR alloimmunisation[tiab])	2590	3,240
#6	(fetus[tiab] OR foetus[tiab] OR fetu*[tiab] OR foetu*[tiab] OR fetal*[tiab] OR foetal*[tiab])	299655	337,336
#5	(#1 OR #2 OR #3 OR #4)	738834	867,966
#4	(postnatal[tiab] OR post natal[tiab] OR post-natal[tiab] OR postpartum[tiab] OR post partum[tiab] OR post-partum[tiab])	153623	183,012
#3	(antenatal[tiab] OR ante natal[tiab] OR ante-natal[tiab] OR prenatal[tiab] OR pre natal[tiab] OR pre-natal[tiab])	115113	139,910
#2	(prepartum[tiab] OR pre partum[tiab] OR pre-partum[tiab] OR intrapartum[tiab] OR intra partum[tiab] OR intra-partum[tiab] OR perinatal[tiab] OR peri natal[tiab] OR peri-natal[tiab])	73169	88,660
#1	(Obstetric[tiab] OR obstetrics[tiab] OR pregnancy[tiab] OR maternal[tiab])	554296	657,008

NA, not applicable

CINAHL

Table A.5 Search results Questions 1 to 4: CINAHL

Searched conducted

#	Query	Results 20 July 2018	Results 27 Sept 2021
S1	(MH "Obstetrics") or (MH "Obstetric Care+") or (MH "Pregnancy+") or "pregnancy disorder" or "prenatal disorder"	128,235	240,984
S2	TI (obstetric or obstetrics or pregnancy or maternal) OR AB (obstetric or obstetrics or pregnancy or maternal) or ("obstetric" or "obstetrics" or "pregnancy" or "maternal")	157,109	308,803
S3	TI (obstetric or obstetrics or pregnancy or maternal) OR AB (obstetric or obstetrics or pregnancy or maternal) or ("obstetric" or "obstetrics" or "pregnancy" or "maternal")	157,109	187,886
S4	TI (antenatal or ante natal or ante-natal or prenatal or pre natal or pre-natal) OR AB (antenatal or ante natal or ante-natal or prenatal or pre natal or pre-natal) OR ("antenatal" or "ante natal" or "ante-natal" or "prenatal" or "pre natal" or "pre-natal")	29,500	66,056
S5	TI (postnatal or post natal or post-natal or postpartum or post partum or post-partum) OR AB (postnatal or post natal or post-natal or postpartum or post partum or post-partum) OR ("postnatal" or "post natal" or "post-natal" or "postpartum" or "post partum" or "post-partum")	23,198	50,224
S6	S1 OR S2 OR S3 OR S4 OR S5	172,767	345,785
S7	(MH "Fetus+")	17,301	26,623
S8	TI (fetu* or fetal* or f#etu* or f#etal*) OR AB (fetu* or fetal* or f#etu* or f#etal*) OR ("fetu*" or "fetal*" or "f#etu*" or "f#etal*")	2,598,213	82,570
S9	S7 OR S8	2,598,237	82,893
S10	"alloimmuni?ation"	343	692
S11	(MH "RH Isoimmunization")	275	458
S12	TI (Rh* Isoimmuni?ation or Rh* D Isoimmuni?ation) OR AB (Rh* Isoimmuni?ation or Rh* D Isoimmuni?ation)	29	61
S13	TI (Rh* alloimmuni?ation or Rh* D alloimmuni?ation) OR AB (Rh* alloimmuni?ation or Rh* D alloimmuni?ation)	25	111
S14	TI (Rh* incompatibility or Rh* D incompatibility or blood group incompatibility) OR AB (Rh* incompatibility or Rh* D incompatibility or blood group incompatibility)	49	115
S15	TI ((Rh* N3 incompatib*) OR (Rh* D N3 incompatibl*)) OR AB ((Rh* N3 incompatib*) OR (Rh* D N3 incompatibl*))	37	82
S16	TI ((Rh or RhD or rhesus) N5 sensiti*) OR AB ((Rh or RhD or rhesus) N5 sensiti*)	60	124
S17	TI (fetomaternal or feto-maternal or foetomaternal or foeto-maternal) N2 immuni?ation) OR AB (fetomaternal or feto-maternal or foetomaternal or foeto-maternal) N2 immuni?ation)	2	2
S18	TI (((rh or RhD or rhesus) N2 (immuni?ation or autoimmuni?ation))) OR AB (((rh or rhesus) N2 (immuni?ation or autoimmuni?ation)))	10	17
S19	S10 OR S11 OR S12 OR S13 OR S14 OR S15 OR S16 OR S17 OR S18	656	1,247
S20	"rhesus D antigen"	2	4
S21	TI rhesus D antigen OR AB rhesus D antigen	3	9
S22	TI rh* D antigen OR AB rh* D antigen	36	64
S23	TI (RhD or rhesus D or Rh D or Rh-D) OR AB (RhD or rhesus D or Rh D or Rh-D)	528	1,117
S24	TI (Rh negative OR Rh positive) OR AB (Rh negative OR Rh positive))	88	194
S25	TI (Rhesus negative or Rhesus positive) OR AB (Rhesus negative or Rhesus positive)	32	78
S26	TI (rh or rhesus) N2 (factor or factors or antigen* or system or group)) OR AB (rh or rhesus) N2 (factor or factors or antigen* or system or group))	156	439
S27	S20 OR S21 OR S22 OR S23 OR S24 OR S25 OR S26	688	1,551
S28	TI (Macaca mulatta or Simian Immunodeficiency Virus or zika or macaque#) OR AB (Macaca mulatta or Simian Immunodeficiency Virus or zika or macaque#)	1,516	4,005
S29	S27 NOT S28	683	1,526
S30	TI (isoimmuni?ation or alloimmuni?ation) OR AB (isoimmuni?ation or alloimmuni?ation) OR ("isoimmuni?ation" or "alloimmuni?ation")	579	1,099
S31	TI (isoimmuni* or iso-immuni* or isoimmune or iso-immune) OR AB (isoimmuni* or iso-immuni* or isoimmune or iso-immune) OR ("isoimmuni*" or "iso-immuni*" or "isoimmune" or "iso-immune")	310	552
S32	TI (alloimmuni* or allo-immuni* or alloimmune or allo-immune) OR AB (alloimmuni* or allo-immuni* or alloimmune or allo-immune) OR ("alloimmuni*" or "allo-immuni*" or "alloimmune" or "allo-immune")	607	1,251

#	Query	Results 20 July 2018	Results 27 Sept 2021
S33	TI (unsensiti?ed or un-sensiti?ed or non-sensiti?ed) OR AB (unsensiti?ed or un-sensiti?ed or non-sensiti?ed) OR ("unsensiti?ed" or "un-sensiti?ed" or "non-sensiti?ed")	20	61
S34	TI (sensiti?ation* or sensiti?ed) OR AB (sensiti?ation* or sensiti?ed) OR ("sensiti?ation*" or "sensiti?ed")	3,426	8,596
S35	S30 OR S31 OR S32 OR S33 OR S34	4,227	10,186
S36	(MH "Erythroblastosis, Fetal+")	616	1,202
S37	TI (((erythroblastoses or erythroblastosis) N2 (fetal* or f#etal*))) OR AB (((erythroblastoses or erythroblastosis) N2 (fetal* or f#etal*))) OR ((("erythroblastoses" or "erythroblastosis") N2 ("fetal*" or "f#etal*")))	240	437
S38	TI ((h#emolytic disease* or h#emolytic disorder*)) OR AB ((h#emolytic disease* or h#emolytic disorder*)) OR (("h#emolytic disease*" or "h#emolytic disorder*"))	376	825
S39	TI (HDFN or HDN) OR AB (HDFN or HDN) OR ("HDFN" or "HDN")	57	141
S40	S36 OR S37 OR S38 OR S39	873	1,824
S41	S6 OR S9 OR S19 OR S35 OR S40	2,610,641	371,868
S42	"Rh D immunoglobulin"	1	6
S43	"Rho D Immune Globulin"	222	341
S44	(MH "Rho(D) Immune Globulin")	219	338
S45	"anti-D immunoglobulin"	34	70
S46	TI Rh* D Immune Globulin OR AB Rh* D Immune Globulin	27	36
S47	TI (rh* immunoglobulin or rh* d immunoglobulin) OR AB (rh* immunoglobulin or rh* d immunoglobulin)	56	155
S48	TI (rh* immuni?ation or rh* d immuni?ation) OR AB (rh* immuni?ation or rh* d immuni?ation)	24	49
S49	S42 OR S43 OR S44 OR S45 OR S46 OR S47 OR S48	298	536
S50	"rhesus D antibody"	0	0
S51	TI rhesus D antibody OR AB rhesus D antibody	4	12
S52	TI (rh* D antibody or rh*D antibody) OR AB (rh* D antibody or rh*D antibody)	409	1,108
S53	TI (anti-D or anti D or anti?D) OR AB (anti-D or anti D or anti?D)	466	1,332
S54	S50 OR S51 OR S52 OR S53	849	2,373
S55	“rhogam”	7	17
S56	TI rhogam OR AB rhogam	7	17
S57	"winrho"	6	7
S58	TI winrho OR AB winrho	6	7
S59	TI rhophylac OR AB rhophylac OR "rhophylac"	2	2
S60	TI RhD immunoglobulin vf OR AB RhD immunoglobulin vf' OR "RhD immunoglobulin vf"	0	0
S61	TI MICRhoGam OR AB MICRhoGam OR "MICRhoGam"	0	0
S62	TI BayRHo-D OR AB BayRHo-D OR "BayRHo-D"	0	0
S63	TI rhesonativ OR AB rhesonativ OR "rhesonativ"	0	0
S64	TI RhD immunoglobulin vf OR AB RhD immunoglobulin vf	0	0
S65	S55 OR S56 OR S57 OR S58 OR S59 OR S60 OR S61 OR S62 OR S63 OR S64	15	26
S66	S49 OR S54 OR S65	1,019	2,660
S67	S41 AND S66	973	657
S68	PT (Editorial or letter or comment or historical article)	364,194	689,076
S69	S67 NOT S68	920	610
S70	S67 NOT S68 Limiters - Date Published: 20180101-20211231	NA	**147**

NA, not applicable

A2 Subquestion 3

Embase

Table A.6 Search results subquestion 3: Embase (via Ovid) for Level I, Level II and Level III studies

#	Searches	Results [a] 19 July 2018	Results [b] 28 Sept 2021
1	exp Prenatal Diagnosis/	100703	114220
2	Maternal Serum Screening Tests/	232	301
3	Hematologic Tests/	12148	14896
4	((prenatal or pre-natal or antenatal or ante-natal) adj3 (test* or screen* or diagnos* or determin* or detect*)).ti,ab.	47618	55601
5	(f?etal adj3 (test* or screen* or diagnos* or determin* or detect*)).ti,ab.	23936	27181
6	((non-invasive adj7 screening) or (non?invasive adj7 screening)).ti,ab.	4412	6066
7	(NIPD or NIPT or NIPS or NIPA).ti,ab.	2110	3497
8	or/1-7	144660	167126
9	Cell-Free Nucleic Acids/	0	1254
10	(cffCDNA or cell-free f?etal DNA).ti,ab.	1056	1354
11	((cell free dna or cfDNA) adj3 (obstetric or obstetrics or pregnancy or maternal)).ti,ab.	443	672
12	((cell free dna or cfDNA) adj3 (fetu* or fetal* or f?etu* or f?etal*)).ab,ti.	267	425
13	Genotyping Techniques/	5856	8971
14	((genotype* or genotyping) adj3 (obstetric or obstetrics or pregnancy or maternal)).ti,ab.	1424	1685
15	((genotype* or genotyping) adj3 (fetu* or fetal* or f?etu* or f?etal*)).ti,ab.	1113	1352
16	(RHD adj3 gene).ti,ab.	667	794
17	or/9-16	9881	15162
18	8 or 17	152700	179708
19	exp "obstetrics"/ or exp "obstetric care"/ or exp "pregnancy"/ or exp "pregnancy disorder"/ or exp "prenatal disorder"/	1138381	1224786
20	(obstetric or obstetrics or pregnancy or maternal).kw,ab,ti.	688764	798197
21	(prepartum or pre partum or pre-partum or intrapartum or intra partum or intra-partum or perinatal or peri natal or peri-natal).kw,ab,ti.	98043	115994
22	(antenatal or ante natal or ante-natal or prenatal or pre natal or pre-natal).kw,ab,ti.	151208	178341
23	(postnatal or post natal or post-natal or postpartum or post partum or post-partum).kw,ab,ti.	194268	231147
24	or/19-23	1433924	1566408
25	exp "fetus"/	189793	199261
26	(fetu* or fetal* or f?etu* or f?etal*).kw,ab,ti.	375396	416482
27	or/25-26	428926	466603
28	exp alloimmunization/	4372	5319
29	exp Rh Isoimmunization/	1604	1500
30	(Rh* Isoimmuni?ation or Rh* D Isoimmuni?ation).ti,ab.	719	588
31	(Rh* alloimmuni?ation or Rh* D alloimmuni?ation).ti,ab.	381	456
32	(Rh* incompatibility or Rh* D incompatibility or blood group incompatibility).ti,ab.	1102	1022
33	(((Rh* adj3 incompatib*) or Rh* D) adj3 incompatibl*).ti,ab.	203	194
34	((Rh or RhD or rhesus) adj5 sensiti*).ti,ab.	1325	265
35	((fetomaternal or feto-maternal or foetomaternal or foeto-maternal) adj2 immuni?ation).ti,ab.	81	54
36	((rh or rhesus) adj2 (immuni?ation or autoimmuni?ation)).ti,ab.	770	541
37	or/28-36	9210	8692
38	exp rhesus D antigen/	1158	1532
39	rhesus D antigen.ti,ab.	55	57
40	rh* D antigen.ti,ab.	234	241
41	(RhD or rhesus D or Rh?D or Rh-?D or Rh D).ti,ab.	7551	9309
42	(Rh-negative or Rh-positive).ti,ab.	1311	1376
43	(Rhesus negative or Rhesus positive).ti,ab.	362	382
44	((rh or rhesus) adj2 (factor or factors or antigen* or system or group)).ti,ab.	4806	5015

#	Searches	Results [a] 19 July 2018	Results [b] 28 Sept 2021
45	or/38-44	12533	14306
46	(Macaca mulatta or Simian Immunodeficiency Virus or zika or macaque?).ti,ab.	38059	48547
47	45 not 46	12358	14114
48	(isoimmuni?ation or alloimmuni?ation).ti,ab,kw.	5920	6513
49	(isoimmuni* or iso-immuni* or isoimmune or iso-immune).ti,ab,kw.	2122	1570
50	(alloimmuni* or allo-immuni* or alloimmune or allo-immune).ti,ab,kw.	11071	13031
51	(unsensiti?ed or un-sensiti?ed or non-sensiti?ed).ti,ab,kw.	2409	2617
52	(sensiti?ation* or sensiti?ed).ti,ab,kw.	119495	134779
53	or/48-52	132300	148922
54	exp Erythroblastosis, Fetal/	11404	9270
55	((erythroblastoses or erythroblastosis) adj2 (fetal* or f?etal*)).kw,ab,ti.	1103	266
56	(h?emolytic disease* or h?emolytic disorder*).ti,ab,kw.	5204	4940
57	(HDFN or HDN).ti,ab,kw.	1169	1533
58	or/54-57	14942	12898
59	24 or 27	1568681	1707662
60	37 or 47 or 53 or 58	156987	172822
61	59 and 60	23151	21809
62	18 and 61	5024	5588
63	(diagnos*.mp. and (exp performance/ or yield.mp.)) or accura*.mp. or exp accuracy/ or exp diagnostic accuracy/ or sensitivity.mp. or specificity.mp. or exp "sensitivity and specificity"/ or exp "specificity and sensitivity"/ or exp precision/ or exp positive predictive value/ or exp negative predictive value/ or positive likelihood ratio.mp. or exp negative predictive value/ or positive likelihood ratio.mp. or negative likelihood ratio.mp. or receiver operating.mp. or diagnostic odds.mp. or ppv.mp. or npv.mp. or plr.mp. or nlr.mp. or roc.mp. or exp sroc/ or dor.mp. or exp reliability/ or repeatability.mp. or exp reproducibility/ or reference standard.mp. or index test.mp. or reference test.mp. or exp gold standard/ or exp false positive result/ or exp false negative result/ or true positive.mp. or true negative.mp. or false positive.mp. or false negative.mp. or concord*.mp. or agreement.mp. or correlat*.mp. or accord*.mp. or (predictive adj4 value).mp.	5845182	7304460
64	62 and 63	1442	1702
65	(editorial or letter or comment or historical article).pt.	1600105	1895537
66	64 not 65	1415	1675
67	(animals/ or nonhuman/) not humans/	6124874	6443842
68	66 not 67	1402	1659
69	limit 68 to yr="2018 -Current"	NA	312

NA, not applicable

a. Embase <1974 to 2018 July 17>

b. Embase <1974 to 2021 September 24>

MEDLINE

Table A.7 Search results subquestion 3: Medline (via Ovid) for Level I, Level II and Level III studies

#	Searches	Results [a] 19 July 2018	Results [b] 28 Sept 2021
1	exp Prenatal Diagnosis/	68829	76931
2	Maternal Serum Screening Tests/	330	531
3	Hematologic Tests/	8696	9685
4	((prenatal or pre-natal or antenatal or ante-natal) adj3 (test* or screen* or diagnos* or determin* or detect*)).ti,ab.	36975	42346
5	(f?etal adj3 (test* or screen* or diagnos* or determin* or detect*)).ti,ab.	17863	20221
6	((non-invasive adj7 screening) or (non?invasive adj7 screening)).ti,ab.	2893	3832
7	(NIPD or NIPT or NIPS or NIPA).ti,ab.	1344	2172
8	or/1-7	104364	118015
9	Cell-Free Nucleic Acids/	198	1982
10	(cffCDNA or cell-free f?etal DNA).ti,ab.	666	847

#	Searches	Results [a] 19 July 2018	Results [b] 28 Sept 2021
11	((cell free dna or cfDNA) adj3 (obstetric or obstetrics or pregnancy or maternal)).ti,ab.	273	263
12	((cell free dna or cfDNA) adj3 (fetu* or fetal* or f?etu* or f?etal*)).ab,ti.	157	410
13	Genotyping Techniques/	5403	7844
14	((genotype* or genotyping) adj3 (obstetric or obstetrics or pregnancy or maternal)).ti,ab.	1115	1305
15	((genotype* or genotyping) adj3 (fetu* or fetal* or f?etu* or f?etal*)).ti,ab.	779	922
16	(RHD adj3 gene).ti,ab.	323	363
17	or/9-16	8282	12920
18	8 or 17	111426	129150
19	exp "obstetrics"/ or exp "obstetric care"/ or exp "pregnancy"/ or exp "pregnancy disorder"/ or exp "prenatal disorder"/	846400	945998
20	(obstetric or obstetrics or pregnancy or maternal).kw,ab,ti.	542881	643633
21	(prepartum or pre partum or pre-partum or intrapartum or intra partum or intra-partum or perinatal or peri natal or peri-natal).kw,ab,ti.	72582	87299
22	(antenatal or ante natal or ante-natal or prenatal or pre natal or pre-natal).kw,ab,ti.	113470	137293
23	(postnatal or post natal or post-natal or postpartum or post partum or post-partum).kw,ab,ti.	152751	181566
24	or/19-23	1127977	1280398
25	exp "fetus"/	151416	161305
26	(fetu* or fetal* or f?etu* or f?etal*).kw,ab,ti.	296787	333936
27	or/25-26	368773	409865
28	exp alloimmunization/	0	0
29	exp Rh Isoimmunization/	1672	1753
30	(Rh* Isoimmuni?ation or Rh* D Isoimmuni?ation).ti,ab.	602	614
31	(Rh* alloimmuni?ation or Rh* D alloimmuni?ation).ti,ab.	215	256
32	(Rh* incompatibility or Rh* D incompatibility or blood group incompatibility).ti,ab.	909	973
33	(((Rh* adj3 incompatib*) or Rh* D) adj3 incompatibl*).ti,ab.	155	166
34	((Rh or RhD or rhesus) adj5 sensiti*).ti,ab.	1195	1225
35	((fetomaternal or feto-maternal or foetomaternal or foeto-maternal) adj2 immuni?ation).ti,ab.	78	78
36	((rh or rhesus) adj2 (immuni?ation or autoimmuni?ation)).ti,ab.	713	780
37	or/28-36	4718	5020
38	exp rhesus D antigen/	0	38
39	rhesus D antigen.ti,ab.	37	0
40	rh* D antigen.ti,ab.	183	189
41	(RhD or rhesus D or Rh?D or Rh-?D or Rh D).ti,ab.	4499	5412
42	(Rh-negative or Rh-positive).ti,ab.	951	1026
43	(Rhesus negative or Rhesus positive).ti,ab.	238	258
44	((rh or rhesus) adj2 (factor or factors or antigen* or system or group)).ti,ab.	3881	4358
45	or/38-44	8683	10069
46	(Macaca mulatta or Simian Immunodeficiency Virus or zika or macaque?).ti,ab.	32790	41471
47	45 not 46	8526	9889
48	(isoimmuni?ation or alloimmuni?ation).ti,ab,kw.	3791	4313
49	(isoimmuni* or iso-immuni* or isoimmune or iso-immune).ti,ab,kw.	2000	2064
50	(alloimmuni* or allo-immuni* or alloimmune or allo-immune).ti,ab,kw.	6616	7647
51	(unsensiti?ed or un-sensiti?ed or non-sensiti?ed).ti,ab,kw.	1629	1750
52	(sensiti?ation* or sensiti?ed).ti,ab,kw.	90214	103333
53	or/48-52	98682	112853
54	exp Erythroblastosis, Fetal/	11580	12015
55	((erythroblastoses or erythroblastosis) adj2 (fetal* or f?etal*)).kw,ab,ti.	858	908
56	(h?emolytic disease* or h?emolytic disorder*).ti,ab,kw.	4554	4970
57	(HDFN or HDN).ti,ab,kw.	552	722
58	or/54-57	13562	14346
59	24 or 27	1260402	1423630
60	37 or 47 or 53 or 58	118578	134150

#	Searches	Results [a] 19 July 2018	Results [b] 28 Sept 2021
61	59 and 60	17363	18649
62	18 and 61	2898	3176
63	(diagnos*.mp. and (exp performance/ or yield.mp.)) or accura*.mp. or exp accuracy/ or exp diagnostic accuracy/ or sensitivity.mp. or specificity.mp. or exp "sensitivity and specificity"/ or exp "specificity and sensitivity"/ or exp precision/ or exp positive predictive value/ or exp negative predictive value/ or positive likelihood ratio.mp. or exp negative predictive value/ or positive likelihood ratio.mp. or negative likelihood ratio.mp. or receiver operating.mp. or diagnostic odds.mp. or ppv.mp. or npv.mp. or plr.mp. or nlr.mp. or roc.mp. or exp sroc/ or dor.mp. or exp reliability/ or repeatability.mp. or exp reproducibility/ or reference standard.mp. or index test.mp. or reference test.mp. or exp gold standard/ or exp false positive result/ or exp false negative result/ or true positive.mp. or true negative.mp. or false positive.mp. or false negative.mp. or concord*.mp. or agreement.mp. or correlat*.mp. or accord*.mp. or (predictive adj4 value).mp.	4531794	5539793
64	62 and 63	716	816
65	(editorial or letter or comment or historical article).pt.	1970016	2342764
66	64 not 65	702	802
67	(animals/ or nonhuman/) not humans/	4441716	4856723
68	66 not 67	699	799
69	limit 68 to yr="2018 -Current"	NA	**106**

NA, not applicable

a. Ovid MEDLINE(R) Epub Ahead of Print, In-Process & Other Non-Indexed Citations, Ovid MEDLINE(R) Daily, Ovid MEDLINE and Versions(R) <1946 to May 30, 2018>

b. Ovid MEDLINE(R) ALL <1946 to September 24, 2021>

Evidence-Based Medicine Reviews

Table A.8 Search results subquestion 3: EBM Reviews

#	Searches	Results [a] 19 July 2018	Results [b] 28 Sept 2021
1	exp Prenatal Diagnosis/	939	1055
2	Maternal Serum Screening Tests/	8	9
3	Hematologic Tests/	204	228
4	((prenatal or pre-natal or antenatal or ante-natal) adj3 (test* or screen* or diagnos* or determin* or detect*)).ti,ab.	852	1287
5	(f?etal adj3 (test* or screen* or diagnos* or determin* or detect*)).ti,ab.	692	1025
6	((non-invasive adj7 screening) or (non?invasive adj7 screening)).ti,ab.	159	260
7	(NIPD or NIPT or NIPS or NIPA).ti,ab.	137	281
8	or/1-7	2540	3613
9	Cell-Free Nucleic Acids/	3	18
10	(cffCDNA or cell-free f?etal DNA).ti,ab.	14	18
11	((cell free dna or cfDNA) adj3 (obstetric or obstetrics or pregnancy or maternal)).ti,ab.	11	16
12	((cell free dna or cfDNA) adj3 (fetu* or fetal* or f?etu* or f?etal*)).ab,ti.	11	12
13	Genotyping Techniques/	60	85
14	((genotype* or genotyping) adj3 (obstetric or obstetrics or pregnancy or maternal)).ti,ab.	31	57
15	((genotype* or genotyping) adj3 (fetu* or fetal* or f?etu* or f?etal*)).ti,ab.	13	24
16	(RHD adj3 gene).ti,ab.	3	4
17	or/9-16	122	2-Jun
18	8 or 17	2636	3779
19	exp "obstetrics"/ or exp "obstetric care"/ or exp "pregnancy"/ or exp "pregnancy disorder"/ or exp "prenatal disorder"/	19993	23474
20	(obstetric or obstetrics or pregnancy or maternal).kw,ab,ti.	37479	71083
21	(prepartum or pre partum or pre-partum or intrapartum or intra partum or intra-partum or perinatal or peri natal or peri-natal).kw,ab,ti.	4694	7790
22	(antenatal or ante natal or ante-natal or prenatal or pre natal or pre-natal).kw,ab,ti.	5834	10176
23	(postnatal or post natal or post-natal or postpartum or post partum or post-partum).kw,ab,ti.	8586	15414

#	Searches	Results [a] 19 July 2018	Results [b] 28 Sept 2021
24	or/19-23	50733	88340
25	exp "fetus"/	1614	1812
26	(fetu* or fetal* or f?etu* or f?etal*).kw,ab,ti.	8812	15176
27	or/25-26	9664	16152
28	exp alloimmunization/	0	0
29	exp Rh Isoimmunization/	30	32
30	(Rh* Isoimmuni?ation or Rh* D Isoimmuni?ation).ti,ab.	13	18
31	(Rh* alloimmuni?ation or Rh* D alloimmuni?ation).ti,ab.	6	14
32	(Rh* incompatibility or Rh* D incompatibility or blood group incompatibility).ti,ab.	25	52
33	(((Rh* adj3 incompatib*) or Rh* D) adj3 incompatibl*).ti,ab.	2	4
34	((Rh or RhD or rhesus) adj5 sensiti*).ti,ab.	23	28
35	((fetomaternal or feto-maternal or foetomaternal or foeto-maternal) adj2 immuni?ation).ti,ab.	0	0
36	((rh or rhesus) adj2 (immuni?ation or autoimmuni?ation)).ti,ab.	27	33
37	or/28-36	113	164
38	exp rhesus D antigen/	0	0
39	rhesus D antigen.ti,ab.	0	0
40	rh* D antigen.ti,ab.	0	0
41	(RhD or rhesus D or Rh?D or Rh-?D or Rh D).ti,ab.	139	217
42	(Rh-negative or Rh-positive).ti,ab.	23	46
43	(Rhesus negative or Rhesus positive).ti,ab.	17	22
44	((rh or rhesus) adj2 (factor or factors or antigen* or system or group)).ti,ab.	117	162
45	or/38-44	283	418
46	(Macaca mulatta or Simian Immunodeficiency Virus or zika or macaque?).ti,ab.	151	281
47	45 not 46	283	418
48	(isoimmuni?ation or alloimmuni?ation).ti,ab,kw.	175	262
49	(isoimmuni* or iso-immuni* or isoimmune or iso-immune).ti,ab,kw.	46	65
50	(alloimmuni* or allo-immuni* or alloimmune or allo-immune).ti,ab,kw.	266	406
51	(unsensiti?ed or un-sensiti?ed or non-sensiti?ed).ti,ab,kw.	49	71
52	(sensiti?ation* or sensiti?ed).ti,ab,kw.	2899	4409
53	or/48-52	3200	4861
54	exp Erythroblastosis, Fetal/	70	76
55	((erythroblastoses or erythroblastosis) adj2 (fetal* or f?etal*)).kw,ab,ti.	14	10
56	(h?emolytic disease* or h?emolytic disorder*).ti,ab,kw.	106	157
57	(HDFN or HDN).ti,ab,kw.	21	32
58	or/54-57	150	210
59	24 or 27	52330	90952
60	37 or 47 or 53 or 58	3577	5408
61	59 and 60	367	565
62	18 and 61	38	48
63	(diagnos*.mp. and (exp performance/ or yield.mp.)) or accura*.mp. or exp accuracy/ or exp diagnostic accuracy/ or sensitivity.mp. or specificity.mp. or exp "sensitivity and specificity"/ or exp "specificity and sensitivity"/ or exp precision/ or exp positive predictive value/ or exp negative predictive value/ or positive likelihood ratio.mp. or exp negative predictive value/ or positive likelihood ratio.mp. or negative likelihood ratio.mp. or receiver operating.mp. or diagnostic odds.mp. or ppv.mp. or npv.mp. or plr.mp. or nlr.mp. or roc.mp. or exp sroc/ or dor.mp. or exp reliability/ or repeatability.mp. or exp reproducibility/ or reference standard.mp. or index test.mp. or reference test.mp. or exp gold standard/ or exp false positive result/ or exp false negative result/ or true positive.mp. or true negative.mp. or false positive.mp. or false negative.mp. or concord*.mp. or agreement.mp. or correlat*.mp. or accord*.mp. or (predictive adj4 value).mp.	233823	340240
64	62 and 63	25	24
65	(editorial or letter or comment or historical article).pt.	7477	8703
66	64 not 65	25	24
67	(animals/ or nonhuman/) not humans/	25	27
68	66 not 67	25	24

#	Searches	Results [a] 19 July 2018	Results [b] 28 Sept 2021
69	limit 68 to yr="2018 -Current"	NA	2

NA, not applicable

a. EBM Reviews combines several resources into a single database and includes the following: ACP Journal Club <1991 to June 2018>; Cochrane Database of Systematic Reviews <2005 to July 18, 2018>; Database of Abstracts of Reviews of Effects <1st Quarter 2016>; Cochrane Clinical Answers <June 2018>; Cochrane Central Register of Controlled Trials <June 2018>; Cochrane Methodology Register <3rd Quarter 2012>; Health Technology Assessment <4th Quarter 2016>; NHS Economic Evaluation Database <1st Quarter 2016>.

b. EBM Reviews - Cochrane Database of Systematic Reviews <2005 to September 23, 2021>; EBM Reviews - ACP Journal Club <1991 to August 2021>; EBM Reviews - Database of Abstracts of Reviews of Effects <1st Quarter 2016>; EBM Reviews - Cochrane Clinical Answers <September 2021>; EBM Reviews - Cochrane Central Register of Controlled Trials <August 2021>; EBM Reviews - Cochrane Methodology Register <3rd Quarter 2012>; EBM Reviews - Health Technology Assessment <4th Quarter 2016>; EBM Reviews - NHS Economic Evaluation Database <1st Quarter 2016>

PubMed

The PubMed search is restricted to records that are not indexed for MEDLINE (i.e. in-process citations and citations from journals (or parts of journals) that are not currently MEDLINE-indexed) and to records added to PubMed since January 2006.

The search comprises free-text terms only and replicates the free-text sets in the Embase search (converted from the Ovid syntax).

Table A.9 Search results subquestion 3: Pubmed (not MEDLINE)

#	Search terms	Results 20 July 2018	Results 28 Sept 2021
#1	(Maternal[tiab] OR obstetric[tiab] OR obstetrics[tiab] OR pregnant[tiab] OR pregnancy[tiab] OR prenatal[tiab] OR pre-natal[tiab]) AND (serum[tiab] OR sera[tiab]) AND (test[tiab] OR tests[tiab] OR testing[tiab] OR screen*[tiab] OR diagnos*[tiab] OR determin*[tiab] OR detect*[tiab])	21171	31,755
#2	(Blood[tiab] OR serum[tiab] OR sera[tiab] OR haematologic*[tiab] OR hematologic*[tiab]) AND (test[tiab] OR tests[tiab] OR testing[tiab])	344458	413,112
#3	(prenatal[tiab] OR pre-natal[tiab] OR antenatal[tiab] OR ante-natal[tiab]) AND (test[tiab] OR tests[tiab] OR testing[tiab] OR screen*[tiab] OR diagnos*[tiab] OR determin*[tiab] OR detect*[tiab])	70051	85,001
#4	(foetal[tiab] OR fetal[tiab]) AND (test[tiab] OR tests[tiab] OR testing[tiab] OR screen*[tiab] OR diagnos*[tiab] OR determin*[tiab] OR detect*[tiab])	103239	119,205
#5	(noninvasive[tiab] OR non-invasive[tiab]) AND (screening[tiab])	8460	11,434
#6	NIPD[tiab] OR NIPT[tiab] OR NIPS[tiab] OR NIPA[tiab]	1368	2,356
#7	#1 OR #2 OR #3 OR #4 OR #5 OR #6	502555	603,704
#8	cffCDNA[tiab] OR cell free fetal DNA[tiab] OR cell free foetal DNA[tiab]	706	919
#9	(cell free dna[tiab] OR cfDNA[tiab]) AND (obstetric[tiab] OR obstetrics[tiab] OR pregnancy[tiab] OR maternal[tiab])	576	950
#10	(cell free dna[tiab] OR cfDNA[tiab]) AND (fetu*[tiab] OR fetal*[tiab] OR foetu*[tiab] OR foetal*[tiab])	593	976
#11	(genotype[tiab] OR genotyping[tiab] OR allele[tiab] OR alleles[tiab]) AND (test[tiab] OR tests[tiab] OR testing[tiab] OR screen*[tiab] OR diagnos*[tiab] OR determin*[tiab] OR detect*[tiab])	180704	216,412
#12	(genotype*[tiab] OR genotyping[tiab]) AND (obstetric[tiab] OR obstetrics[tiab] OR pregnancy[tiab] OR maternal[tiab])	7324	8,971
#13	(genotype*[tiab] OR genotyping[tiab]) AND (fetu*[tiab] OR fetal*[tiab] OR foetu*[tiab] OR foetal*[tiab])	3628	4,398
#14	(RHD[tiab] AND gene[tiab])	607	730
#15	#8 OR #9 OR #10 OR #11 OR #12 OR #13 OR #14	186624	223,843
#16	#7 OR #15	675038	810,096
#17	Obstetric[tiab] OR obstetrics[tiab] OR pregnancy[tiab] OR maternal[tiab]	554296	657,138
#18	prepartum[tiab] OR pre partum[tiab] OR pre-partum[tiab] OR intrapartum[tiab] OR intra partum[tiab] OR intra-partum[tiab] OR perinatal[tiab] OR peri natal[tiab] OR peri-natal[tiab]	73169	88,677
#19	antenatal[tiab] OR ante natal[tiab] OR ante-natal[tiab] OR prenatal[tiab] OR pre natal[tiab] OR pre-natal[tiab]	115113	139,946
20#	postnatal[tiab] OR post natal[tiab] OR post-natal[tiab] OR postpartum[tiab] OR post partum[tiab] OR post-partum[tiab]	153623	183,050
#21	#17 OR #18 OR #19 OR #20	738834	868,132

#	Search terms	Results 20 July 2018	Results 28 Sept 2021
#22	fetus[tiab] OR foetus[tiab] OR fetu*[tiab] OR foetu*[tiab] OR fetal*[tiab] OR foetal*[tiab]	299655	337,391
#23	alloimmunization[tiab] OR alloimmunisation[tiab]	2590	3,241
#24	(rh[tiab] OR rhd[tiab] rhesus[tiab]) AND (isoimmunization[tiab] OR isoimmunisation[tiab])	63	82
#25	(rh[tiab] OR rhd[tiab] OR rhesus[tiab]) AND (incompatibility[tiab]) OR (blood group incompatibility[tiab])	1636	1,747
#26	(rh[tiab] OR rhd[tiab] OR rhesus[tiab]) AND (incompatib*[tiab])	1307	1,428
#27	(Rh[tiab] OR RhD[tiab] OR rhesus[tiab]) AND (sensiti*[tiab])	4933	5,735
#28	(fetomaternal[tiab] OR feto-maternal[tiab] OR foetomaternal[tiab] OR foeto-maternal[tiab]) AND (immunisation[tiab] OR immunization[tiab])	166	174
#29	(rh[tiab] OR RhD[tiab] OR rhesus[tiab]) AND (immunisation[tiab] OR immunization[tiab] OR autoimmunisation[tiab] OR autoimmunization[tiab])	2055	2,251
#30	#23 OR #24 OR #25 OR #26 OR #27 OR #28 OR #29	11065	12,723
#31	(rh[tiab] OR rhd[tiab] OR rhesus[tiab]) AND antigen[tiab]	3429	3,816
#32	RhD[tiab] OR rhesus d[tiab] OR rh-d[tiab] OR Rh D[tiab]	3887	4,753
#33	rh-negative[tiab] OR rh-positive[tiab] OR rh negative[tiab] OR rh positive[tiab]	949	1,038
#34	rhesus negative[tiab] OR rhesus positive[tiab]	240	261
#35	(rh[tiab] OR rhesus[tiab]) AND (factor[tiab] OR factors[tiab] OR antigen*[tiab] OR antigens[tiab] OR system[tiab] OR group[tiab])	24249	27,871
#36	#31 OR #32 OR #33 OR #34 OR #35	27458	31,809
#37	Macaca mulatta[tiab] OR Simian Immunodeficiency Virus[tiab] OR zika[tiab] OR macaque[tiab] OR macaques[tiab]	33079	42,084
#38	#36 NOT #37	23409	27,075
#39	isoimmuni*[tiab] OR iso-immuni*[tiab] OR isoimmune[tiab] OR iso-immune[tiab]	2006	2,084
#40	alloimmuni*[tiab] OR allo-immuni*[tiab] OR alloimmune[tiab] OR allo-immune[tiab]	6637	7,695
#41	(rh[tiab] OR rho[tiab] OR rhesus[tiab]) AND (sensitising[tiab] OR sensitizing[tiab] OR sensitisation[tiab] OR sensitization[tiab] OR sensitised[tiab] OR sensitized[tiab])	1599	1,735
#42	sensitisation*[tiab] OR sensitization*[tiab] OR sensitised[tiab] OR sensitized[tiab]	90526	104,472
#43	#39 OR #40 OR #41 OR #42	98817	113,847
#44	(erythroblastoses[tiab] OR erythroblastosis[tiab]) AND (fetal[tiab] OR foetal[tiab] OR fetalis[tiab] OR foetalis[tiab])	3161	3,172
#45	(hemolytic OR haemolytic) AND (disorder[tiab] OR disorders[tiab] OR disease[tiab] OR diseases[tiab])	18492	19,246
#46	hdfn[tiab] OR hdn[tiab]	553	750
#47	#44 OR #45 OR #46	20920	21,750
#48	#21 OR #22	887315	1,028,420
#49	#30 OR #38 OR #43 OR #47	143560	163,294
#50	#48 AND #49	12773	13,944
#51	#16 AND #50	3926	4,511
#52	Diagnos*[tiab] AND (performance[tiab] or yield[tiab]) OR accura*[tiab] OR diagnostic accuracy[tiab] OR sensitivity[tiab] OR specificity [tiab] OR precision[tiab] OR positive predictive value [tiab] OR negative predictive value[tiab] OR positive likelihood ratio[tiab] OR negative likelihood ratio[tiab] OR receiver operating[tiab] OR diagnostic odds[tiab] OR ppv[tiab] OR npv[tiab] OR plr[tiab] OR nlr[tiab] OR ROC[tiab] OR sroc[tiab] OR dor[tiab] OR reliability[tiab] OR repeatability[tiab] OR reproducibility[tiab] OR reference standard[tiab] OR index test[tiab] OR reference test[tiab] OR gold standard[tiab] OR false positive[tiab] OR false negative[tiab] OR true positive[tiab] OR true negative[tiab] OR concord*[tiab] OR agreement[tiab] OR correlate*[tiab] OR accord*[tiab] OR (predictive[tiab] AND value[tiab])	3585762	4,486,125
#53	#51 AND #52	981	1,150
#54	#53 AND pubmednotmedline[sb]	40	72
#55	#53 AND pubmednotmedline[sb] from 2018 - 2021	NA	**28**

NA, not applicable

CINAHL

Table A.10Search results subquestion 3: CINAHL

#	Query	Results 19 July 2018	Results 28 Sept 2021
S1	(MH "Prenatal Diagnosis+")	8232	20,759
S2	"Maternal Serum Screening Tests"	0	5
S3	(MH "Hematologic Tests+")	22714	49,125
S4	TI (((prenatal or pre-natal or antenatal or ante-natal) N3 (test* or screen* or diagnos* or determin* or detect*))) OR AB (((prenatal or pre-natal or antenatal or ante-natal) N3 (test* or screen* or diagnos* or determin* or detect*)))	4126	11,292
S5	TI ((f#etal N3 (test* or screen* or diagnos* or determin* or detect*))) OR AB ((f#etal N3 (test* or screen* or diagnos* or determin* or detect*)))	2104	5,707
S6	TI (((non-invasive N7 screening) or (non#invasive N7 screening))) OR AB (((non-invasive N7 screening) or (non#invasive N7 screening)))	333	954
S7	TI (NIPD or NIPT or NIPA or NIPS) OR AB (NIPD or NIPT or NIPA or NIPS)	222	896
S8	S1 OR S2 OR S3 OR S4 OR S5 OR S6 OR S7	34153	78,684
S9	"Cell Free Nucleic Acids" OR "Cell Free dna"	247	1,519
S10	TI ((cffCDNA or cell free f#etal DNA)) OR AB ((cffCDNA or cell free f#etal DNA))	134	495
S11	TI (((cell free dna or cfDNA) N3 (obstetric or obstetrics or pregnancy or maternal))) OR AB (((cell free dna or cfDNA) N3 (obstetric or obstetrics or pregnancy or maternal)))	96	408
S12	TI (((cell free dna or cfDNA) N3 (fetu* or fetal* or f#etu* or f#etal*))) OR AB (((cell free dna or cfDNA) N3 (fetu* or fetal* or f#etu* or f#etal*)))	378	491
S13	(MH "Molecular Diagnostic Techniques")	729	2,491
S14	TI (((genotype* or genotyping) N3 (obstetric or obstetrics or pregnancy or maternal))) OR AB (((genotype* or genotyping) N3 (obstetric or obstetrics or pregnancy or maternal)))	128	284
S15	TI (((genotype* or genotyping) N3 (fetu* or fetal* or f#etu* or f#etal*))) OR AB (((genotype* or genotyping) N3 (fetu* or fetal* or f#etu* or f#etal*)))	3291	226
S16	TI RHD N3 gene OR AB RHD N3 gene	49	84
S17	S9 OR S10 OR S11 OR S12 OR S13 OR S14 OR S15 OR S16	4459	4,728
S18	S8 OR S17	38260	82,409
S19	(MH "Obstetrics") or (MH "Obstetric Care+") or (MH "Pregnancy+") or “pregnancy disorder” or “prenatal disorder”	128201	240,985
S20	TI (obstetric or obstetrics or pregnancy or maternal) OR AB (obstetric or obstetrics or pregnancy or maternal) or (“obstetric” or “obstetrics” or “pregnancy” or “maternal”)	157074	311,683
S21	TI (prepartum or pre partum or pre-partum or intrapartum or intra partum or intra-partum or perinatal or peri natal or peri-natal) OR AB (prepartum or pre partum or pre-partum or intrapartum or intra partum or intra-partum or perinatal or peri natal or peri-natal) OR ("prepartum" or "pre partum" or "pre-partum" or "intrapartum" or "intra partum" or "intra-partum" or "perinatal" or "peri natal" or "peri-natal")	20549	39,685
S22	TI (antenatal or ante natal or ante-natal or prenatal or pre natal or pre-natal) OR AB (antenatal or ante natal or ante-natal or prenatal or pre natal or pre-natal) OR ("antenatal" or "ante natal" or "ante-natal" or "prenatal" or "pre natal" or "pre-natal")	29496	66,056
S23	TI (postnatal or post natal or post-natal or postpartum or post partum or post-partum) OR AB (postnatal or post natal or post-natal or postpartum or post partum or post-partum) OR ("postnatal" or "post natal" or "post-natal" or "postpartum" or "post partum" or "post-partum")	23191	50,224
S24	S19 OR S20 OR S21 OR S22 OR S23	176163	352,395
S25	(MH "Fetus+")	17287	26,623
S26	TI (fetu* or fetal* or f#etu* or f#etal*) OR AB (fetu* or fetal* or f#etu* or f#etal*) OR ("fetu*" or "fetal*" or "f#etu*" or "f#etal*")	2597789	82,572
S27	S25 OR 26	40390	82,928
S28	"alloimmuni?ation" TI alloimmuni?ation OR AB alloimmuni?ation OR "alloimmuni?ation"	343	692
S29	(MH "RH Isoimmunization")	275	458
S30	TI (Rh* Isoimmuni?ation or Rh* D Isoimmuni?ation) OR AB (Rh* Isoimmuni?ation or Rh* D Isoimmuni?ation)	29	61
S31	TI (Rh* alloimmuni?ation or Rh* D alloimmuni?ation) OR AB (Rh* alloimmuni?ation or Rh* D alloimmuni?ation)	55	111
S32	TI (Rh* incompatibility or Rh* D incompatibility or blood group incompatibility) OR AB (Rh* incompatibility or Rh* D incompatibility or blood group incompatibility)	49	115

#	Query	Results 19 July 2018	Results 28 Sept 2021
S33	TI ((Rh* N3 incompatib*) OR (Rh* D N3 incompatibl*)) OR AB ((Rh* N3 incompatib*) OR (Rh* D N3 incompatibl*))	37	82
S34	TI ((Rh or RhD or rhesus) N5 sensiti*) OR AB ((Rh or RhD or rhesus) N5 sensiti*)	60	124
S35	TI (fetomaternal or feto-maternal or foetomaternal or foeto-maternal) N2 immuni?ation) OR AB (fetomaternal or feto-maternal or foetomaternal or foeto-maternal) N2 immuni?ation)	2	2
S36	TI (((rh or RhD or rhesus) N2 (immuni?ation or autoimmuni?ation))) OR AB (((rh or rhesus) N2 (immuni?ation or autoimmuni?ation)))	10	17
S37	S28 OR S29 OR S30 OR S31 OR S32 OR S33 OR S34 OR S35 OR S36	656	1,247
S38	"rhesus D antigen"	2	4
S39	TI rhesus D antigen OR AB rhesus D antigen	3	9
S40	TI rh* D antigen OR AB rh* D antigen	36	64
S41	TI (RhD or rhesus D or Rh D or Rh-D) ORAB (RhD or rhesus D or Rh D or Rh-D)	528	1,117
S42	TI (Rh negative OR Rh positive) OR AB (Rh negative OR Rh positive))	88	194
S43	TI (Rhesus negative or Rhesus positive) OR AB (Rhesus negative or Rhesus positive)	32	78
S44	TI (rh or rhesus) N2 (factor or factors or antigen* or system or group)) OR AB (rh or rhesus) N2 (factor or factors or antigen* or system or group))	156	439
S45	S38 OR S39 OR S40 OR S41 OR S42 OR S43 OR S44	688	1,551
S46	TI (Macaca mulatta or Simian Immunodeficiency Virus or zika or macaque#) OR AB (Macaca mulatta or Simian Immunodeficiency Virus or zika or macaque#)	1514	4,005
S47	S45 NOT S46	683	1,526
S48	TI (isoimmuni?ation or alloimmuni?ation) OR AB (isoimmuni?ation or alloimmuni?ation) OR ("isoimmuni?ation" or "alloimmuni?ation")	579	1,099
S49	TI (isoimmuni* or iso-immuni* or isoimmune or iso-immune) OR AB (isoimmuni* or iso-immuni* or isoimmune or iso-immune) OR ("isoimmuni*" or "iso-immuni*" or "isoimmune" or "iso-immune")	310	552
S50	TI (alloimmuni* or allo-immuni* or alloimmune or allo-immune) OR AB (alloimmuni* or allo-immuni* or alloimmune or allo-immune) OR ("alloimmuni*" or "allo-immuni*" or "alloimmune" or "allo-immune")	607	1,251
S51	TI (unsensiti?ed or un-sensiti?ed or non-sensiti?ed) OR AB (unsensiti?ed or un-sensiti?ed or non-sensiti?ed) OR ("unsensiti?ed" or "un-sensiti?ed" or "non-sensiti?ed")	20	61
S52	TI (sensiti?ation* or sensiti?ed) OR AB (sensiti?ation* or sensiti?ed) OR ("sensiti?ation*" or "sensiti?ed")	3421	8,596
S53	S48 OR S49 OR S50 OR S51 OR S52	4222	10,186
S54	(MH "Erythroblastosis, Fetal+")	616	1,202
S55	TI (((erythroblastoses or erythroblastosis) N2 (fetal* or f#etal*))) OR AB (((erythroblastoses or erythroblastosis) N2 (fetal* or f#etal*))) OR ((("erythroblastoses" or "erythroblastosis") N2 ("fetal*" or "f#etal*")))	240	437
S56	TI ((h#emolytic disease* or h#emolytic disorder*)) OR AB ((h#emolytic disease* or h#emolytic disorder*)) OR (("h#emolytic disease*" or "h#emolytic disorder*"))	376	825
S57	TI (HDFN or HDN) OR AB (HDFN or HDN) OR ("HDFN" or "HDN")	57	141
S58	S54 OR S55 OR S56 OR S57	873	1,824
S59	S24 OR S27	199864	408,390
S60	S37 OR S47 OR S53 OR S58	5450	12,886
S61	S59 AND S60	1142	2,427
S62	S18 AND S61	361	806
S63	(diagnos* and (performance or yield)) or (accura* or “diagnostic accuracy”) or “sensitivity” or “specificity” or (MH "Sensitivity and Specificity") or (MH "Precision") or (MH "Predictive Value of Tests") or “positive predictive value” or “negative predictive value” or “positive likelihood ratio” or “negative likelihood ratio” or (MH "ROC Curve") or "receiver operating" or “diagnostic odds” or ppv or npv or plr or nlr or roc or sroc or dor or reliability or repeatability or reproducibility or “reference standard” or “index test” or “reference test” or “gold standard” or “false positive result” or (MH "False Positive Results") or “false negative result” or (MH “False Negative Results”) or “true positive” or “true negative” or “false positive” or “false negative” or concord* or agreement or correlate* or accord* or (predictive N4 value) or (MH "Predictive Validity")	374650	891,666
S64	S62 and S63	96	235
S65	PT (Editorial or letter or comment or historical article)	364150	689,083
S66	S64 NOT S65	94	230
S67	S64 NOT S65 Limiters - Date Published: 20180101-20211231	NA	49

NA, not applicable

Ovid syntax

Exp explodes controlled vocabulary term (i.e. includes all narrower terms in the hierarchy)
* denotes a term that has been searched as a major subject heading
/ denotes controlled vocabulary terms (EMTREE)
$ truncation character (unlimited truncation)
$n truncation limited to specified number (n) of characters (e.g. time$1 identifies time, timed, timer, times but not timetable)
* truncation character (unlimited truncation)
? substitutes any letter (e.g. oxidi?ed identifies oxidised and oxidized)
adjn search terms within a specified number (n) of words from each other in any order
.ti. limit to title field
.ti,ab. limit to title and abstract fields
.kw,ti,ab. limit to keyword, title and abstract field
.pt limit to publication type

PubMed syntax

* truncation character (unlimited truncation)
[TI] limit to title field
[TIAB] limit to title and abstract fields
[EDAT] date citation added to PubMed
[SB] PubMed subset

CINHAL syntax

* truncation character (unlimited truncation)
# wildcard character will replace 1 or 0 characters (e.g. f#etus will retrieve fetus and foetus)
? wildcard character will replace one character (e.g. wom?n will retrieve women and woman)
MH - Search the exact CINAHL® subject heading; searches both major and minor headings
MH"heading"+ Search an exploded subheading
TI search title fields
AB search abstract fields
Nn – Proximity "near" operator will find a result if the terms are within a certain number (n) words of each other, regardless of the order in which they appear. (e.g. eating N5 disorders for results that contain eating disorders, as well as mental disorders and eating pathology.)
PT limit to publication type

Appendix B Literature screening results (2021 update)

This appendix documents the updated literature search screening results for a systematic review on the prophylactic use of Rh D Immunoglobulin (Anti-D) in pregnant women.

A PRIMSA flow illustrating the screening results is provided in **Figure 1** (all questions) and **Figure 2** (subquestion 3, diagnostic accuracy).

Table B.1 Literature search and title/abstract screening results

Number of citations identified	Questions 1-4		Q3	
	Level I [a]	Level II (not Level I)	Level III (not Level II)	Diagnostic accuracy
Database				
Embase 1974 to 18 July 2018	27	106	302	312
MEDLINE 1946 to 18 July 2018	12	37	53	106
Cochrane 18 July 2018	0	19	0	2
PubMed	0	0	108	28
CINAHL	0	0	147	49
TOTAL	**39**	**162**	**610**	**497**
Date limit [b]	2018 to current	2018 to current	2018 to current	2018 to current
Duplicates removed in Endnote (across databases)	10	44	133	143
Duplicates removed by Covidence [c]	6	22	79	31
TITLE/ABSTRACT SCREENING				
Number of citations screened in Covidence	**23**	**96**	**398**	**323**
Additional duplicates identified	1	3	2	1
Nonhuman	0	0	14	0
Population out of scope	5	36	112	32
Intervention out of scope	8	27	202	214
Comparator out of scope	0	0	0	0
Outcome out of scope	0	0	0	0
Publication type out of scope. Not a systematic review.	0	0	0	2
Publication type out of scope. Opinion piece.	0	2	6	7
Publication type out of scope. Editorial.	0	0	0	0
Publication type out of scope. Other.	1	0	0	0
Study type out of scope. Level IV or below.	0	8	19	23
TOTAL irrelevant	**15**	**76**	**355**	**279**

a. NHMRC evidence level filters were applied in the Ovid interface. Studies identified in the Cochrane Collection and those retrieved via PubMed and CINAHL did not have filters applied but were screened in the first pass. (see Technical report, volume one)

b. A date limit was applied to studies based on the previous literature search date (July 2018), with the prior six months included to account for potential database changes (see Technical report, volume one).

c. https://www.covidence.org/home

Table B.2 Full text screening results

Number of citations identified	Questions 1-4			Q3
	Level I [a]	Level II (not Level I)	Level III (not Level II)	Diagnostic accuracy
FULL TEXT REVIEW				
Number of citations screened in Covidence [b]	**8**	**20**	**43**	**44**
Duplicate citation	0	1	2	2
Not available in English	0	0	0	0
Population out of scope	0	1	3	3
Intervention out of scope	0	5	12	8
Comparator out of scope	0	2	0	0
Outcome out of scope	1	2	1	5
Publication type out of scope. Simple review.	0	0	1	0
Publication type out of scope. Opinion piece.	0	0	3	1
Publication type out of scope. Editorial.	0	0	2	0
Level II or III study already included in Level I	0	0	0	0
Study design out of scope (Level IV or below)	0	3	17	9
No usable data (conference abstract etc.)	0	0	2	9
Superseded	0	0	0	0
Duplicate data (published elsewhere)	0	0	0	0
Small sample size	0	3	0	4
Not comparable to the Australian context	0	1	0	0
TOTAL EXCLUDED	**1**	**18**	**43**	**41**
TOTAL INCLUDED	**7**	**2**	**0**	**3**

a. NHMRC evidence level filters were applied in the Ovid interface. Studies identified in the Cochrane Collection and those retrieved via PubMed and CINAHL did not have filters applied but were screened in the first pass. (see Technical report, volume one)

b. https://www.covidence.org/home

Appendix C Excluded studies

This appendix documents studies that are awaiting cliassification or those that met the prespecified inclusion criteria for a systematic review on the prophylactic use of Rh D Immunoglobulin (Anti-D) in pregnant women but were later excluded. These studies, and their reasons for exclusion, are listed below.

C1 Studies relevant to all Questions

No usable data (conference abstracts etc.)

Donohoe, O (2021). Cost-effectiveness of targeted antenatal anti-d in ireland. *BJOG: An International Journal of Obstetrics and Gynaecology* **128**(SUPPL 2): 125.

Donohoe, O, L Mulvany, E O'Connor*, et al.* (2019). One-year audit of targeted routine antenatal anti-d prophylaxis in portiuncula university hospital. *BJOG: An International Journal of Obstetrics and Gynaecology* **126**(Supplement 1): 99-100.

Gordon, L, R Flower and C Hyland (2018). Non-invasive fetal rhd genotyping of rhd negative pregnant women for targeted anti-d therapy in australia: A cost-effectiveness analysis. *Value in Health* **21**(Supplement 2): S93.

Matteocci, A, G Nespoli, K Castagna*, et al.* (2020). Cost and saving analysis of rhd genotyping and anti-d immuno-prophylaxis in d-variant women of childbearing age in central italy. *Vox Sanguinis* **115**(SUPPL 1): 279.

C2 Studies relevant to Question 3 (or subquestion 3)

No usable data (conference abstracts etc.)

Balsalobre, EL, RR Sanchez, MdMV Penas*, et al.* (2019). Implementation of the rhd fetal protocol in rhd negative gestants. *Clinica Chimica Acta* **493**(Supplement 1): S585-S586.

Bingulac-Popovic, J, V Dogic, I Babic*, et al.* (2018). Prenatal rhd genotyping: Automated extraction of cell-free fetal DNA using the qiasymphony sp platform. *Clinical Chemistry and Laboratory Medicine* **56**(6): eA111.

Choo, BL, M Williamson, EA Martindale*, et al.* (2019). Provision of a fetal rhd genotyping service: The east lancashire experience. *BJOG: An International Journal of Obstetrics and Gynaecology* **126**(Supplement 1): 83.

Doescher, A and C Vogt (2018). Pitfalls in prenatal diagnosis of fetal rhd: Frequency of maternal rhd variants as cause for a false positive genotype of the fetus. *Transfusion Medicine and Hemotherapy* **45**(Supplement 1): 37.

Joshi, N, S Bassiony, A Mathyalakan*, et al.* (2021). Re-audit of cell free foetal DNA (cffdna) screen to avoid administration of anti-d immunoglobulin in rhd-negative pregnant women with rhd-negative foetus. *British Journal of Haematology* **193**(SUPPL 1): 14.

Londero, D, D Bolzicco, M Candolini*, et al.* (2018). First trimester noninvasive fetal rhd genotyping using frozen DNA samples: Validation and optimization of the test to implement a screening program. *Vox Sanguinis* **113**(Supplement 1): 276-277.

Maric, I, K Zeleznik, I Bricl*, et al.* (2018). Targeted prophylaxis program for d-negative pregnant women based on genotyping fetal rhd from maternal blood. *Vox Sanguinis* **113**(Supplement 1): 277.

Small sample size (N<200)

Addai-Mensah, O, EY Afriyie, ME Annani-Akollor*, et al.* (2020). Fetal rhesus d genotyping and sex determination from maternal plasma of rhesus d-negative antenatal population: The usefulness of conventional polymerase chain reaction in resource-limited settings. *Obstetrics and Gynecology International* **2020**: 4913793.

Ahmadi, MH and N Amirizadeh (2018). Evaluation the sry to confirm the presence of fetal DNA in the fetal rhd genotyping using cffdna. *Vox Sanguinis* **113 (Supplement 1)**: 277.

Bingulac-Popovic, J, I Babic, V Dogic*, et al.* (2021). Prenatal rhd genotyping in croatia: Preliminary results. *Transfusion Clinique et Biologique* **28**(1): 38-43.

Blanco, S, MC Frutos, SV Gallego*, et al.* (2018). Usefulness of non-invasive fetal rhd genotyping towards immunoprophylaxis optimization. *Transfusion Medicine and Hemotherapy* **45**(6): 423-428.

Londero, D, D Bolzicco, M Candolini*, et al.* (2019). Fetal rhd detection from circulating cell-free fetal DNA in maternal plasma: Validation of a diagnostic kit using automatic extraction and frozen DNA. *Transfusion Medicine* **29**(6): 408-414.

Plesinac, S, D Plecas and I Babovic (2018). The determination of fetal rhd status from maternal blood in serbia. *Indian Journal of Hematology and Blood Transfusion* **34**(3): 486-490.

Rather, R, S Saha and V Dhawan (2019). Non-invasive prenatal rhesus d genotyping using cell-free foetal DNA. *Indian Journal of Medical Research* **150**(1): 62-66.

Not comparable to the Australian context

Bohmova, J, R Kratochvilova, E Krejcirikova*, et al.* (2020). Two reliable methodical approaches for non-invasive rhd genotyping of a fetus from maternal plasma. *Diagnostics* **10**(8): 564.

Appendix D Critical appraisal

D1 Question 1

Level I – Systematic review (of RCTs and cohort studies)

Question	Xie 2020	
1. Did the research questions and inclusion criteria for the review include the components of the PICO?	Yes	PICO and inclusion criteria provided.
2. Did the report of the review contain an explicit statement that the review methods were established prior to the conduct of the review and did the report justify any significant deviations from the protocol?	Partial yes	The authors refer to predetermined research objectives which note that they are searching for RCTs. They include cohort studies but do not provide a reason for this change.
3. Did the review authors explain their selection of the study designs for inclusion in the review?	No	No explanation provided
4. Did the review authors use a comprehensive literature search strategy?	Yes	A comprehensive search strategy was employed to search the PubMed, EMBASE, Web of Science, China National Knowledge Infrastructure (CNKI) and Wanfang databases. T
5. Did the review authors perform study selection in duplicate?	Yes	Two authors independently assessed all studies for inclusion and data extraction (p2)
6. Did the review authors perform data extraction in duplicate?	Yes	Two investigators collected data independently in accordance with predesigned tables (p3)
7. Did the review authors provide a list of excluded studies and justify the exclusions?	No	Reasons for exlcusion provided, but study details not provided.
8. Did the review authors describe the included studies in adequate detail?	Partial Yes	Table of study characteristics but not further described.
9. Did the review authors use a satisfactory technique for assessing the risk of bias (RoB) in individual studies that were included in the review?	No	RoB of included studies not conducted
10. Did the review authors report on the sources of funding for the studies included in the review?	No	Not reported
11. If meta-analysis was performed, did the review authors use appropriate methods for statistical combination of results?	Yes	random-effects model with parameters estimated using the Markov chain Monte Carlo method of Gibbs sampling
12. If meta-analysis was performed, did the review authors assess the potential impact of RoB in individual studies on the results of the meta-analysis or other evidence synthesis?	No	No RoB reported.
13. Did the review authors account for RoB in individual studies when interpreting/ discussing the results of the review?	Yes	No RoB reported, but authors note the inadequacies and age of the studies in their conclusions.
14. Did the review authors provide a satisfactory explanation for, and discussion of, any heterogeneity observed in the results of the review?	Yes	Fig 3 shows the inconsistency plot used to identify heterogeneity among studies in the closed loop of this network meta-analysis
15. If they performed quantitative synthesis did the review authors carry out an adequate investigation of publication bias (small study bias) and discuss its likely impact on the results of the review?	Yes	Funnel plots provided and discussed.
16. Did the review authors report any potential sources of conflict of interest, including any funding they received for conducting the review?	Yes	The authors have declared that no competing interests exist.
Overall risk of bias of the review	***Moderate***	***More than one non-critical weakness – the systematic review has more than one weakness but no critical flaws. It may provide an accurate summary of the results of the available studies that were included in the review.***

Source: Shea et al. 2017. BMJ 358: j4008 doi:10.1136 (https://doi.org/10.1136/bmj.j4008)

Level II- RCT

Study ID	White 2019		
Domain	**Judgement**	**Description**	**Source**
Random sequence generation (selection bias)	Low risk	Consenting participants were randomised 1:1 in blocks of ten. Presumed to be computer generated, but not reported.	p.262
Allocation concealment (selection bias)	Low risk	Sealed envelopes were used.	p.262
Blinding of participants and personnel (performance bias)	Unclear risk	No blinding used in study. Participants, clinicians, and researchers were aware of treatment allocation. Altough unlikely, this may have affected how participants were treated in followup routine care, including compliance. Twelve women in the single dose group (9%) received only 625 IU anti-D at 28–30 weeks; they were therefore given a second dose at 34–36 weeks, consistent with standard practice, to avoid potential late antenatal sensitisation.	p.262
Blinding of outcome assessment (detection bias)	Low risk	Not blinded but nature of the outcomes (objective measures relating to anti-D levels) makes it unlikely to have created bias in the results	p.262
Incomplete outcome data addressed (attrition bias)	Unclear risk	3/280 (1%) women lost to follow-up (low risk). Antibody screens were available for 254/277 (92%). No imputations/adjustments for missing data were made.	p.263
Selective reporting (reporting bias)	Low risk	Some outcomes missing as per trial registry (see ACTRN12613000661774) (total amount of Rh D IgG used per participant)	
Other sources of bias*	Low risk	Bias could exist in the results due to the high level of participants who did not receive the intervention in the required timeframe (high levels of non-compliance). A sensitivity analysis suggested this did not influence the primary outcome.	p.263
Overall risk of bias of the review	***Unclear risk***	***One domain has some concerns raised, but none are found to be at high risk of bias***	

Source: Chapter 8 of the Cochrane Handbook for Systematic Reviews of Interventions, version 5.1.0

Note: a more recent version of the Cochrane Risk of bias tool is available (see www.riskofbias.info) however, we chose to use the tool specified and used in the 2018 review.

D2 Question 2

Level I-Systematic review of observational studies

Study ID	Schmidt-Hansen 2020	
Question		
1. Did the research questions and inclusion criteria for the review include the components of the PICO?	Yes	PICO elements are outlined (p2)
2. Did the report of the review contain an explicit statement that the review methods were established prior to the conduct of the review and did the report justify any significant deviations from the protocol?	Partial yes	No explicit statement was made about the establishment of prior review methods – except reference to the NICE guidelines. – which links to the full evidence review.
3. Did the review authors explain their selection of the study designs for inclusion in the review?	Partial yes	The study lists that it was able to select RCTs and observational studies but there wasn't an explanation of this. Full details provided in the NICE report.
4. Did the review authors use a comprehensive literature search strategy?	Yes	Yes details of the search strategy (p2)
5. Did the review authors perform study selection in duplicate?	Parital yes	Initial screening was only done by one author will full text screening performed by two
6. Did the review authors perform data extraction in duplicate?	No	Data extraction was to be performed by one author (p2)
7. Did the review authors provide a list of excluded studies and justify the exclusions?	Partial yes	Included in supplementary appendix 2 which was not to be seen in journal links. Found via NICE.
8. Did the review authors describe the included studies in adequate detail?	Partal yes	No studies were included
9. Did the review authors use a satisfactory technique for assessing the risk of bias (RoB) in individual studies that were included in the review?	Yes	Used the GRADE approach to assessing bias
10. Did the review authors report on the sources of funding for the studies included in the review?	Yes	Sources of funding are disclosed (p.5)
11. If meta-analysis was performed, did the review authors use appropriate methods for statistical combination of results?	Partial yes	No meta-analysis was performed but was planned. the The techniques planned were the Mantel-Haenszel statistical method for RRs and the inverse variance statistical method for MDs and SMDs
12. If meta-analysis was performed, did the review authors assess the potential impact of RoB in individual studies on the results of the meta-analysis or other evidence synthesis?	No	No plan for this and no meta-analysis actually performed
13. Did the review authors account for RoB in individual studies when interpreting/ discussing the results of the review?	Partial yes	No individual studies included but potentially would have used the ROB assessment from GRADE to discuss individual study bias in the results
14. Did the review authors provide a satisfactory explanation for, and discussion of, any heterogeneity observed in the results of the review?	No	No heterogeneity discussed
15. If they performed quantitative synthesis did the review authors carry out an adequate investigation of publication bias (small study bias) and discuss its likely impact on the results of the review?	No	No investigation of publication bias (no studies found)
16. Did the review authors report any potential sources of conflict of interest, including any funding they received for conducting the review?	Yes	Authors declared no conflicts of interest and funding details discussed (p5)
Overall risk of bias of the review	***Low***	***No or one non-critical weakness – the systematic review provides an accurate and comprehensive summary of the results of the available studies that address the question of interest.***

Source: Shea et al. 2017. BMJ 358: j4008 doi:10.1136 (https://doi.org/10.1136/bmj.j4008)

D3 Question 3

Level I – Systematic review of RCT, cohrot studies and/or diagnostic accuracy studies

Study ID	**Alshehri 2021**	
Question		
1. Did the research questions and inclusion criteria for the review include the components of the PICO?	Yes	PICO elements are outlined (p19)
2. Did the report of the review contain an explicit statement that the review methods were established prior to the conduct of the review and did the report justify any significant deviations from the protocol?	Yes	This health technology assessment was registered in PROSPERO, the international prospective register of systematic reviews (CRD42019128547), available at https://www.crd.york.ac.uk/PROSPERO.
3. Did the review authors explain their selection of the study designs for inclusion in the review?	Yes	Justification for the included study types is geven (p19)
4. Did the review authors use a comprehensive literature search strategy?	Yes	The comprehensie strategy used is outlined on p18. Multiple data bases were used with appropriate inclusion criteria
5. Did the review authors perform study selection in duplicate?	No	Only one reviewer screened the studies (p19)
6. Did the review authors perform data extraction in duplicate?	Partial yes	It is suggested that data extraction was performed by more than one reviewer but it isn't explicitly said (p20)
7. Did the review authors provide a list of excluded studies and justify the exclusions?	Partial yes	There is a list of excluded studies but the list is not complete. Justification for exclusion of studies is gien
8. Did the review authors describe the included studies in adequate detail?	Yes	The included studies are described in a good level of depth (p26/p33)
9. Did the review authors use a satisfactory technique for assessing the risk of bias (RoB) in individual studies that were included in the review?	Yes	Yes QUADAS-2 was used to assess the bias of included studies
10. Did the review authors report on the sources of funding for the studies included in the review?	No	No explicit statement around sources of funding
11. If meta-analysis was performed, did the review authors use appropriate methods for statistical combination of results?	No	No meta analysis performed
12. If meta-analysis was performed, did the review authors assess the potential impact of RoB in individual studies on the results of the meta-analysis or other evidence synthesis?	No	No meta analysis performed
13. Did the review authors account for RoB in individual studies when interpreting/ discussing the results of the review?	Yes	Appendix 2 demonstrates the authors suffieintly discussing RoB and how this could've affected results
14. Did the review authors provide a satisfactory explanation for, and discussion of, any heterogeneity observed in the results of the review?	Partial yes	Heterogeneity was investigated through the SROC plots for individual SRs. No discussion about heterogeneity in their own results was discussed
15. If they performed quantitative synthesis did the review authors carry out an adequate investigation of publication bias (small study bias) and discuss its likely impact on the results of the review?	Yes	Publication bias was adequatley investigated and the impact of such was considered (p23/p135)
16. Did the review authors report any potential sources of conflict of interest, including any funding they received for conducting the review?	Partial yes	The impact of conflicts of interest in the individual studies was investigated. However the conflicts of interest that could've occurred in the overarching study was not considered
Overall risk of bias of the review	***Moderate***	

Source: Shea et al. 2017. BMJ 358: j4008 doi:10.1136 (https://doi.org/10.1136/bmj.j4008)

Study ID	**Ontario Health 2020**	
Question		
1. Did the research questions and inclusion criteria for the review include the components of the PICO?	Yes	PICO elements are outlined (p19)
2. Did the report of the review contain an explicit statement that the review methods were established prior to the conduct of the review and did the report justify any significant deviations from the protocol?	No	There is no explicit statement about the review methods
3. Did the review authors explain their selection of the study designs for inclusion in the review?	Yes	Justification for the included study types is geven (p19)
4. Did the review authors use a comprehensive literature search strategy?	Yes	The comprehensie strategy used is outlined on p18. Multiple data bases were used with appropriate inclusion criteria
5. Did the review authors perform study selection in duplicate?	No	Only one reviewer screened the studies (p19)
6. Did the review authors perform data extraction in duplicate?	Partial yes	It is suggested that data extraction was performed by more than one reviewer but it isn't explicitly said (p20)
7. Did the review authors provide a list of excluded studies and justify the exclusions?	Partial yes	There is a list of excluded studies but the list is not complete. Justification for exclusion of studies is gien
8. Did the review authors describe the included studies in adequate detail?	Yes	The included studies are described in a good level of depth (p26/p33)
9. Did the review authors use a satisfactory technique for assessing the risk of bias (RoB) in individual studies that were included in the review?	Yes	Yes QUADAS-2 was used to assess the bias of included studies
10. Did the review authors report on the sources of funding for the studies included in the review?	No	No explicit statement around sources of funding
11. If meta-analysis was performed, did the review authors use appropriate methods for statistical combination of results?	No	No meta analysis performed
12. If meta-analysis was performed, did the review authors assess the potential impact of RoB in individual studies on the results of the meta-analysis or other evidence synthesis?	No	No meta analysis performed
13. Did the review authors account for RoB in individual studies when interpreting/ discussing the results of the review?	Yes	Appendix 2 demonstrates the authors suffieintly discussing RoB and how this could've affected results
14. Did the review authors provide a satisfactory explanation for, and discussion of, any heterogeneity observed in the results of the review?	Partial yes	Heterogeneity was investigated through the SROC plots for individual SRs. No discussion about heterogeneity in their own results was discussed
15. If they performed quantitative synthesis did the review authors carry out an adequate investigation of publication bias (small study bias) and discuss its likely impact on the results of the review?	Yes	Publication bias was adequatley investigated and the impact of such was considered (p23/p135)
16. Did the review authors report any potential sources of conflict of interest, including any funding they received for conducting the review?	Partial yes	The impact of conflicts of interest in the individual studies was investigated. However the conflicts of interest that could've occurred in the overarching study was not considered
Overall risk of bias of the review	***Low***	

Source: Shea et al. 2017. BMJ 358: j4008 doi:10.1136 (https://doi.org/10.1136/bmj.j4008)

Study ID	**Runkel 2020**	
Question		
1. Did the research questions and inclusion criteria for the review include the components of the PICO?	Yes	PICO compoenents are outlined (p86)
2. Did the report of the review contain an explicit statement that the review methods were established prior to the conduct of the review and did the report justify any significant deviations from the protocol?	No	No statement that depicts when the review methods were established
3. Did the review authors explain their selection of the study designs for inclusion in the review?	Partial yes	The authors mention study designs that were excluded but have not described the reasoning behind exclusion
4. Did the review authors use a comprehensive literature search strategy?	Yes	Multiple databases were searched with search strategy shown.
5. Did the review authors perform study selection in duplicate?	No	There is no mention as to whether study selection was done in duplication or not
6. Did the review authors perform data extraction in duplicate?	No	The authors did not mention if data extraction was done in duplication or not
7. Did the review authors provide a list of excluded studies and justify the exclusions?	No	No list of excluded studies was given
8. Did the review authors describe the included studies in adequate detail?	Yes	Table one provides all of the relevant study characteristics (p87)
9. Did the review authors use a satisfactory technique for assessing the risk of bias (RoB) in individual studies that were included in the review?	Yes	Used the STROBE checklist to assess bias in individual studies
10. Did the review authors report on the sources of funding for the studies included in the review?	Yes	Authors declared no external sources of funding (p93)
11. If meta-analysis was performed, did the review authors use appropriate methods for statistical combination of results?	Partial yes	The method of meta-analysis is depicted but there isn't sufficient detail given for compatiblility of studies or why they wanted a single pooled effect
12. If meta-analysis was performed, did the review authors assess the potential impact of RoB in individual studies on the results of the meta-analysis or other evidence synthesis?	Partial yes	The authors described the potential bias in the individual studies to be quite low. This led to no discussion around the impact of individual studies on the meta-analysis
13. Did the review authors account for RoB in individual studies when interpreting/ discussing the results of the review?	Partial	Risk of bias in all included studies was deemed to be low. This created a lack of discussion about potential bias in indvividual studies in the results of the review
14. Did the review authors provide a satisfactory explanation for, and discussion of, any heterogeneity observed in the results of the review?	No	No discussion around potential heterogeneity in results
15. If they performed quantitative synthesis did the review authors carry out an adequate investigation of publication bias (small study bias) and discuss its likely impact on the results of the review?	No	No mention of publication bias
16. Did the review authors report any potential sources of conflict of interest, including any funding they received for conducting the review?	Yes	Authors declared no conflicts of interest (p93)
Overall risk of bias of the review	***Low***	

Source: Shea et al. 2017. BMJ 358: j4008 doi:10.1136 (https://doi.org/10.1136/bmj.j4008)

Study ID	**Yang 2019**	
Question		
1. Did the research questions and inclusion criteria for the review include the components of the PICO?	Yes	PICO components listed (p3)
2. Did the report of the review contain an explicit statement that the review methods were established prior to the conduct of the review and did the report justify any significant deviations from the protocol?	No	There was no explicit statement saying that the review methods were established prior to the review
3. Did the review authors explain their selection of the study designs for inclusion in the review?	No	The review authors did not mention the selection of study designs in the inclusion criteria
4. Did the review authors use a comprehensive literature search strategy?	Yes	The search strategy is comprehensive (p2-3, additional file 1)
5. Did the review authors perform study selection in duplicate?	Yes	Two authors involved with a third author used to settle disputes
6. Did the review authors perform data extraction in duplicate?	Partial yes	Data extraction was done by one author, with another author checked by another reviewer. Any disputes between these two authors was resolved by a third author
7. Did the review authors provide a list of excluded studies and justify the exclusions?	No	Reasons for exclusion are listed, but an actual list of excluded studies is not provided
8. Did the review authors describe the included studies in adequate detail?	Yes	PICO elements of included studies provided in table 1 (p5)
9. Did the review authors use a satisfactory technique for assessing the risk of bias (RoB) in individual studies that were included in the review?	Yes	Risk of bias of included studies was checked through the QUADAS-2 checklist
10. Did the review authors report on the sources of funding for the studies included in the review?	Yes	Yes details of funding are listed (p9)
11. If meta-analysis was performed, did the review authors use appropriate methods for statistical combination of results?	Yes	I values listed for combining results to assess heterogeneity. Pooled results from included studies listed (p6)
12. If meta-analysis was performed, did the review authors assess the potential impact of RoB in individual studies on the results of the meta-analysis or other evidence synthesis?	Partial yes	The effect of bias is assessed individually for each study but the effect on the meta analysis is not assessed
13. Did the review authors account for RoB in individual studies when interpreting/ discussing the results of the review?	Yes	The risk of bias is mentioned in the discussion, with certain studies valued more highly due to less bias (p8)
14. Did the review authors provide a satisfactory explanation for, and discussion of, any heterogeneity observed in the results of the review?	Yes	Reasons for the heterogeneity of results is listed (p6)
15. If they performed quantitative synthesis did the review authors carry out an adequate investigation of publication bias (small study bias) and discuss its likely impact on the results of the review?	No	No analysis for small study effects or publication bias was performed because there were too few studies identified to justify (p4)
16. Did the review authors report any potential sources of conflict of interest, including any funding they received for conducting the review?	Yes	Got external funding but declared no conflict of interest (p9)
Overall risk of bias of the review	***Low***	

Source: Shea et al. 2017. BMJ 358: j4008 doi:10.1136 (https://doi.org/10.1136/bmj.j4008)

Level III- Comparative Observational Studies

Study ID	**Jernman 2021**	
Domain	**Judgement**	**Description**
Bias due to failure to develop and apply appropriate eligibility criteria	Low risk	Controls are from the same population as the exposed group – being all pregnant women with anti-D antibodies detected in the Finnish Red Cross (FRC) Blood Service between January 1, 2014 and December 31, 2017. There is a low risk for selection bias, with the intervention and outcome clearly defined.
Bias due to flawed measurement of both exposure and outcome	Low risk	Outcomes are objective (alloimmunisation, HDFN) and comparable across groups, therefore unlikely to be significantly affected by bias. Any potential for flawed measurement of intervention status is carefully accounted for. Any deviations rom intended intervention likley refect usual practise.
Bias due to failure to adequately control confounding	Serious risk	There are numerous potential confouding variables, some of which are not matched between groups (e.g. age, gravidity, parity). Details on BMI and potential sensitising events are not fully captured. There is also risk of potential time varying confouding (including change in obstetric practise) that are not accounted for. The authors conduct both univariate and multivariate analysis to identify potential risk factors associated with sensitisation, but numbers are low and residual confounding is expected.
Bias due to incomplete or inadequately short follow-up	Moderate risk	Follow up was long enough to accurately determine the relevant outcomes (e.g. immunization incidence) but it is possible there is missing data that does not truly reflect the incidence of sensitisation prior to the introduction of targeted RAADP (ie the proportion of missing data between groups slightly differs)
Overall risk of bias	***Serious risk***	***The study appears to provide sound evidence for a non-randomised study but cannot be considered comparable to a well-performed randomised trial. There is potential for some serious residual confounding.***

Source: Table 5.5 GRADE handbook http://gdt.guidelinedevelopment.org/app/handbook/handbook.html#h.m9385o5z3li7

D4 Question 3b

Level II – Consecutive patients with valid reference standard

Study ID	Parchure 2021	
Domain	**Risk of bias**	**Applicability**
Patient selection	Yes (consecutive selection, case-control design was avoided and the study did not inappropriately exclude participants)	Yes (patients match those targetted by the review)
Index test	Yes (reference standard was interpreted without knowledge of the reference standard, prespecified threshold used)	Yes (No difference in the interpretation or variability of test technology)
Reference standard	Yes (Reference standard is likely to correctly classfiy target audience and the result of the reference standard was not known beforehand)	Yes (the target condition as defined by the reference standard matches the index test)
Patient flow	No (All patients received the same reference standard but not all patients were tested at the same point in time)	

Source: QUADAS-2 (Whiting et al., 2011)

Level III-1 – Non-consecutive patients with valid reference standard

Study ID	Legler 2021	
Domain	**Risk of bias**	**Applicability**
Patient selection	No (case-control design, no innappropriate exclusions, consecutive selection)	Yes (patients are applicable to research question)
Index test	Yes (Knowledge of reference standard was known but not likely to have impacted results, prespecified threshold used)	Yes (no real variation in test technology or difference in interpretation)
Reference standard	Unclear (Reference standard is likely to correctly classify target audience but the result of the reference standard was known prior to the index test)	Yes (the target condition as defined by the reference standard matches the index test)
Patient flow	Unclear (All patients received the same reference standard but not all patients tested were the exact same)	

Source: QUADAS-2 (Whiting et al., 2011)

D5 Question 4

Level III- Retrospective Cohort studies

Study ID	Wikman 2021	
Domain	**Judgement**	
Bias due to failure to develop and apply appropriate eligibility criteria	Critical	Both cohorts are from different places and from different times. Large disparity between the numbers of each cohort group
Bias due to flawed measurement of both exposure and outcome	Moderate risk	The measurement of the outcome could have potentially been different in both cohorts but it is unlikely that this introduced significant bias
Bias due to failure to adequately control confounding	Serious risk	Confounders are accounted for in the intervention group (BMI) but not discussed in the comparative group
Bias due to incomplete or inadequately short follow-up	Low risk	Both groups were followed up at the same time
Overall risk of bias	***Critical risk***	***The study is too problematic to provide any useful evidence on the outcome of interest.***

Source: Table 5.5 GRADE handbook http://gdt.guidelinedevelopment.org/app/handbook/handbook.html#h.m9385o5z3li7

Appendix E Data extraction forms

E1 Question 1

Level I – Systematic review (RCTs and cohort studies)

STUDY DETAILS: SR/MA			
Citation			
Xie 2020 Xie, X., Qiurong, F., Bao, Z., Zhang, Y. & Zhou, D. (2020). Clinical value of different anti-D immunoglobulin strategies for preventing Rh hemolytic disease of the fetus and newborn: A network meta-analysis. *PLoS ONE 15*(3). pp. 1-14. https://doi.org/10.1371/journal.pone.0230073			
Affiliation/Source of funds			
Department of Obstetrics and Gynecology, the First People's Hospital of Neijiang, Neijiang, Sichuan Province, P. R. China (X.X. & D.Z.)., Department of Nursing, The first Affiliated Hospital of Hainan Medical University, Haikou, Hainan Province, P. R. China (Q.F.)., Department of medicine, Southwest Medical University, Luzhou, Sichuan Province, P. R. China (Z.B.). & Department of General Surgery, the First People's Hospital of Neijiang, Neijiang, Sichuan Province, P. R. China (Y.Z.)			
Study design	**Level of evidence**	**Location**	**Setting**
SR and MA of Level II and III studies (RCTs and cohoet studies)	Level I-III	USA, Canada, Scotland, Holland, England, France, Denmark, Sweden	Obstetrics and maternal care
Intervention		**Comparator**	
Various dosage amounts of Rh D immunoglobulin *administered antenatal or postpartum*		No treatment; or a placebo; or comparisons of different anti-D regimens.	
Population characteristics			
Rh negative women with Rh positive fetuses, reported positive incidence of anti-D antibody in postpartum mothers			
Length of follow-up		**Outcomes measured**	
Studies published between 1968-2004		Effectiveness of dose and timing of anti-D immunoglobin in preventing maternal antibody sensitisation	
INTERNAL VALIDITY			
Overall risk of bias (descriptive)			
Rating: Moderate Description: Network meta-analysis of low quality studies. The authors do not provide risk of bias assessments or consider the quality of the cohort studies within the analysis. The review may provide an accurate assessment of the available evidence, but the results should be interpreted with caution. . .			
RESULTS:			

Outcome **No. patients (No. trials)** **N = 64860 (24 studies)**	**RAADP** **n/N (%)**	**No therapy** **n/N (%)**	**Risk estimate** **OR (95% CI)**	***Statistical significance*** ***p-value*** **Heterogeneity [a]** **I^2 (*p*-value)**
One dose (250 µg within 28 weeks' gestation) v placebo/no treatment				
Incidence of Rh D alloimmunisation N = 9295 (1 study)	NR	NR	0.05 (0.01, 0.18)	*Favours intervention* *p < 0.05*
SUCRA (surface area under the cumulative ranking curve)	NR	NR	50.3%	Rank = 5
One dose (300 µg within 28 weeks' gestation) v placebo/no treatment				
Incidence of Rh D alloimmunisation N = 16 639 (4 studies)	NR	NR	0.01 (0.00, 0.01)	*Favours intervention* *p < 0.05*

SUCRA (surface area under the cumulative ranking curve)	NR	NR	89.2%	Rank = 2
Two dose (50 µg within 28 and 34 weeks' gestation) v placebo/no treatment				
Incidence of Rh D alloimmunisation N = 1180 (1 study)	NR	NR	0.15 (0.09, 0.24)	*Favours intervention* *p < 0.05*
SUCRA (surface area under the cumulative ranking curve)	NR	NR	17.5%	Rank = 8
Two dose (100 µg between 28 and 34 weeks' gestation) v placebo/no treatment				
Incidence of Rh D alloimmunisation N = 19 684 (4 studies)	NR	NR	0.01 (0.01, 0.03)	*Favours intervention* *p < 0.05*
SUCRA (surface area under the cumulative ranking curve)	NR	NR	75.1%	Rank = 3
Two dose (300 µg between 28 and 34 weeks' gestation) v placebo/no treatment				
Incidence of Rh D alloimmunisation N = 2361 (1 study)	NR	NR	0.00 (0.00, 0.04)	*Favours intervention* *p < 0.05*
SUCRA (surface area under the cumulative ranking curve)	NR	NR	96.8%	Rank = 1
Administered 100 µg ≤ dosage < 200 µg within 72 h postpartum v placebo/no treatment				
Incidence of Rh D alloimmunisation NR (NR)	NR	NR	NR	*NR*
SUCRA (surface area under the cumulative ranking curve)	NR	NR	40.1%	Rank = 6
Administered 200 µg ≤ dosage < 300 µg within 72 h postpartum v placebo/no treatment				
Incidence of Rh D alloimmunisation NR (NR)	NR	NR	0.11 (0.04, 0.31)	*Favours intervention* *p < 0.05*
SUCRA (surface area under the cumulative ranking curve)	NR	NR	24.1%	Rank = 7
Administered 300 µg ≤ dosage < 500 µg within 72 h postpartum v placebo/no treatment				
Incidence of Rh D alloimmunisation NR (NR)	NR	NR	0.04 (0.02, 0.06)	*Favours intervention* *p < 0.05*
SUCRA (surface area under the cumulative ranking curve)	NR	NR	57.0%	Rank = 4
EXTERNAL VALIDITY				
Generalisability (relevance of the study population to the Guidelines target population)				
The evidence is generalisable to the target population with few caveats. All studies were conducted in 'Western' countries.				
Applicability (relevance of the evidence to the Australian health care system)				
The evidence is applicable the Australian health care context with some caveats.				
Additional comments				
Statistical analysis Each closed loop in the network was assessed for inconsistency. Inconsistency factor (IF) was 0.11</= 2.13. 95% CI crossed line of no effect (contained 0, $p > 0.05$). Node analysis showed direct and indirect effect estimates also did not differ. Treatments were ranked using SUCRA analysis of cumulative probability of preventing Rh D alloimmunisation. *Authors conclusions* In conclusion, this study showed that the current first-line recommendation is two 300-µg prenatal immunizations at 28 and 34 gestational weeks. If the anti-D immunoglobulin supply is inadequate, the second alternative should be a single 300-µg prenatal immunization at 28 gestational weeks.				

Included studies
Ascari 1968, Ascari 1969, Bryant 1969, Jennings 1968, Pollack 1968, Robertson 1969, Stenchever 1971, White 1970, Dudok 1968, Clarke 1968, Buchanan 1969, Chown 1969, John 1969, Tovey 1983, Huchet 1987, Bowam 1987, Trolle 1989, Mayne 1997, Mackenzie 1999, Mackenzie 2004, Lee 1995, Bowam 1978, Bowam 1978, Hermann 1984

CI, confidence interval; IU international units; MA, meta-analysis; µg, microgram; RAADP, routine antenatal anti-D prophylaxis; RCT, randomised controlled trial; RR, relative risk; SD, standard deviation; SR, systematic review; UK, United Kingdom

a. Heterogeneity defined as follows: (i) no significant heterogeneity if $P_{het} > 0.1$ and $I^2 < 25\%$; (ii) mild heterogeneity if $I^2 < 25\%$; moderate heterogeneity if I^2 between 25–50%; substantial heterogeneity $I^2 > 50\%$

Level II- RCT

STUDY DETAILS: RCT			
Citation			
White 2019 White, SW., Cheng, JC., Penova-Vaselinovic, B., Wang, C., White, M., Ingleby, B., Arnold, C. & Pennell, CE. (2019). Single dose v two-dose antenatal anti-D prophylaxis: a randomised controlled trial. *Medical Journal of Australia. 221*(6). pp.261-265. Doi:10.5694/mja2.50266			
Affiliation/Source of funds			
Author Affiliations: University of Western Australia, Perth, WA (SWW. & BPV)., King Edward Memorial Hospital for Women, Perth, WA (SWW., BI. & CA)., Royal Perth Hospital, Perth, WA (JCC)., University of Newcastle, Newcastle, NSW (CW., MW. & CEP)., Hunter Medical Research Institute, Newcastle, NSW (CEP). Sources of Funding: The study was funded in part by a grant to Scott White from the Women and Infants Research Foundation (Perth). Conflicts of Interest: Authors declared no conflicts of interest			
Study design	**Level of evidence**	**Location**	**Setting**
RCT	Level II	King Edward Memorial Hospital, WA, Australia	Obstetrics and maternity care
Intervention		**Comparator**	
1500 IU Rh(D) Immunoglobulin-VF at 28 weeks gestation		625 IU Rh(D) Immunoglobulin-VF at 28 and 34 weeks gestation	
Population characteristics			
277 women who attended a tertiary obstetric referral hospital in Perth for antenatal care and were at least 18 years of age, less than 30 weeks pregnant and yet to receive RAADP, Rh(D)-negative (negative antibody screen), and who intended to deliver their baby at the hospital. Exclusion criteria were prior anti-D sensitisation, any contraindication of anti-D administration, and a history of isolated IgA deficiency. Mean age of 30.9 and 31.2 years, 2% to 3% with multiple pregnancy, median BMI of 26.2 and 24.3 and 27% to 31 % had caesarean delivery.			
Length of follow-up		**Outcomes measured**	
Between May 2013 and November 2015.		- Detectability anti-D levels in maternal blood at the time of delivery - Non-compliance with allocated Rh(D) immunoglobulin prophylaxis regimen	
INTERNAL VALIDITY			
Overall risk of bias (descriptive)			
Rating: Unclear Description: One domain, relating ot blinding of the participants and researchers has some concerns raised, but none are found to be at high risk of bias. 9% of women in the single dose group were given a second dose, which may bias the results in favour of the single dose.			
RESULTS			

Population analysed	**Intervention (one dose)**		**Comparator (two dose)**	
Randomised	140		140	
Efficacy analysis (ITT)	125		129	
Efficacy analysis (PP)	65		75	
Safety analysis	138		139	
Outcome	**1500 IU Rh D IgG at 28 weeks n/N (%)**	**625 IU Rh D Ig G at 28 and 34 weeks n/N (%)**	**Risk estimate (95% CI)**	***Statistical significance p-value***
One-dose (1500 IU at 28 weeks) versus two-dose (625 IU at 28 and 34 weeks)				
Proportion with detectable anti-D at delivery (ITT) N = 254	70/125 (56%)	111/129 (86%)	OR 4.91 (2.67, 9.02)	*Favours two-dose* $p < 0.001$
	Univariate analyses: increasing maternal weight [per kg] interval between final dose and birth [per day] gestaton at birth [per day]		 OR 0.84 (0.76, 0.93) OR 0.96 (0.95, 0.98) OR 0.99 (0.99, 1.01)	 $p < 0.001$ $p < 0.001$ $p = 0.20$

	Multivariate analysis (adjusting for maternal weight and interval between final dose and birth)		OR 1.55 (0.62, 3.87)	*No difference* *p* = 0.35
Proportion with detectable anti-D at delivery (PP) N = 140	57/65 (88%)	42/75 (56%)	NNR	*NR*
Non compliant (total)	52/138 (38%)	69/139 (50%)	NR	*No significant difference* *p* = 0.06
Safety	No major adverse events observed. The greater injection volume (> 5 mL) for the single dose group initially made it more painful than for the standard regimen; which was alleviated by using a more concentrated product, delivering the same dose in a smaller volume (2 mL). Twelve women in the single dose group (9%) received only 625 IU anti-D at 28–30 weeks; they were therefore given a second dose at 34–36 weeks, consistent with standard practice, to avoid potential late antenatal sensitisation.			
EXTERNAL VALIDITY				
Generalisability (relevance of the study population to the Guidelines target population)				
The evidence is directly generalisable to the Guideline target population.				
Applicability (relevance of the evidence to the Australian health care system)				
The evidence is directly applicable to the Australian health care system.				
Additional comments				
This is the final published report of the previously included conference abstract (Pennell 2017) that was considered in the 2018 review.				

ANZCTR, Australian New Zealand Clinical Trials Registry; CI, confidence interval; IgG, immunoglobulin; ITT, intent to treat; IU, international units; NR, not reported; OR, odds ratio; PP, per-protocol; RCT, randomised controlled trial; WA, Western Australia

E2 Question 2

Level I – Systematic review of observational studies

STUDY DETAILS: SR/MA			
Citation			
Schmidt-Hansen, 2020 Schmidt-Hansen, M., Lord, J., Hawkins, J., Cameron, S., Pandey, A., Hasler, E. & Regan, F. (2020). Anti-D prophylaxis for rhesus D (RhD)-negative women having an abortion of a pregnancy up to 13^{+6} weeks' gestation: a systematic review and new NICE consensus guidelines. *BMJ Sexual Reporductive Health 0*(0), 1-6, doi:10.1136/bmjsrh-2019-200536			
Affiliation/Source of funds			
National Guideline Alliance, Royal College of Obstetricians & Gynaecologists, London UK (MSH, JH, EH) Department of Obstetrics & Gynaecology, Royal Cornwall Hospitals NHS Trust, Truro, UK (JL) Sexual and Reproductive Health Services, NHS Lothian, Edinburgh, UK (SC) Department of Haematology, Imperial College Healthcare NHS Trust and NHS Blood & Transplant, London, UK (FR) Funding: The study was undertaken by the National Guideline Alliance (NGA) at the Royal College of Obstetricians & Gynaecologists (RCOG), which received funding from the National Institute for Health and Care Excellence (NICE). The authors declared no conflict of interest			
Study design	**Level of evidence**	**Location**	**Setting**
SR and MA of Level II and Level III studies (RCTs and non-randomised trials)	Level I-III	NR	Obstetrics and maternal care
Intervention		**Comparator**	
Intramuscular anti-D prophylaxis (minimum dose of 250 IU/50 µg within 72 hours of medical or surgical abortion)		No anti-D prophylaxis	
Population characteristics			
Women who are RhD (or D) negative and undergoing either medical abortion with mifepristone and misoprostol or surgical abortion using vacuum aspiration of a pregnancy up to 13^{+6} weeks' gestation.			
Length of follow-up		**Outcomes measured**	
Searched Embase, Medline and the Cochrane Library on 19 October 2018. Studies ranged from 1947-2018		- anti-D isoimmunisation/sensitisation or subsequent affected pregnancy.	
INTERNAL VALIDITY			
Overall risk of bias (descriptive)			
Rating: Low Description: No critical weaknesses – the systematic review provides an accurate and comprehensive summary of the results of the available studies that address the question of interest.			
RESULTS:			

Outcome **No. patients (No. trials)**	**RAADP** **n/N (%)**	**No therapy** **n/N (%)**	**Risk estimate** **RR (95% CI)**	***Statistical significance*** ***p-value*** **Heterogeneity [a]** **I^2 (*p*-value)**
No studies found.				

Recommendation 1
Offer anti-D prophylaxis to women who are rhesus D negative who are having an abortion after 10^{+0} weeks' gestation
Recommendation 2
Do not offer anti-D prophylaxis to women who are having a medical abortion up to and including 10^{+0} weeks' gestation
Recommendation 3
Consider anti-D prophylaxis for women who are rhesus D negative and are having a surgical abortion up to and including 10^{+0} weeks' gestation
Recommendation 4

Providers should ensure that: rhesus status testing and anti-D prophylaxis supply does not cause any delays to women having an abortion
Recommendation 5
Providers should ensure that anti-D prophylaxis is availableat the time of the abortion
EXTERNAL VALIDITY
Generalisability (relevance of the study population to the Guidelines target population)
The evidence is generalisable to the target population with some caveats.
Applicability (relevance of the evidence to the Australian health care system)
The evidence is applicable the Australian health care context with some caveats.
Additional comments
The systematic review ended up producing 0 studies that were relevant to the inclusion material. Outcomes were to be analysed as risk ratios in Review Manager 5.3 using the Mantel-Haenszel statistical method and a fixed or random effect model. The overall quality of the evidence was planned to be assessed using GRADE. The results were based off an expert committee that generate the 2019 NICE guidelines on abortion care

CI, confidence interval; IU international units; MA, meta-analysis; µg, microgram; RAADP, routine antenatal anti-D prophylaxis; RCT, randomised controlled trial; RR, relative risk; SD, standard deviation; SR, systematic review; UK, United Kingdom

a. Heterogeneity defined as follows: (i) no significant heterogeneity if $P_{het} > 0.1$ and $I^2 < 25\%$; (ii) mild heterogeneity if $I^2 < 25\%$; moderate heterogeneity if I^2 between 25–50%; substantial heterogeneity $I^2 > 50\%$

E3 Question 3

Level I – Systematic review (of RCTs, cohort studies and/or diagnostic studies)

STUDY DETAILS: Systematic review of diagnostic studies			
Citation			
Alshehri 2021 Alshehri, AA. & Jackson, DE. 2021. Non-Invasive Prenatal Fetal Blood Group Genotype and Its Application in the Management of Hemolytic Disease of Fetus and Newborn: Systematic Review and Meta-Analysis. *Transfusion Medicine Reviews 35*(1). 85-94. https://doi.org/10.1016/j.tmrv.2021.02.001			
Study design	**Level of evidence**	**Location**	**Setting**
Systematic review and meta-analysis of diagnostic studies	Level I	India, France, Netherlands, Great Britain, Denmark, Spain, Sweden, Belgium	Obstetrics and maternity
Index test	**Exon(s) sequenced**	**Internal control(s)**	**Reference standard or comparator**
High-throughput, NIPT cell-free fetal DNA tests of maternal plasma	4, 5, 7, 10 (depends on the study)	Not specified	Serologic cord blood testing
Population characteristics			
Pregnant Rh negative women who could be alloimmunised			
Number of studies		**Outcomes measured**	
16 studies investigating NIPT 11 sudies included in the meta-analysis		Specificity, sensitivity	
Method of analysis			
Meta-analysis of sensitivity and specificity was done through DerSimonian-Liard random effect model			
INTERNAL VALIDITY			
Overall risk of bias (descriptive)			
Rating: Moderate Description: More than one non-critical weakness – the systematic review has more than one weakness but no critical flaws. It may provide an accurate summary of the results of the available studies that were included in the review. Included studies: All studies assessed by the authors to fulfill STROBE quality standards, but details not provided.			

RESULTS							
Outcome	**Sensitivity % (95% CI)**	**Specificity % (95% CI)**	**PPV % (95% CI)**	**NPV % (95% CI)**	**LR+ % (95% CI)**	**LR- % (95% CI)**	**Diagnostic accuracy % (95% CI)**
Diagnostic performance NIPT against birth blood sample (inconclusive as positive)							
N= 31 441 (11 studies)	99.3% (98.7, 99.7)	98.4% (97.4, 99.0)	NR	NR	NR	NR	NR
Rather 2019 (India)	99.2% (99.4, 99.9)	92.3% (60.9, 98.9)	NR	NR	12.88 (NR)	0.0087 (NR)	NR
Darlington 2018	99.7% (98.1, 100)	93.2% (87.7, 96.3)	NR	NR	14.66 (NR)	0.0032 (NR)	NR
Soothill 2015	99.8% (97.5, 100)	99.2% (96.1, 99.8)	NR	NR	124.75 (NR)	0.0020 (NR)	NR
Banch-Clausen 2014	99.5% (99.3, 99.6)	99.8% (99.6, 99.9)	NR	NR	497.5 (NR)	0.0050 (NR)	NR
Chitty 2014	99.3% (99.0, 99.6)	99.1% (98.6, 99.4)	NR	NR	110.33 (NR)	0.0071 (NR)	NR
Grande 2013	99.7% (96.0, 100)	98.4% (92.3, 99.7)	NR	NR	62.31 (NR)	0.0030 (NR)	NR
Wikman 2012	97.6%	98.9%	NRR	NR	88.73 (NR)	0.0242 (NR)	NR

	(96.9, 98.2)	(98.2, 99.3)					
Akolekar 2011	98.1% (95.9, 99.1)	99.7% (95.3, 100)	NR	NR	327 (NR)	0.0191 (NR)	NR
Minon 2008	99.9% (97.8, 100)	99.7% (95.9, 100)	NR	NR	333 (NR)	0.0010 (NR)	NR
Finning 2008	99.7% (99.2, 99.9)	98.0% (96.7, 98.8)	NR	NR	49.85 (NR)	0.0031 (NR)	NR
Finning 2007	98.1% (91.0, 99.6)	99.5% (91.8, 100)	NR	NR	196.2 (NR)	0.0191 (NR)	NR
EXTERNAL VALIDITY							
Generalisability (relevance of the study population to the Guidelines target population)							
The evidence is generalisable to the Australian population with some caveats. Studies enrolled Rh D negative pregnant women but some may not be directly applicable in terms of RHD prevalence.							
Applicability (relevance of the evidence to the Australian health care system)							
The evidence is applicable to the Australian health care context with few caveats. Includes both high throughput studies (automated) and those with manual DNA extraction.							
Additional comments							
Included studies: Rather 2019; Darlington 2018; Soothill 2015; Banch-Clausen 2014; Grande 2013; Wikman 2012; Akolekar 2011; Minon 2008; Finning 2008; Finning 2007							

--. data not reported; cffDNA, cell free fetal DNA; CI, confidence interval; DNA, deoxyribonucleic acid; LR+, positive likelihood ratio; LR-, negative likelihood ratio; NIPT, non-invasive prenatal testing; NPV, negative predictive value; NR, not reported; PPV, positive predictive value; RNA, ribonucleic acid; RT-PCR, real-time polymerase chain reaction.

STUDY DETAILS: Systematic review of diagnostic studies			
Citation			
Ontario Health 2020 Ontario Health. 2020. Noninvasive fetal RhD blood group genotyping: a health technology assessment. *Ontario Health Technology Assessment Series* [Internet]. *20*(15), 1–160. Available from: https://www.hqontario.ca/evidence-to-improve-care/health-technology-assessment/reviews-and-recommendations/noninvasive-fetal-rhd-blood-group-genotyping			
Study design	**Level of evidence**	**Location**	**Setting**
Systematic review and meta-analysis of diagnostic studies	Level I	UK, France, Finland, Cyprus, Netherlands, Denmark, Sweden, Spain, US	Obstetrics and maternity
Index test	**Exon(s) sequenced**	**Internal control(s)**	**Reference standard or comparator**
cffDNA NIPT testing including laboratory-developed tests or commercial test kits	4, 5, 7, 10 (depends on the study)	Not specified	Serologic cord blood testing
Population characteristics			
Pregnant Rh negative women (who could be alloimmunised) with singleton or multiple pregnancies.			
Number of studies		**Outcomes measured**	
6 systematic reviews 11 cohort studies		Diagnostic accuracy, sensitivity, specificity, PPV, NPV, diagnostic accuracy, Unnecessary RhIG avoided; Risk of alloimmunization; Compliance with RhIG prophylaxis; Maternal quality of life; Adverse effects such as infections from or reactions to RhIG; Implementation outcomes such as uptake of testing, uptake of RhIG; Avoidance of cord blood RhD testing	
Method of analysis			
No meta-analysis, the type of analysis differs relevant to the individual study			
INTERNAL VALIDITY			
Overall risk of bias (descriptive)			
Rating: Low Description: No critical weaknesses – the systematic review provides an accurate and comprehensive summary of the results of the available studies that address the question of interest. Risk of bias of included systematic reviews assessed using the Risk of Bias in Systematic Reviews (ROBIS) tool. For nonrandomized studies, the risk of bias of each included study using the Risk of Bias Assessment tool for Non-randomized Studies (RoBINS). Assessments included in GRADE summary of findings			

RESULTS

Outcome	**Sensitivity % (95% CI)**	**Specificity % (95% CI)**	**PPV % (95% CI)**	**NPV % (95% CI)**	**LR+ % (95% CI)**	**LR- % (95% CI)**	**Diagnostic accuracy % (95% CI)**
Diagnostic performance of cffDNA NIPT v blood sample at birth							
Mackie 2017 N = 10 290 tests	99.3% (98.2, 99.7)	98.4% (96.4, 99.3)	NR	NR	61 (22,167)	–0.007 (0.003, 0.186)	NR
Zhu 2014 N = 10 777 tests (excludes 352 inconclusive tests)	98.5% (98.2, 98.7)	97.7% (0.87, 1.83)	98.7 (98.4, 98.9)	98.0 (97.5, 99.0)	42.83	0.015	98.5% (98.2, 98.7)
Geifman-Holtzman 2006 N = 3 078 tests (excludes 183 duplicate samples, studies with N<10, and where	95.4% (90.6, 97.8)	98.6% (96.4, 99.5)	99% (97.9, 99.6)	92.1% (80.9, 97.0)	17.42 (NR)	0.002 (NR)	94.8% (NR)

excluded by primary studies)							
Bivariate meta-analysis	**False-positive rate % (95% CI)**		**False-negative rate % (95% CI)**		**Inconclusive results % (95% CI)**		
Yang 2018 Inconclusive (8 studies) treated as positive exlcuded	 3.86 (2.54–5.82) 1.26 (0.87–1.83)		 0.34 (0.15–0.76) 1.26 (0.87–1.83)				
Universal v targeted anti-D							
Outcome	**Targeted RAADP n/N (%) % (95% CI)**	**No RAADP n/N (%) % (95% CI)**	**Risk estimate OR (95% CI)**				
Incidence of Rh D alloimmunisation (1 study, N=27 926) Tiblad 2013	24/9380 0.26% (0.15, 0.36)	86/18 546 0.46% (0.37, 0.56)	RR 0.55 (0.35, 0.87) Absolute RD: 0.20 NNT 500		The risk of alloimmunization was 45% lower in the genotyping cohort compared with the historic reference cohort that received postnatal and antenatal Rh D immunoglobulin prophylaxis following any potentially sensitising events.		
Utilisation of Rh D immunoglobulin (8 studies)	Pregnancies Carrying RhD negative Fetus (% women who avoid Rh D immunoglobulin)	All Pregnancies, % (n/N)	**Narrative summary (results not pooled)**				
Darlington 2018 (N=850) Haimila 2017 (N=10 814) Papasavva 2016 (N=71) Soothill 2015 (N=529) Clausen 2014 (N=12 668) Tiblad 2013 (N= 27 926) Grande 2013 (N=302) Damkjaer 2012 (N=239)	479/515 (93%) 3626/3641 (99.6%) 18/18 (100%) 17/18 (94%) NR (97.3%) NR (100%) 90/95 (95%) 68/69 (98.6%)	90/335 (27%) 3626/10 814 (33.7%) 18/71 (25.3%) NR (35%) NR (37.1%) 3270/8374 (39%) NR 68/216 (31.5%)	Across studies, 25.3% to 39% of all RhD− pregnancies (with an RhD+ or RhD− fetus) avoided unnecessary RhIG after noninvasive fetal RhD blood group genotyping. Among the RhD− pregnancies carrying an RhD− fetus (i.e., not RhD incompatible nor at risk for alloimmunization), over 90% avoided unnecessary RhIG. Darlington et al reported 93% of not-at-risk RhD− pregnancies avoided unnecessary RhIG in the genotyping arm, compared with only 27% in the control arm (P value or confidence intervals not provided). After noninvasive fetal RhD blood group genotyping in the studies, a small proportion of people (range: 0.4%–10%) received RhIG upon request45 or when test results were inconclusive.				
EXTERNAL VALIDITY							
Generalisability (relevance of the study population to the Guidelines target population)							
The evidence is generalisable to the Australian population with some caveats.							
Applicability (relevance of the evidence to the Australian health care system)							
The evidence is applicable to the Australian health care context.							
Additional comments							
Included studies Diagnostic Accuracy: Mackie 2017; Zhu 2014; Geifman-Holtzman 2006; Yang 2019 Clinical Utility: Darlington 2018; Haimila 2017; Papasavva 2016; Soothill 2015; Clausen 2014; Tiblad 2013; Grande 2013; Damkjaer 2012							

--. data not reported; cffDNA, cell free fetal DNA; CI, confidence interval; DNA, deoxyribonucleic acid; LR+, positive likelihood ratio; LR-, negative likelihood ratio; NIPT, non-invasive prenatal testing; NPV, negative predictive value; NR, not reported; PPV, positive predictive value; RNA, ribonucleic acid; RT-PCR, real-time polymerase chain reaction.

STUDY DETAILS: Systematic review of RCTs and diagnostic studies			
Citation			
Runkel 2020 Runkel, B., Bein, G., Sieben, W., Sow, D., Polus, S. & Fleer, D. 2020. Targeted antenatal anti-D prophylaxis for RhD-negative pregnant women: a systematic review. *BMC Pregnancy and Childbirth 20*(83). 1-10. https://doi.org/10.1186/s12884-020-2742-4			
Affiliation/Source of funds			
Author affiliations: Institute for Quality and Efficiency in Health Care, Cologne Germany, (BR, WS, DS & DF)., Institute for Clinical Immunology and Transfusion Medicine, Justus-Liebig-University, Giessen, Germany, (GB)., Institute for Research in Operative Medicine, Witten/Herdece University, Cologne, Germany (SP).			
Study design	**Level of evidence**	**Location**	**Setting**
Systematic review and meta-analysis of RCTs and diagnostic studies	Level I	France, UK, Netherlands, Denmark, Finland, Sweden, Germany, Spain, Australia, Belgium	Obstetrics and maternity
Index test	**Exon(s) sequenced**	**Internal control(s)**	**Reference standard or comparator**
NIPT testing with subsequent administration of anti-D prophylaxis depending on the result	4, 5, 7, 10 (depends on the study)	Not specified	Universal anti-D prophylaxis for all non-sensitzed rh D-negative women
Population characteristics			
Non-sensitized Rh D negative pregnant women			
Number of studies		**Outcomes measured**	
2 RCTs (Rh D prophylaxis) Identified 70 relevant diagnostic accuracy studies - 58 had small numbers (between 2 and 467), therefore only 12 included in meta-analysis.		Sensitivity, specificity	
Method of analysis			
Meta-analysis was conducted of all the included studies			
INTERNAL VALIDITY			
Overall risk of bias (descriptive)			
Rating: Low Description: No critical weaknesses – the systematic review provides an accurate and comprehensive summary of the results of the available studies that address the question of interest. Included studies: Both off-label studies on anti-D prophylaxis showed a high risk of bias on the study and outcome level. In 11 of the 12 diagnostic accuracy studies, the risk of bias was high in the total score. However, the pooled estimate of all studies were similar to the results of the study with the low risk of bias			
RESULTS			

Outcome **No. patients (No. trials)**	**RAADP** **n/N (%)**	**No therapy** **n/N (%)**	**Risk estimate** **OR (95% CI)**	***Statistical significance*** ***p-value*** **Heterogeneity [a]** **I^2 (*p*-value)**
Incidence of Rh D alloimunisation N = 2297 (2 studies)	NR	NR	Knapp-Hartung method OR 0.33 (0, 123851) Mantel-Haenszel method OR 0.37 (0.13, 1.06) Beta-binomial model OR 0.30 (0.07, 1.26)	*p* = not significant I^2 = 52% (NR) I^2 = 51% (NR)

Outcome	**Sensitivity** **% (95% CI)**	**Specificity** **% (95% CI)**	**PPV** **% (95% CI)**	**NPV** **% (95% CI)**	**LR+** **% (95% CI)**	**LR-** **% (95% CI)**	**Diagnostic accuracy** **% (95% CI)**

Diagnostic performance of cffDNA NIPT v blood sample at birth							
bivariate meta-analysis N = 60 011 (12 studies)	99.9% (99.5; 100)	99.2% (98.5; 99.5)	NR	NR	NR	NR	NR
De Haas 2016 N = 25789	99.9 (99.9, 100)	97.7 (97.4, 98.0)	NR	NR	NR	NR	NR
Clausen 2014 N = 12668	99.9 (99.7, 99.9)	99.1 (98.8, 99.4)	NR	NR	NR	NR	NR
Haimila 2017 N = 10814	100 (99.9, 100)	99.8 (99.6, 99.9)	NR	NR	NR	NR	NR
Wikman 2012 N = 3652	97.6 (96.9, 98.2)	98.9 (98.2, 99.4)	NR	NR	NR	NR	NR
Chitty 2014 N = 2288	99.3 (98.9, 99.6)	99.1 (98.5, 99.4)	NR	NR	NR	NR	NR
Finning 2008 N = 1869	99.7 (99.2, 99.9)	98.0 (96.6, 98.9)	NR	NR	NR	NR	NR
Muller 2008 N = 1022	99.7 (98.9, 100)	99.2 (97.6, 99.8)	NR	NR	NR	NR	NR
Macher 2012 N = 2012	100 (99.4, 100)	98.2 (96.4, 99.3)	NR	NR	NR	NR	NR
Hyland 2017 N = 599	100 (99, 100)	99.6 (97.6, 100)	NR	NR	NR	NR	NR
Akolekar 2011 N = 586	98.2 (96.2, 99.3)	100 (97.8, 100)	NR	NR	NR	NR	NR
Minon 2008 N = 545	100 (99, 100)	100 (98, 100)	NR	NR	NR	NR	NR
Soothill 2015 N = 499	100 (98.6, 100)	99.4 (96.8, 100)	NR	NR	NR	NR	NR
EXTERNAL VALIDITY							
Generalisability (relevance of the study population to the Guidelines target population)							
The evidence is generalisable to the Australian population with some caveats.							
Applicability (relevance of the evidence to the Australian health care system)							
The evidence is applicable to the Australian health care context.							
Additional comments Evidence is to inform the German Institute for Quality and Efficiency in Health Care (IQWiG). The current policy of universal antenatal anti-D administration leads to approximately 50,000 RhD negative pregnant women per year in Germany receiving anti-D prophylaxis even though they are carrying an RhD negative fetus.							
Included studies: Effectiveness: Hutchet 1987; Lee 1995 Diagnostic accuracy: De Haas 2016; Clausen 2014; Haimila 2017; Wikman 2012; Chitty 2014; Finning 2008; Muller 2008; Macher 2012; Hyland 2017; Akolekar 2011; Minon 2008; Soothill 2015							

--. data not reported; cffDNA, cell free fetal DNA; CI, confidence interval; DNA, deoxyribonucleic acid; LR+, positive likelihood ratio; LR-, negative likelihood ratio; NIPT, non-invasive prenatal testing; NPV, negative predictive value; NR, not reported; PPV, positive predictive value; RNA, ribonucleic acid; RT-PCR, real-time polymerase chain reaction.

STUDY DETAILS: Systematic review of diagnostic studies

Citation

Yang, 2019

Yang, H., Llewellyn, A., Walker, R., Harden, M., Saramago, P., Griffin, S. & Simmonds, M. (2019). High-throughput, non-invasive prenatal testing for fetal rhesus D status in RhD negative women: a systematic review and meta-analysis. *BMC Medicine 17*(37). pp. 1-10. https://doi.org/10.1186/s12916-019-1254-4

Study design	**Level of evidence**	**Location**	**Setting**
Systematic review and meta-analysis of diagnostic studies	Level I	London, Denmark, Bristol, Spain, Netherlands, Sweden	Obstetrics and maternity
Index test	**Exon(s) sequenced**	**Internal control(s)**	**Reference standard or comparator**
High-throughput, NIPT cell-free fetal DNA tests of maternal plasma	4, 5, 7, 10 (depends on the study)	Not specified	Serologic cord blood testing

Population characteristics

Pregnant Rh negative women who could be alloimmunised

Number of studies	**Outcomes measured**
Diagnostic accuracy: 8 studies included in the review. Combined sample of 42491.	Diagnostic accuracy, sensitivity, specificity, PPV, NPV, accuracy at gestational age

Method of analysis

Meta-analysis of all eight studies to determine overall false positive and false negative rates.

INTERNAL VALIDITY

Overall risk of bias (descriptive)

Rating : Low

Description: No critical flaws. The systematic review provides an accurate and comprehensive summary of the results of the available studies that address the question of interest.

RESULTS

Outcome	**Sensitivity % (95% CI)**	**Specificity % (95% CI)**	**PPV % (95% CI)**	**NPV % (95% CI)**	**LR+ % (95% CI)**	**LR- % (95% CI)**	**Diagnostic accuracy % (95% CI)**
Diagnostic performance NIPT against birth blood sample (inconclusive as positive)							
N = NR	99.66% (0.15-0.76)	96.14% (2.54-5.82)	NR	NR	25.82 (NR)	0.004 (NR)	NR
Diagnostic performance NIPT against birth blood sample (excluding inconclusive)							
N = NR	99.65% (0.15-0.82)	98.74% (0.87-1.83)	NR	NR	79.09 (NR)	0.004 (NR)	NR
Diagnostic performance NIPT against birth blood sample (only including Bristol population)							
N = NR	99.79% (0.09-0.48)	94.27% (4.58-7.16)	NR	NR	17.42 (NR)	0.002 (NR)	NR

EXTERNAL VALIDITY

Generalisability (relevance of the study population to the Guidelines target population)

The evidence is generalisable to the Australian population with some caveats.

Studies enrolled Rh D negative pregnant women but some may not be directly applicable in terms of *RHD* prevalence

Applicability (relevance of the evidence to the Australian health care system)

The evidence is probably applicable to the Australian health care system with some caveats.

Only high throughput studies were included. This may overestimate the sensitivity of the test.

Additional comments

Duplicate Data (this is published report of data included in our original 2018 search - see Saramago 2018

Included studies

Akolekar 2011; Banch-Clausen, 2014; Chitty 2014; Finning 2008; Grande 2013; Soothill 2015; Thurik 2015; Wikman 2012

--. data not reported; cffDNA, cell free fetal DNA; CI, confidence interval; DNA, deoxyribonucleic acid; LR+, positive likelihood ratio; LR-, negative likelihood ratio; NIPT, non-invasive prenatal testing; NPV, negative predictive value; NR, not reported; PPV, positive predictive value; RNA, ribonucleic acid; RT-PCR, real-time polymerase chain reaction.

Level III- Comparative Observational Studies

STUDY DETAILS: Cohort / Case-control			
Citation			
Jernman 2021 Jernman, R., Isaksson, C., Haimila, K., Kuosmanen, M., Makikallio-Anttila, K., Toivonen, S., Orden, MR., Sulin, K., Tihtonen, K., Vaarasmaki. & Sainio, S. (2021). Time points and risk factors for RhD immunizations after the implementation of targeted routine antenatal anti-D prophylaxis: A retrospective nationwide cohort study. *Acta Obstetricia et Gynecologica Scandinavica. 100*(10). pp. 1868-1875. doi: 10.1111/aogs.14216			
Affiliation/Source of funds			
Funding: Grants were received from the Päivikki and Sakari Sohlberg Foundation and Helsinki University Hospital Obstetrics Department Research Funding The authors declared no conflict of interest Author Affiliations: Department of Obstetrics and Gynecology, University of Helsinki and Helsinki University Hospital, Helsinki, Finland (RJ. & CI)., Finnish Red Cross Blood Service, Helsinki, Finland (KI., MK., ST., KS. & SS)., Department of Obstetrics and Gynecology, Turku University Hospital, Turku, Finland (KM)., Department of Obstetrics and Gynecology, Kuopio University Hospital, Kuopio, Finland (MR)., Department of Obstetrics and Gynecology, Tampere University Hospital, Tampere, Finland (KT)., Department of Obstetrics and Gynecology, Oulu University Hospital, Oulu, Finland (MV).			
Study design	**Level of evidence**	**Location**	**Setting**
Retrospective cohort study	Level III-3	Finland	Obstetrics and maternity
Intervention		**Comparator**	
National screening program of Finland routine antenatal anti-D prophylaxis *Risk-based prophylaxis:* 250-300 mcg anti-D immunoglobin given in the event of a sensitising event (spontaneous abortions after 8 weeks, all terminations of pregnancy, extrauterine pregnancies, chorionic villous sampling, amniocentesis, abdominal trauma, antenatal haemorrhage, external version, intrauterine death *Targeted:* 250-300 mcg anti-D immunoglobin given to RhD-negative mothers with an RHD-positive fetus or if the fetal RhD status is unknown at 28-30 weeks of gestation. *Postnatal:* 250-300 mcg given within 72 hours of delivery to RhD-negative mothers with an RhD-positive newborn or unclear RhD status of the newborn		Pre-introduction of routine anti-D immunoglobin (no routine anti-D screening), general population of pregnant women in Finland during the same period (obtained from the Finnish Institute for Health and Welfare without matching for parity).	
Population characteristics			
RhD negative pregnant women with detected anti-D antibodies who gave birth in Finland (ave age of 27.3), median BMI of 24.5			
Length of follow-up		**Outcomes measured**	
Between 2014-2017		Incidence of anti-D immunization	
Method of analysis			
A nationwide cohort study was conducted of all pregnant women with anti-D antibodies detected in the Finnish Red Cross (FRC) Blood Service between January 1, 2014 and December 31, 2017. The data were analysed using SPSS Version 25.0 (IBM Corporation, Armonk, NY, USA). A p-value <0.05 was considered statistically significant, and 95% CI was used. The number of observations in the study group and controls was compared using Pearson's chi-squared test, Fisher's exact test and Student's t test, depending on the variable. Logistic regression was used to sort out risk factor proportion before and after 2014 and between time-points of immunization			
INTERNAL VALIDITY			
Overall risk of bias (descriptive)			
Rating: Serious Description: The study appears to provide sound evidence for a non-randomised study but cannot be considered comparable to a well-performed randomised trial. There is potential for some serious residual confounding.			
RESULTS			
Population analysed	**Intervention**	**Comparator**	
Available	197	215048	

Outcome	**Targeted RAADP** **n/N** **% (95% CI)**	**No RAADP** **n/N** **% (95% CI)**	**Risk estimate (95% CI)**	***Statistical significance*** ***p-value***
Targeted RAADP vs no RAADP				
Prevalence of anti-D sensitisation among pregnant women (274 pregnancies of 228 women)	54 0.88% (0.68%, 1.14%)	174 1.52% (1.26%, 1.84%)	NR	*Favours intervention* *p* = 0.0009
- Screening at 8-10 weeks - Screening at 24-26 weeks - Screening at 36 weeks	10/54 (18.5%) 27/54 (50%) 17/54 (28%)	NR (52%) NR (20%) NR (28%)		
Incidence of anti-D sensitisation among pregnant women (NR pregnancies of 197 women)	0.10% (0.05%, 0.22%)	0.33% (0.22%, 0.48%)	RR 0.29 (0.10, 0.71) [new sensitisations] Absolute RD 0.20%	*Favours intervention* *p* = 0.0037
	Univariate analysis suggested the following risk factors for sensitisation: PPH ≥ 1000 mL, RBC transfusion in previous pregnancy, twins in ongoing pregnancy. Multivariate analysis: contributions of risk factors did not reach statistical significance. (low numbers may prevent other factors reaching statistical significance)			
EXTERNAL VALIDITY				
Generalisability (relevance of the study population to the Guidelines target population)				
The evidence is generalisable to the Australian population with some caveats.				
Applicability (relevance of the evidence to the Australian health care system)				
The evidence is applicable to the Australian health care context with some caveats.				
Additional comments				
*There were significant baseline differences between the intervention and comparator groups in relation to mean age (27.36 vs 30.7); gravidity (G1: 18.2% vs 29.6%); parity (P0: 25.5% vs 41.3%); and delivery complications (assisted delivery, transfusion, bleeding ≥1000mL, postmaturity ≥41 weeks). *There is insufficient information on the incidence of potential sensitising events. *It is noted that none of the sensitising events were attributed to false-negative fetal *RHD* typing.				

CI, confidence interval; HDFN, haemolytic disease of the fetus and newborn; IgG, immunoglobulin; ITT, intention to treat; IU, international units; IUT, intrauterine transfusion; NNT, number needed to treat; PP, per-protocol; RAADP, routine antenatal anti-D prophylaxis; RCT, randomised controlled trial; RR, relative risk; SD, standard deviation

E4 Question 3b

Level II – Consecutive patients with valid reference standard

STUDY DETAILS: Diagnostic study			
Citation			
Parchure 2021 Parchure, D., Madkaikar. & Kulkarni, S. 2021. Algorithm development and diagnostic accuracy testing for non-invasive foetal RHD genotyping: an Indian experience. *Blood Transfusion.* 1-11. doi: 10.2450/2021.0022-21			
Affiliation/Source of funds			
Author Affliations: Department of Transfusion Medicine, ICMR-National Institute of Immunohaematology, Mumbai, India (DP, SK)., Department of Pediatric Immunology and Leukocyte Biology, ICMR-National Institute of Immunohaematology, Mumbai, India (MM). Funding was sort through an intramural grant received from the Indian Council of Medical Research. The authors declared no sources of conflict			
Study design	**Level of evidence**	**Location and study date**	**Setting**
Prospective observational study	Level II	Mumbai, India	Obstetrics and maternity
Index test	**Exon(s) sequenced**	**Internal control(s)**	**Reference standard or comparator**
PCR method in the extraction of cffDNA from maternal plasma in various weeks of gestation (10-34)	RHD exons 4, 5 and 10 (initial 54 samples) RHD exons 5 and 10 (163 samples)	CCR5, SRY and RASSF1A genes were used as controls	Cord blood serology at delivery
Population characteristics			
RhD negative pregnant Indian women aged between 19-42 with a mean age of 32.5			
Number of studies or samples		**Outcomes measured**	
217		Sensitivity, specificity, diagnostic accuracy, alloimmunisation	
Method of analysis			
Specificity, sensitivity and diagnostic accuracy values of the diagnostic methods were calculated			
INTERNAL VALIDITY			
Overall risk of bias (descriptive)			
RESULTS			
2x2 table with inconclusive results counted as test positive[a]			
N = 217	**Reference standard positive** n = 175 (86.21%)	**Reference standard negative** n = 28 (13.79%)	**Inconclusive results** n = 14
Index text positive n = 175 (86.21%)	175	0	NR
Index text negative n = 28 (13.79%)	0	28	NR
Index test inconclusive			

Outcome	**Sensitivity % (95% CI)**	**Specificity % (95% CI)**	**PPV % (95% CI)**	**NPV % (95% CI)**	**LR+ (95% CI)**	**LR- (95% CI)**	**Diagnostic accuracy % (95% CI)**
Diagnostic performance NIPT against birth blood sample[d]							
	100% (NR)	100%(NR)	NR	NR	NR	NR	100%

EXTERNAL VALIDITY
Generalisability (relevance of the study population to the Guidelines target population)
The evidence is generalisable to the Australian population with some caveats
Applicability (relevance of the evidence to the Australian health care system)
The evidence is not applicable to the Australian health care system

Additional comments

cffDNA, cell free fetal DNA; CI, confidence interval; DNA, deoxyribonucleic acid; GW, gestational week; LR+, positive likelihood ratio; LR-, negative likelihood ratio; NIPT, non-invasive prenatal testing; NPV, negative predictive value; NR, not reported; PPV, positive predictive value; RT-PCR, real-time polymerase chain reaction.

Level III-1 – Non-consecutive patients with a valid reference standard

STUDY DETAILS: Diagnostic study			
Citation			
Legler 2021 Legler, TJ., Luhrig, S., Korschineck, I. & Schwartz, D. (2021). Diagnostic performance of the noninvasive prenatal FetoGnost RhD assay for the prediction of the fetal RhD blood group status. *Archives of Gynecology and Obstetrics (304)*1. pp. 1191-1196. https://doi.org/10.1007/s00404-021-06055-1			
Affiliation/Source of funds			
Open Access funding enabled and organized by Projekt DEAL. Author affiliations: Department of Transfusion Medicine, University Medical Center Göttingen, Robert Koch Str. 40, 37075 Göttingen, Germany (TJL & SL)., Ingenetix GmbH, Vienna, Austria (IK)., Department of Blood Group Serology and Transfusion Medicine, Medical University of Vienna, Vienna, Austria (DS). Sources of conflict: T.L. receives consultation fees from LADR GmbH and participates in the revenue of his employer. I.K. is the owner and manager of the company Ingenetix GmbH. S.L. and D.S. do not have any conflicts of interest/competing interests to declare			
Study design	**Level of evidence**	**Location and study date**	**Setting**
Retrospective observational study	Level III-1	Vienna Medical University Obstetrics department. Between 2009-2020	Obstetrics and maternity
Index test	**Exon(s) sequenced**	**Internal control(s)**	**Reference standard or comparator**
FetoGnost RhD assay	RHD exon 5, exon 7	NR	Cord blood serology
Population characteristics			
Pregnant women aged between 16-50			
Number of studies or samples		**Outcomes measured**	
2968 pregnant women		Sensitivity, specificity, PPV, NPV, diagnostic accuracy	
Method of analysis			
Samples of EDTA blood of RhD negative women were received in the genetics laboratory within a maximum of 6 h of venipuncture. Plasma was separated by centrifugation at 3000 rpm/10 min and stored frozen at - 20C until the insulation. Free-floating DNA from the plasma was isolated from Macherey–Nagel commercial NucleoSpin Plasma kit according to the manufacturer's instructions. In parallel with the isolation of plasma sample in duplicate,and was isolated by the same amount of RNAse free water as a negative control monitored the entire procedure. CffDNA is eluted with 30 ll of the elution buffer. Statistically evaluated by reviewing NIPT-RhD results from the FetoGnost RhD assay with the reference standard of RhD blood group serology results from newborns from the Medical University of Vienna in a retrospective analysis			
INTERNAL VALIDITY			
Overall risk of bias (descriptive)			
Rating: Some concerns Description:			
RESULTS			
2x2 table with inconclusive results counted as test positive[a]			
N = 2968	**Reference standard positive** n = 1475 (63.71%)	**Reference standard negative** n = 769 (33.59%)	**Inconclusive results** n = 644
Index text positive n = 1891 (65.48%)	1474	3	414
Index text negative n = 997 (34.52%)	1	766	230
Index test inconclusive n = 80 (2.70%)	NR	NR	NR

Outcome	**Sensitivity % (95% CI)**	**Specificity % (95% CI)**	**PPV % (95% CI)**	**NPV % (95% CI)**	**LR+ (95% CI)**	**LR- (95% CI)**	**Diagnostic accuracy % (95% CI)**

Diagnostic performance NIPT against birth blood sample[d]							
	99.93% (99.61, 99.99)	99.61% (98.86, 99.87)	99.80 (NR)	99.87 (NR)	256.16 (NR)	0.0007 (NR)	99.82% (99.54, 99.93)
EXTERNAL VALIDITY							
Generalisability (relevance of the study population to the Guidelines target population)							
The evidence is generalisable to the Australian population with some caveats							
Applicability (relevance of the evidence to the Australian health care system)							
The evidence is not applicable to the Australian health care system							
Additional comments							

cffDNA, cell free fetal DNA; CI, confidence interval; DNA, deoxyribonucleic acid; GW, gestational week; LR+, positive likelihood ratio; LR-, negative likelihood ratio; NIPT, non-invasive prenatal testing; NPV, negative predictive value; NR, not reported; PPV, positive predictive value; RT-PCR, real-time polymerase chain reaction.

E5 Question 4

Level III- Retrospective cohort studies

STUDY DETAILS: Case-control			
Citation			
Wikman, 2021 Wikman, A., Mortberg, A., Jalkesten, E., Jansson, Y., Karlsson, A., Tiblad, E. & Ajne, G. 2021. Altered strategy of prophylactic anti-D administration in pregnancy to cover term and post-term – a pilot study. *The international journal of transfusion medicine 116*(1) 1005-1011. https://doi.org/10.1111/vox.13092			
Affiliation/Source of funds			
Author Affiliations: Department of Clinical Immunology and Transfusion Medicine, Karolinska University Hospital, Stockholm, Sweden (AW, AM, EJ & AK)., Division of Immunology, Department of CLINTEC, Karolinska Institute, Stockholm, Sweden (AW & AM)., Pregnancy Care & Delivery, Karolinska University Hospital, Stockholm, Sweden (YJ & GA)., Center for Fetal Medicine, Department of Obstetrics and Gynecology, Karolinska University Hospital, Stockholm, Sweden (ET)., Clinical Epidemiology Division, Department of Medicine Solna, Karolinska Institute, Stockholm, Sweden (ET)., Division of Obstet & Gynecol, Department of CLINTEC, Karolinska Institute, Stockholm, Sweden (GA). Funding: The study was supported by Stockholms Lans Landsting FOU 2018-2019 The authors declared no conflict of interest			
Study design	**Level of evidence**	**Location**	**Setting**
Case-control	Level III	Sweden, Germany	Maternity and obstetrics
Intervention		**Comparator**	
RAADP of 1500 IU of anti-D given at GA 28 and another 1500 IU dose given at GA 38		RAADP with 1250 IU of anti-D given at GA 28-29	
Population characteristics			
RhD negative women with a RhD positive fetus			
Length of follow-up		**Outcomes measured**	
Retrospective cohort was collected between October 2010 and October 2012 in Sweden The prospective cohort was collected between 2016 and 2018 in Germany		- Effect of BMI on anti-D IgG detection in week 38 - Detection of anti-D prophylaxis at delivery	
Method of analysis			
Linear regression analysis was conducted to show the effect of BMI on anti-D detection			
INTERNAL VALIDITY			
Overall risk of bias (descriptive)			
Rating: Moderate Description: The study appears to provide sound evidence for a non-randomised study but cannot be considered comparable to a well-performed randomised trial. Of key concern is an over-representation of women from the primary setting (midwives, GPs) vs obstetric setting (3:1) in the controls compared with cases. Weighted data were used in the analysis.			
RESULTS			

Population analysed	**Cases**		**Controls**	
Available	39		4280	
Analysed	39		4280	
Outcome	**Cases** **n/N (%)**	**Controls** **n/N (%)**	**Risk estimate (95% CI)**	***Statistical significance*** ***p-value***
Linear Regression Analysis				
Detectability of anti-D at delivery	7/39 (18%)	856/4280 (20.5%)	NR	*NR*
Incidence of FMH (>1mL) at delivery	None detected 0/25 (0%)			
BMI	23.9 (18.8, 34.8)	NR	NR	*NR*

median (min, max)	Linear regression analysis showed a significant correlation to body mass index (p = 0.0118)			
EXTERNAL VALIDITY				
Generalisability (relevance of the study population to the Guidelines target population)				
The results are somewhat generalisable to the Australian population				
Applicability (relevance of the evidence to the Australian health care system)				
The results are the study are applicable to the Australian context with some caveats				
Additional comments				

BMI, body mass index; CI, confidence interval; GP, general practitioner; im, intramuscular; IU, international units; OR, odds ratio; RAADP, routine antenatal anti-D prophylaxis; RBC, red blood cell; RCT, randomised controlled trial; SD, standard deviation

a. By design, the controls under primary care were overrepresented (with lower prevalence of potential risk factors for example previous medical intervention), which could overestimate the effect of potential risk factors. The authors therefore weighted the primary care controls (0.35) to restore the proportion of primary care pregnancies to the control group. All p-values are based on n=146.

References

Alshehri, AA and DE Jackson (2021). Non-invasive prenatal fetal blood group genotype and its application in the management of hemolytic disease of fetus and newborn: Systematic review and meta-analysis. *Transfusion Medicine Reviews* **35**(2): 85-94.

Bichler J., Schondorfer G., Pabst G.*, et al.* (2003). Pharmacokinetics of anti-d igg in pregnant rhd-negative women. *BJOG: An International Journal of Obstetrics & Gynaecology* **110**(1): 39.

Bowman, JM, B Chown, M Lewis*, et al.* (1978). Rh isoimmunization during pregnancy: Antenatal prophylaxis. *Can Med Assoc J* **118**(6): 623-627.

Bowman, JM and JM Pollock (1978). Antenatal prophylaxis of rh isoimmunization: 28-weeks'-gestation service program. *Can Med Assoc J* **118**(6): 627-630.

Bowman, JM and JM Pollock (1987). Failures of intravenous rh immune globulin prophylaxis: An analysis of the reasons for such failures. *Transfus Med Rev* **1**(2): 101-112.

Chilcott, J, M Lloyd Jones, J Wight*, et al.* (2003). A review of the clinical effectiveness and cost-effectiveness of routine anti-d prophylaxis for pregnant women who are rhesus-negative. *Health Technol Assess* **7**(4): iii-62.

Gavin, PS (1972). Rhesus sensitization in abortion. *Obstet Gynecol* **39**(1): 37-40.

Geifman-Holtzman, O, CA Grotegut and JP Gaughan (2006). Diagnostic accuracy of noninvasive fetal rh genotyping from maternal blood-a meta-analysis. *American Journal of Obstetrics and Gynecology* **195**(4): 1163-1173.

Haimila, K, K Sulin, M Kuosmanen*, et al.* (2017). Targeted antenatal anti-d prophylaxis program for rhd-negative pregnant women - outcome of the first two years of a national program in finland. *Acta Obstetricia et Gynecologica Scandinavica* **96**(10): 1228-1233.

Hermann, M, H Kjellman and C Ljunggren (1984). Antenatal prophylaxis of rh immunization with 250 micrograms anti-d immunoglobulin. *Acta Obstet Gynecol Scand Suppl* **124**: 1-15.

Huchet, J, S Dallemagne, C Huchet*, et al.* (1987). [ante-partum administration of preventive treatment of rh-d immunization in rhesus-negative women. Parallel evaluation of transplacental passage of fetal blood cells. Results of a multicenter study carried out in the paris region]. *J Gynecol Obstet Biol Reprod (Paris)* **16**(1): 101-111.

Hyland, CA, GM Millard, H O'Brien*, et al.* (2017). Non-invasive fetal rhd genotyping for rhd negative women stratified into rhd gene deletion or variant groups: Comparative accuracy using two blood collection tube types. *Pathology* **49**(7): 757-764.

Jakobsen, MA, HK Rosbach, C Dellgren*, et al.* (2018). Results of noninvasive prenatal rhd testing in gestation week 25 are not affected by maternal body mass index. *Transfusion*.

Jernman, R, C Isaksson, K Haimila*, et al.* (2021). Time points and risk factors for rhd immunizations after the implementation of targeted routine antenatal anti-d prophylaxis: A retrospective nationwide cohort study. *Acta Obstetricia et Gynecologica Scandinavica* **100**(10): 1868-1875.

Karanth, L, SH Jaafar, S Kanagasabai*, et al.* (2013). Anti-d administration after spontaneous miscarriage for preventing rhesus alloimmunisation. *Cochrane Database of Systematic Reviews*(3).

Koelewijn, JM, M de Haas, TG Vrijkotte*, et al.* (2008). One single dose of 200 [mu]g of antenatal rhig halves the risk of anti-d immunization and hemolytic disease of the fetus and newborn in the next pregnancy. *Transfusion* **48**(8): 1721-1729.

Koelewijn, JM, M de Haas, TG Vrijkotte*, et al.* (2009). Risk factors for rhd immunisation despite antenatal and postnatal anti-d prophylaxis. *BJOG: An International Journal of Obstetrics & Gynaecology* **116**(10): 1307-1314.

Lee, D and VI Rawlinson (1995). Multicentre trial of antepartum low-dose anti-d immunoglobulin. *Transfus Med* **5**(1): 15-19.

Legler, TJ, S Luhrig, I Korschineck*, et al.* (2021). Diagnostic performance of the noninvasive prenatal fetognost rhd assay for the prediction of the fetal rhd blood group status. *Archives of Gynecology and Obstetrics* **304**(5): 1191-1196.

Macher, HC, P Noguerol, P Medrano-Campillo*, et al.* (2012). Standardization non-invasive fetal rhd and sry determination into clinical routine using a new multiplex rt-pcr assay for fetal cell-free DNA in pregnant women plasma: Results in clinical benefits and cost saving. *Clinica Chimica Acta* **413**(3-4): 490-494.

MacKenzie, IZ, J Bichler, GC Mason*, et al.* (2004). Efficacy and safety of a new, chromatographically purified rhesus (d) immunoglobulin. *Eur J Obstet Gynecol Reprod Biol* **117**(2): 154-161.

MacKenzie, IZ, P Bowell, H Gregory*, et al.* (1999). Routine antenatal rhesus d immunoglobulin prophylaxis: The results of a prospective 10 year study. *Br J Obstet Gynaecol* **106**(5): 492-497.

Mackie, FL, K Hemming, S Allen*, et al.* (2017). The accuracy of cell-free fetal DNA-based non-invasive prenatal testing in singleton pregnancies: A systematic review and bivariate meta-analysis. *BJOG: An International Journal of Obstetrics and Gynaecology* **124**(1): 32-46.

Manfroi, S, C Calisesi, P Fagiani*, et al.* (2018). Prenatal non-invasive foetal rhd genotyping: Diagnostic accuracy of a test as a guide for appropriate administration of antenatal anti-d immunoprophylaxis. *Blood transfusion = Trasfusione del sangue*(101237479): 1-11.

Mayne, S, JH Parker, TA Harden*, et al.* (1997). Rate of rhd sensitisation before and after implementation of a community based antenatal prophylaxis programme. *BMJ* **315**(7122): 1588.

McBain, RD, CA Crowther and P Middleton (2015). Anti-d administration in pregnancy for preventing rhesus alloimmunisation. *Cochrane Database Syst Rev*(9): CD000020.

Moise, KJ, Jr., M Gandhi, NH Boring*, et al.* (2016). Circulating cell-free DNA to determine the fetal rhd status in all three trimesters of pregnancy. *Obstet Gynecol* **128**(6): 1340-1346.

NCCWCH, Ed. (2012). *Ectopic pregnancy and miscarriage: Diagnosis and initial management in early pregnancy of ectopic pregnancy and miscarriage*. NICE Guidance [NG126]. Royal College of Obstetricians and Gynaecologists, London, National Institute for Health and Clinical Excellence. Accessible at https://www.nice.org.uk/guidance/ng126/evidence/december-2012-full-guideline-pdf-6772587518

NICE, Ed. (2019). *Abortion care*. NICE guideline [NG140], National Institute for Health and Care Excellence. Accessible at https://www.nice.org.uk/guidance/ng140

Ontario Health (2020). Noninvasive fetal rhd blood group genotyping: A health technology assessment. *Ontario Health Technology Assessment Series* **20**(15): 1-160.

Orzińska, A, K Guz, M Dębska*, et al.* (2015). 14 years of polish experience in non-invasive prenatal blood group diagnosis. *Transfusion medicine and hemotherapy : offizielles Organ der Deutschen Gesellschaft fur Transfusionsmedizin und Immunhamatologie* **42**(6): 361-364.

Papasavva, T, P Martin, TJ Legler*, et al.* (2016). Prevalence of rhd status and clinical application of non-invasive prenatal determination of fetal rhd in maternal plasma: A 5 year experience in cyprus. *BMC research notes* **9**((Veldhuisen, van der Schoot) Sanquin Blood Supply, PO Box 9892, 1006 AN, Amsterdam, The Netherlands): 198.

Parchure, D, S Kulkarni and M Madkaikar (2021). Algorithm development and diagnostic accuracy testing for non-invasive foetal rhd genotyping: An indian experience. *Blood transfusion = Trasfusione del sangue*.

Parsons, M, M Van den Hoj, B Armson*, et al.* (1998). A comparison of the rate of rhd alloimmunisation between nova scotia and scotland. . *Br J Obstet Gynaecol* **105** s39.

Pazourkova, E, I Zednikova, M Korabecna*, et al.* (2021). Optimization of diagnostic strategy for non-invasive cell-free foetal rhd determination from maternal plasma. *Vox Sang* **116**(9): 1012-1019.

Picchiassi, E, GC Di Renzo, F Tarquini*, et al.* (2015). Non-invasive prenatal rhd genotyping using cell-free fetal DNA from maternal plasma: An italian experience. *Transfusion medicine and hemotherapy : offizielles Organ der Deutschen Gesellschaft fur Transfusionsmedizin und Immunhamatologie* **42**(1): 22-28.

Pilgrim, H, M Lloyd-Jones and A Rees (2009). Routine antenatal anti-d prophylaxis for rhd-negative women: A systematic review and economic evaluation. *Health Technology Assessment* **13**(37): 1-126.

Runkel, B, W Sieben, D Sow*, et al.* (2020). Targeted antenatal anti-d prophylaxis for rhd-negative pregnant women: A systematic review. *BMC Pregnancy and Childbirth* **20**(1): 83.

Ryan, H, M Lambert, J Mulvany*, et al.* (2017). The identification of maternal rhd variant alleles in rhd-negative pregnant women during the validation of fetal rhd screen in ireland. *Transfus Med* **27**(Supplement 2): 40.

Saramago, P, H Yang, A Llewellyn*, et al.* (2018). High-throughput non-invasive prenatal testing for fetal rhesus d status in rhd-negative women not known to be sensitised to the rhd antigen: A systematic review and economic evaluation. *Health Technology Assessment* **22**(13).

Schmidt-Hansen, M, J Lord, J Hawkins*, et al.* (2020). Anti-d prophylaxis for rhesus d (rhd)-negative women having an abortion of a pregnancy up to 13+6 weeks' gestation: A systematic review and new nice consensus guidelines. *BMJ sexual & reproductive health*.

Simonovits, I, G Bajtai, R Kellner*, et al.* (1974). Immunization of rho(d)-negative secundigravidae whose first pregnancy was terminated by induced abortion. *Haematologia (Budap)* **8**(1-4): 291-298.

Sorensen, K, J Kjeldsen-Kragh, H Husby*, et al.* (2018). Determination of fetal rhd type in plasma of rhd negative pregnant women. *Scand J Clin Lab Inv*((Husby) Department of Obstetrics, Oslo University Hospital, Oslo, Norway): 1-6.

Tovey, LA, A Townley, BJ Stevenson*, et al.* (1983). The yorkshire antenatal anti-d immunoglobulin trial in primigravidae. *Lancet* **2**(8344): 244-246.

Trolle, B (1989). Prenatal rh-immune prophylaxis with 300 micrograms immune globulin anti-d in the 28th week of pregnancy. *Acta Obstet Gynecol Scand* **68**(1): 45-47.

Turner, RM, M Lloyd-Jones, DOC Anumba*, et al.* (2012). Routine antenatal anti-d prophylaxis in women who are rh(d) negative: Meta-analyses adjusted for differences in study design and quality. *PLoS ONE* **7**(2): e30711.

Visscher, RD and HC Visscher (1972). Do rh-negative women with an early spontaneous abortion need rh immune prophylaxis? *Am J Obstet Gynecol* **113**(2): 158-165.

White, SW, JC Cheng, B Penova-Veselinovic*, et al.* (2019). Single dose v two-dose antenatal anti-d prophylaxis: A randomised controlled trial. *Med J Aust* **211**(6): 261-265.

Wikman, A, A Mortberg, E Jalkesten*, et al.* (2021). Altered strategy of prophylactic anti-d administration in pregnancy to cover term and post-term - a pilot study. *Vox Sang* **116**(9): 1005-1011.

Woelfer, B, K Schuchter, M Janisiw*, et al.* (2004). Postdelivery levels of anti-d igg prophylaxis in d-- mothers depend on maternal body weight. *Transfusion* **44**(4): 512-517.

Xie, X, D Zhou, Q Fu*, et al.* (2020). Clinical value of different anti-d immunoglobulin strategies for preventing rh hemolytic disease of the fetus and newborn: A network meta-analysis. *PLoS ONE* **15**(3): e0230073.

Yang, H, A Llewellyn, R Walker*, et al.* (2019). High-throughput, non-invasive prenatal testing for fetal rhesus d status in rhd-negative women: A systematic review and meta-analysis. *BMC Medicine* **17**(1): 37.

Zhu, YJ, YR Zheng, L Li*, et al.* (2014). Diagnostic accuracy of non-invasive fetal rhd genotyping using cell-free fetal DNA: A meta analysis. *Journal of Maternal-Fetal and Neonatal Medicine* **27**(18): 1839-1844.