Hemolytic disease of the fetus and newborn: awareness precedes change

Winter, D.P. de

PART II

EVALUATING OUR CURRENT STANDARDS OF CARE


CHAPTER
2

Hemolytic Disease Of The Fetus And Newborn: Systematic Literature Review Of The Antenatal Landscape

Derek P. de Winter, Allysen Kaminksi, May Lee Tjoa, Dick Oepkes

Published in BMC Pregnancy and Childbirth, 2023 Jan;23:12

ABSTRACT

Background*:* Prevention of pregnancy-related alloimmunization and the management of hemolytic disease of the fetus and newborn (HDFN) has significantly improved over the past decades. Considering improvements in HDFN care, the objectives of this systematic literature review were to assess the prenatal treatment landscape and outcomes of Rh(D)- and K-mediated HDFN in mothers and fetuses, to identify the burden of disease, to identify evidence gaps in the literature, and to provide recommendations for future research.

Methods*:* We performed a systematic search on MEDLINE, EMBASE and clinicaltrials.gov. Observational studies, trials, modelling studies, systematic reviews of cohort studies, and case reports and series of women and/or their fetus with HDFN caused by Rhesus (Rh)D or Kell alloimmunization. Extracted data included prevalence; treatment patterns; clinical outcomes; treatment efficacy; and mortality.

Results*:* We identified 2,541 articles. After excluding 2,482 articles and adding 1 article from screening systematic reviews, 60 articles were selected. Most abstracted data were from case reports and case series. Prevalence was 0.047% and 0.006% for Rh(D)- and K-mediated HDFN, respectively. Most commonly reported antenatal treatment was intrauterine transfusion (IUT; median frequency [interquartile range]: 13.0% [7.2–66.0]). Average gestational age at first IUT ranged between 25 and 27 weeks. weeks. This timing is early and carries risks, which were observed in outcomes associated with IUTs. The rate of hydrops fetalis among pregnancies with Rh(D)-mediated HDFN treated with IUT was 14.8% (range, 0–50%) and 39.2% in K-mediated HDFN. Overall mean±SD fetal mortality rate that was found to be 19.8%±29.4% across 19 studies. Mean gestational age at birth ranged between 34 and 36 weeks.

Conclusion*:* These findings corroborate the rareness of HDFN and frequently needed intrauterine transfusion with inherent risks, and most births occur at a late preterm gestational age. We identified several evidence gaps providing opportunities for future studies.

BACKGROUND

Despite advances in the prevention of pregnancy-related red blood cell immunization and management and treatment of pregnancies affected by hemolytic disease of the fetus and newborn (HDFN) over recent decades, the disease still poses a significant risk in affected pregnancies .[1,2] HDFN is caused by maternal alloimmunization through exposure to incompatible red blood cell antigens of the fetus or through incompatible blood transfusion.[1,3] The then-formed immunoglobulin G (IgG) antibodies are actively transported across the placenta and can cause fetal hemolysis and anemia. When untreated, progressive fetal anemia results in hydrops fetalis and ultimately fetal demise. If the fetus survives, persistent hemolysis causes neonatal anemia and hyperbilirubinemia, which—when untreated—ultimately leads to a severe cerebral condition ("kernicterus").

No cure exists for HDFN. Hence, interventions have focused on its prevention and minimizing adverse effects of associated complications.[1,4] Through transfusing women within the reproductive ages with Kell-negative donor blood, if possible, and through the introduction of Rhesus (Rh) immunoglobulin prophylaxis, the occurrence of red blood cell alloimmunization and the prevalence of Rh(D)- and K-mediated HDFN has decreased[1,4-6]; however, the gap between anti-Rh(D) supply and demand is large in low-income countries and is below the optimal threshold in high-income countries.[7] Additionally, the disease still poses a significant risk for mortality and morbidity in developing countries, whereas it is considered treatable with good outcomes in developed countries. Serological monitoring, ultrasonography, and Doppler imaging decreased the need for risky and invasive diagnostic procedures.[3,8-12] Antenatal treatment, however, still relies predominantly on (often serial) intrauterine transfusion (IUT)—an invasive procedure that carries maternal and fetal risks.[13,14]

Considering improvements in HDFN care, the objectives of this systematic literature review were to assess the prenatal treatment landscape and outcomes of Rh(D)- and K-mediated HDFN in mothers and fetuses to identify the burden of disease, to identify evidence gaps in the literature, and to provide recommendations for future research. Secondarily, we aim to determine the humanistic and economic burden of HDFN.

METHODS

Search strategy

We conducted a systematic literature review according to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) statement[15] and MOOSE Reporting Guidelines for Meta-Analysis of Observational Studies[16] to address prespecified research questions (Supplemental Table 2). To assess the treatment landscape, articles published between January 1, 2005, and March 10, 2021 were searched (Additional file 2: Appendix S1) in the MEDLINE and EMBASE databases and ClinicalTrials.gov using ProQuest (Figure 1). The search strategy included descriptions of the disease, possible interventions and clinical outcomes. No limitations were set on studies reporting on cases managed before January 1, 2005. Searches for clinical outcomes were performed for journal articles and conference abstracts indexed in EMBASE. Duplicates were removed automatically. We also manually searched reference lists of pertinent systematic literature reviews of cohort studies and our personal libraries for potentially relevant articles.

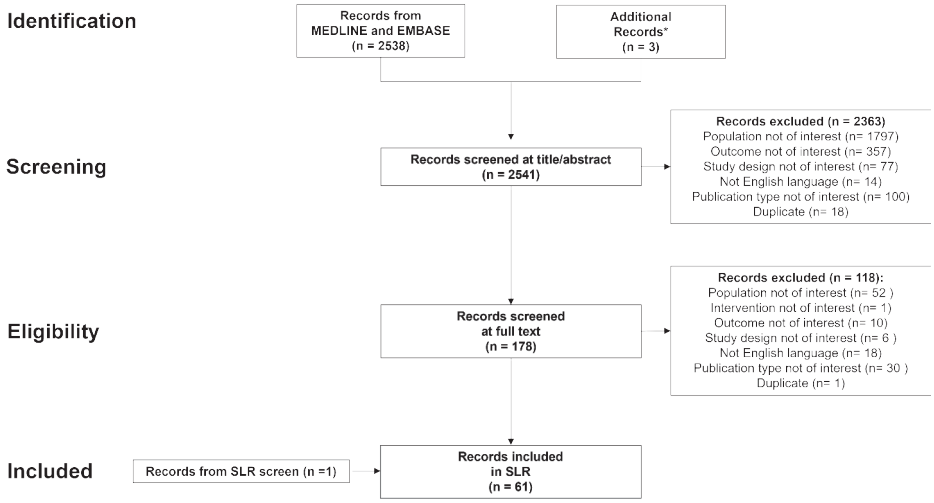


Figure 1: Flowchart of the Article Selection Process. SLR, systematic literature review. *From authors' personal library. †From eligible SLRs of cohort studies.

Study selection

Two independent reviewers (D.P.D.W. and A.K.) (Supplemental Table 3)[17] reviewed the titles/abstracts in Rayyan (https://rayyan.ai/) and then full texts in Microsoft Excel. Citations were independently evaluated to determine whether or not studies fulfilled inclusion and exclusion criteria. The project director (D.O.) and the project

team adjudicated decisions. Randomized or nonrandomized trials; retrospective or prospective observational studies, including cohort, case-control, or cross-sectional studies; modelling studies; systematic reviews of cohort studies (to identify primary studies only); and case reports and case series of women and/or their fetuses, infants, or children experiencing or having experienced Rh(D)- and/or K-mediated HDFN were included. Studies or patient groups within studies where HDFN was caused by alloimmunization to antigens other than Rh(D) only and/or K only, such as c, e, E, Duffy (Fy), Kidd (Jk), MNS (S), or Gerbich, were excluded as the risk of prenatal disease is regarded as relatively low. Non–English-language articles were excluded, as were notes, editorials, and commentaries; nonsystematic reviews; reports of populations, interventions, outcomes, or study designs not of interest; publication types not of interest; indexed conference abstracts; and reports of animal or preclinical studies. The review was registered with PROSPERO before data were abstracted.

Two independent reviewers (D.P.D.W. and A.K.) abstracted data (i.e., study reference; study design; patient characteristics; HDFN treatment patterns; clinical outcomes [eg, fetal anemia, hydrops fetalis, and adverse events]; intravenous immunoglobulin [IVIG] efficacy; mortality; and prevalence) from studies that fulfilled inclusion and exclusion criteria. All abstracted data underwent quality control by the project director (D.O.), who screened 10% of included/excluded articles. The methodological quality (risk of bias) of the selected studies was assessed by 2 independent reviewers (D.P.D.W. and A.K.) using the JBI Critical Appraisal Checklist for Case Reports[18], the JBI Critical Appraisal Checklist for Case Series[19], the Newcastle-Ottawa Scale for retrospective and prospective cohort studies[20], the Checklist for Reporting Results of Internet E-Surveys (CHERRIES) for questionnaires[21], and lastly the NICE checklist for randomized controlled trials (RCTs).

Analyses

Data from eligible studies were characterized as representative, which included data from studies that accurately reflected the characteristics of the larger group (e.g., larger case series, retrospective or prospective studies, RCTs), or were characterized as nonrepresentative, which included data from studies that reflected a small proportion of the characteristics of the larger group (e.g., case reports or small case series, or studies in a subset of the larger group, such as cases treated with IUT or only cases with hydrops fetalis). When possible, we aggregated information reported in a similar manner. For unique outcomes, we highlighted information from generalizable studies. Where appropriate, data were summarized as percentage (mean±standard deviation [SD] or range) or median (interquartile range [IQR]) for patient groups or

patient populations (e.g., Rh[D] or Kell, Rh[D] treated with IVIG or Rh[D] not treated with IVIG). Case reports and case series were excluded from prevalence analyses.

An assessment of the available findings was conducted to identify evidence gaps, and recommendations to fill unmet needs were formulated. Results pertinent to mothers and fetuses are reported herein. Neonatal outcomes will be reported in a separate article.

Humanistic and economic burden

A separate objective of this systematic review is to determine the humanistic and economic burden of HDFN. We conducted a systematic search using the same criteria as previously mentioned (Additional file 2: Appendix S2). We selected studies reporting on quality of life, humanistic burden, economic burden, health care resource use, and direct and indirect costs. The review process was performed according to the PRISMA and MOOSE guidelines and similar as previously stated.

RESULTS

Data sources

In addition to the 2,538 articles identified through searches of MEDLINE and EMBASE, we identified 3 articles from our personal libraries (Figure 1). The search on ClinicalTrials.gov did not yield any additional results to the search on MEDLINE and EMBASE. Overall, 2,363 of the 2,541 total articles were excluded on the basis of title and abstract review, and 119 were excluded on the basis of full-text review. Besides the 59 articles that remained, we identified 1 article from a review of applicable systematic reviews of cohort studies.

Study characteristics

Among the 60 eligible studies that were included in our analysis (Supplemental Table 1)[2,22-80], nearly half were retrospective cohort studies (n=27 [45%]), followed by case reports and case series (n=21 [35%]), prospective cohort studies (n=7 [12%]), observational cohort studies (n=3 [5%]), and RCTs and questionnaires (n=1 [2%] each). More studies included patients with only Rh(D)-mediated HDFN (n=26) than only K-mediated HDFN (n=7); 27 studies included patients with Rh(D)- or K-mediated HDFN. Studies were conducted across 25 countries, most commonly The Netherlands (n=12), followed by Turkey and the United States (n=6 each). The 60 studies comprised 146 patient groups, including mothers, neonates, and fetuses. Of these patient groups,

46 were single patients extracted from case reports. Mean (range) group size, including case reports, was 36.5 (SD±68.7, range 1.0–334.0). The reported patient groups included cases managed between 1985 and 2019.

Methodological quality of the studies

Of the 14 included case reports, 7 received a perfect score (8/8) using the JBI Critical Appraisal Checklist for Case Reports. Median score among case reports was 7.5 [IQR: 6.0–8.0]. Two of the seven included case series received a perfect score among the applicable questions. Median score among case series was 5.0 [IQR: 5.0–8.5]. Twenty-one of 30 retrospective cohort studies were rated as good quality, 8 as fair, and 1 as poor. All 8 included prospective studies were rated as good quality. The randomized controlled trial by Santos et al. was rated as having low risk of bias in all 4 domains—selection bias, performance bias, attrition bias, and detection bias. Supplemental tables 4A-E contains a detailed overview of the methodological quality of the selected studies.

Diagnostic testing

Diagnostic testing data were available for 59 of the 60 studies. The most commonly reported diagnostic testing method was ultrasound (n=20; median [IQR]: 100% [100–100%]), followed by percutaneous umbilical cord sampling (n=17; 100% [100–100%]); anti-D and/or anti-K antibody titer (n=16; 100% [100–100%]); fetal hemoglobin (n=15; 100% [100–100%]); Coombs/antiglobulin testing (n=15; 100%); cell-free DNA testing (n=8; 100% [80–100%]); amniocentesis (n=7; 100% [50–100%]); free antibody testing, antibody release testing, and gel card technique (n=2; 100%); and magnetic resonance imaging (n=2; 100% [56.5–100%]).

Prevalence of D- and K-mediated HDFN

The mean±SD prevalence of Rh(D)-mediated HDFN (requiring any form of treatment) as reported in 5 studies[23,29,45,47,54] was 0.047%±0.037% among all pregnancies that were managed and delivered in the centers of the 5 selected studies (Figure 2). The reported prevalence of K-mediated HDFN (requiring any form of treatment) among all pregnancies managed and delivered in the centers reporting in 2 retrospective studies was 0.006%[45,47]. No data were available for the prevalence of early-onset HDFN (requiring intervention before 24 weeks of pregnancy) in the selected studies. The gestational age at first IUT was between 25 and 27 weeks and the mean gestational age at birth between 34 and 36 weeks (Table 1) amongst all selected studies.[2, 22-80]

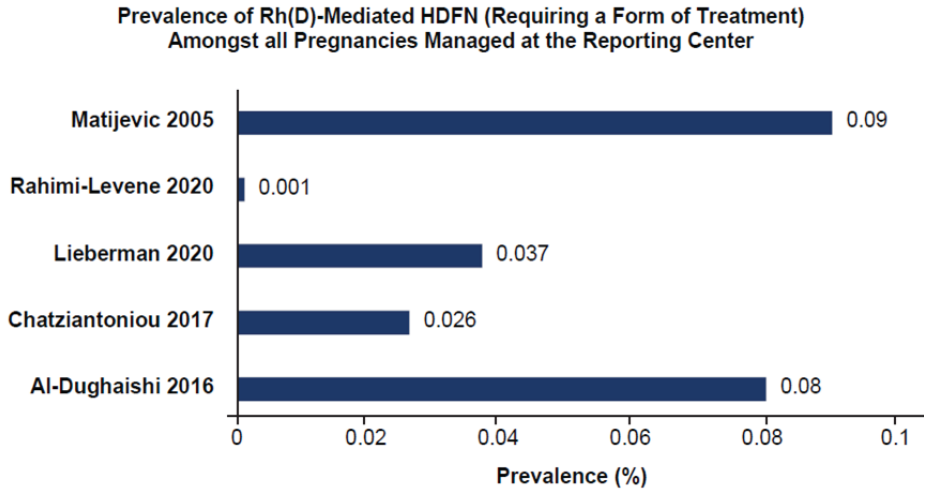


Figure 2: Prevalence of Rh(D)-mediated HDFN (Requiring a Form of Treatment) Among All Referred Pregnancies.[18,24,40,42,49] D, Rh(D); HDFN, hemolytic disease of the fetus and newborn; Rh, Rhesus; SD, standard deviation.

Table 1. Patient Characteristics Among 155 HDFN Groups, Including Mothers, Neonates, and Fetuses, From 60 Included Studies

Characteristic	Mean (±SD, Range)	Patient Groups (n)	No. of Studies (References)
Patient group size, n	36.5 (±68.7, 1.0–334.0)	146[a]	60 (2, 22-80)
Gestational age at birth, weeks[b]			
Mean gestational age	35.1 (±2.1, 33.0–37.4)	13	9 (23, 26, 28, 47, 53, 63, 65, 70, 72)
Median gestational age	35.4 (±2.1, 28.0–37.0)	24	12 (31, 35, 42, 45, 51, 59, 60, 62, 63, 75, 76)
Exact gestational age	34.1 (±2.9, 28.1–38.0)	18	14 (22, 24, 27, 34, 39, 43, 44, 46, 50, 52, 55, 66, 74, 77)
Gestational age at first IUT, weeks			
Mean gestational age	26.6 (±0.1, 26.6–26.7)	4	2 (28, 53)
Median gestational age	25.9 (±3.4, 21.7–36.3)	27	13 (31, 51, 59-61, 63, 64, 69, 73, 75, 79, 80)
Exact gestational age	24.8 (±3.9, 20.0–30.1)	8	5 (44, 52, 66, 68, 76)

HDFN, hemolytic disease of the fetus and newborn; IUT, intrauterine transfusion.
Means, medians, and exact gestational age as reported in each study were used to calculate mean (range).
[a]Of the 155 patient groups, 46 (29.7%) groups were single patients from case reports.
[b]Does not include 1 study (2) in which the reported percentage of the patient group fell within gestational age ranges (ie, <259 days and 259–294 days).

Frequencies of antenatal management strategies

Antenatal treatment data were available for 24 studies (Supplemental Table 8).[24, 25, 29, 30, 33–37, 42, 43, 48, 51, 52, 56, 58–63, 65, 71, 80] The most commonly reported antenatal treatment across these studies was IUT (n=9 with representative data[25, 29, 33, 36, 37, 56, 59, 60, 65]; n=3 with nonrepresentative data).[35, 38, 62]

Intrauterine transfusions

Among the 9 studies with representative data, IUTs were given at a median frequency of 13.0% (IQR: 7.2–66.0) among pregnancies with a positive anti-Rh(D) screening that were monitored prenatally with ultrasonography. Three of these studies monitored pregnancies with a positive anti-Rh(D) screening and a risk-stratification using the antibody titers and/or antibody-dependent cellular cytotoxicity values above cut-off value, and might therefore overestimate the frequency of IUTs.[56, 59, 60] In these three studies, the frequency of IUTs was 64.7% (range 59.2–66).[56, 59, 60] Number of IUTs was reported by only two of these studies with a median of 2 (range 0–4).[56, 59] IUTs were required in 76.8% of pregnancies, as reported in 63/82 collective cases.[36, 56, 60]

The frequency of IUTs in pregnancies with a positive anti-Rh(D) screening monitored without serological cut-off values was 11.2% (range, 4.5–58.6) in the collective cases (42/376) reported by 5 studies.[25, 29, 33, 37, 65] The number of IUTs required was only reported by 1 study with a mean of 2.4 (SD not reported).[33] Data on the need for IUTs in pregnancies with K-alloimmunization without serological cut-offs were reported by 1 study and was 12.5% in 1/8 reported cases.[25]

Alternative management strategies

Use of IVIG alone was reported in 1 study with representative data [48].[48] In this case series of 3 severely affected pregnancies, 2 (Rh[D], n=1; Kell, n=1) resulted in live births without IUT; the third (Rh[D]+anti-C) was treated with IUT but resulted in a post-procedure intrauterine death.

Use of IUT+IVIG was reported in 4 studies (n=1 with representative data[61]; n=3 with nonrepresentative data[35, 42, 62]). In the one study with representative data, 3.2% of the Rh(D) alloimmunization cases and 4.3% of the Kell alloimmunization cases were treated with IUT+IVIG.[61]

Use of other treatments (therapeutic plasma exchange [TPE]; maternal plasma exchange±high-dose IVIG; TPE+IVIG+IUT; TPE+immunoadsorption+IVIG+IUT; plasmapheresis+IUT; and plasmapheresis+IVIG+IUT) were reported in 10 studies with a total of 38 cases.[24, 30, 34, 36, 43, 51, 52, 63, 71, 80] Plasmapheresis+IVIG+IUT was the most

commonly reported treatment regimen across these 10 studies. Two of these studies did not report gestational age at start of the treatment, at first IUT (if applicable) and at birth.[36, 80] The remaining 8 studies, including 20 cases total, reported the mean gestational age at treatment initiation (13.0±5.7 weeks). 17/20 cases required an IUT for fetal anemia. The mean gestational age at first IUT was 24.2±3.1 weeks, with a median of 4 IUTs (range 1–8) administered. Gestational age at birth was 34.4±3.1 weeks.[30, 34, 43, 51, 52, 63, 71] In the series of 20 cases, one patient received plasmapheresis, which was started a week after the first IUT at a gestational age of 27 weeks.[43] In all other cases, the alternative treatment was started prior to the occurrence of fetal anemia. The indications to start the alternative treatment option in the 20 cases were previous intrauterine fetal death (n=11), neonatal hydrops fetalis and/or death (n=4), marked elevation in antibody titer (n=4), and suspected fetal anemia after initial IUT (n=1).

Clinical outcomes of mothers and fetuses

The most commonly reported maternal/fetal clinical outcome across studies was hydrops fetalis (n=19 with representative data[23, 28, 31, 32, 41, 42, 47, 49, 51, 53, 59, 64, 66, 67, 69, 75, 76, 79, 80] (Supplemental Table 8); n=10 with nonrepresentative data[24, 39, 40, 44, 46, 52, 55, 68, 70, 74]. The rate of hydrops fetalis among pregnancies with Rh(D)-mediated HDFN treated with IUT was 14.9% (range, 0–50%) in 72/483 reported cases[31, 32, 49, 53, 66, 76, 79]. The rate of hydrops fetalis among pregnancies with K-mediated HDFN treated with IUT was 39.2% in 49/125 reported cases.[69, 75, 76, 79] Five studies reported on the rate of hydrops fetalis in all pregnancies monitored for Rh(D)- and or K-alloimmunization, with or without the need for antenatal treatment. The rate of hydrops fetalis in these studies was 7.3% in 17/232 collective cases.

Severe fetal anemia was reported in 1 study with representative data[28] (Supplemental Table 8) and 11 studies with nonrepresentative data[30, 39, 42–44, 46, 52, 63, 68, 74, 80]. In the 1 cohort study with representative data, 100% of 22 successful IUTs performed in Rh(D)- or K-mediated HDFN cases within 20 weeks of gestation were considered severely anemic (≥5 SDs from the fetal hemoglobin reference value of 15 g/dL; 1 SD=1 g/dL difference from reference value.[28] Adverse events or procedure-related complications were commonly reported after IUTs or other treatments for Rh(D)- and/or K-mediated HDFN (n=11 studies with representative data[28, 33, 47, 51, 53, 63, 64, 66, 69, 72, 80] (Supplemental Table 8); n=2 studies with nonrepresentative data[68, 73]. Bradycardia was the most frequently reported post-IUT complication per procedure, and adverse serological outcomes were the most frequently reported post-IUT complication per fetus, although adverse serological outcomes were reported in only 1 study[33] (Supplemental Figure 2).

IVIG efficacy

Assessment of IVIG efficacy in women and fetuses affected by HDFN was based on treatment response in 6 studies[24, 30, 34, 48, 62, 80] and associated mortality in 3 studies[35, 42, 63] (Supplemental Table 8). Collectively, findings indicate that IVIG delayed or prevented IUT. IVIG-associated fetal mortality ranged from 0 to 50% across the 3 studies reporting this outcome.[35, 42, 63]

Fetal mortality

The overall mean±SD fetal mortality rate was 19.8% ± 29.4% across 19 studies, including representative case reports[28, 31, 35, 42, 43, 45, 47, 49, 53, 63, 64, 66, 68, 69, 73, 75, 76, 78, 80] (Supplemental Table 8). Head-to-head comparison of mortality rates between the different treatment strategies is limited by variation in potential patient characteristics between the groups. Employed treatment strategies for HDFN take into account previous obstetrical history, for example 75% of cases in the "IUT+other" group had a history of fetal or neonatal death due to HDFN. Together, these will influence the outcomes of HDFN in the current pregnancy and limit our capability of mortality rate comparison.

Humanistic and economic burden

In addition to the 1457 articles identified in the systematic search, one additional article was identified from personal libraries (Supplemental Figure 2). Based on the title/abstract screening 1435 articles were excluded. Full-text screening was performed in the remaining 23 records of which 22 were excluded. Healthcare utilization was reported by only one study with a median of duration of phototherapy of 4-4.5 days and a median length of stay of 6.5–7.5 days.[65]

DISCUSSION

Main findings

We found that the prenatal burden and need for treatment remains relatively high – we estimated that 13% of pregnancies monitored for Rh(D) or K-alloimmunization required one or more IUTs – despite advances in the identification and care for pregnancies at risk of HDFN. Strikingly, the rate of hydrops fetalis in pregnancies requiring an IUT was found to be 14.9% for Rh(D)-mediated HDFN and 39.2% in K-mediated HDFN. As the occurrence of hydrops fetalis was previously found to be associated with impaired neurodevelopmental outcomes[81] still much is to gain in the timely identification of pregnancies at-risk and the timely detection and treatment of fetal anemia to prevent hydrops fetalis. The average

gestational age at first IUT was 27 weeks, which was possibly delayed by using IVIG and/or plasmapheresis although evidence on this in the included studies is limited. Although IUT is a regarded as a relatively safe procedure in experienced hands, its invasive nature still poses serious risks to the mother and fetus. Fetal loss rate increases when procedures need to be done early in gestation (i.e., <22 weeks).[13] It is also noteworthy that the average gestational age at birth in the present analyses was approximately 35 weeks for Rh(D)- and/or K-mediated HDFN, which is considered late preterm and might also represent that early delivery is frequently employed in the management of pregnancies at risk of fetal anemia although we were unable to extract data on this from the included studies. But, late preterm birth has the potential for serious consequences, such as increased risk for short- or long-term respiratory issues[82-84], readmission[82], death[82, 84, 85], and neurocognitive impairment in late adulthood[86, 87].

Strengths and limitations

A strength of this systematic review and corresponding analyses is the minimal limitation on study design criteria. Overall, 35% of the studies included in our analyses were case reports or case series, validating the rarity of HDFN. By including case reports and case series, we were able to identify and aggregate data on treatment types (e.g., plasmapheresis and plasma exchange) that were not typically reported in larger cohort studies. But, inclusion of case reports and case series might also be regarded as a limitation as it may skew and overestimate the results as, given the rarity of HDFN, the most severe cases are generally reported in literature. Our estimates might therefore not truly mirror the population level data.

Also, we were unable to quantify heterogeneity (using e.g. the I2-statistic) due to the descriptive nature of this systematic review and consequent lack of reported comparisons between interventions. However, a certain level of heterogeneity may be expected due to differences in available management options, treatment protocols, prevalence of Rh(D)- and K-negativity, geographical location and sociodemographic differences. These differences may be approached by the varying frequencies of, for instance, IUTs and rate of hydrops fetalis between included studies.

Our analyses are further limited by the strict prespecified inclusion of only Rh(D)- or K-mediated HDFN populations. By applying this criterion, 7 studies were excluded from analyses—despite high quality of evidence and outcomes of interest—because Rh(c) populations were mixed with Rh(D) or Kell populations, or the population was not well defined.[88-94] We were unable to stratify the data per alloimmunization type with the data provided in these articles. One of these studies represented the only prospective

study with long-term outcomes[89], thereby also indicating the lack of data on long-term outcomes and the need for further research on the topic.

Interpretation

Almost all studies included in these analyses were conducted in high-income countries, which have adequate resources for screening, prophylaxis and preventative measures for alloimmunization, and referral to specialized fetal therapy centers. Outcome data on HDFN-complicated pregnancies from less privileged or less organized societies are lacking and, if analyzed, likely are less favorable. This well-known bias in outcome reporting indicates an important evidence gap and signifies the need for international collaboration to gain a better understanding of the global burden of HDFN and to pave the way for potential wide-spread improvements. To add to that, we also found that the evidence for frequency of use and effectiveness of alternative treatment options such as IVIG, plasmapheresis, and plasma exchange on disease severity and the prevention of fetal anemia is limited in the included studies. Also, as previously mentioned it is likely that the most severe cases are reported in literature due to the rarity of the disease. Taken together, future research should aim to gain more exact insight into the employed treatment options and its efficacy and clinical outcomes of mothers, fetuses, and neonates affected by HDFN through an international retrospective and/or prospective registry through the collection of data on diagnostics, antenatal and postnatal treatments and short- and long-term clinical outcomes of mothers, fetuses, and neonates. Such an international effort will pave the way for long sought after answers.

A separate objective of this systematic review, as previously mentioned, was to ascertain the economic and humanistic burden of HDFN. However, through the systematic approach only one study reporting on healthcare utilization was included. This dearth of information indicates another major gap in knowledge, particularly as it relates to the impact of HDFN on a pregnant individual's quality of life and the potential downstream consequences of high-risk pregnancy on family planning decisions, as well as on the healthcare system.

CONCLUSION

To conclude, we found that the clinical burden of Rh(D)- and K-mediated HDFN remains relatively high, with 13% of pregnancies monitored for Rh(D)- or K-alloimmunization requiring an IUT and most births occurring at a late preterm gestational age. We identified several important evidence gaps that provide opportunities for future studies to further improve the clinical care of HDFN.

REFERENCES

1. Jackson ME, Baker JM. Hemolytic Disease of the Fetus and Newborn: Historical and Current State. Clin Lab Med. 2021;41(1):133-51.
2. Raguz MJ, Prce Z, Bjelanovic V, Bjelanovic I, Dzida S, Mabic M. 20 Years of Follow-up Alloimmunization and Hemolytic Disease in Newborn: Has Anything Changed in the Field Over the Years? Klin Padiatr. 2020;232(6):314-20.
3. Dziegiel MH, Krog GR, Hansen AT, Olsen M, Lausen B, Nørgaard LN, et al. Laboratory Monitoring of Mother, Fetus, and Newborn in Hemolytic Disease of Fetus and Newborn. Transfus Med Hemother. 2021;48(5):306-15.
4. Legler TJ. RhIg for the prevention Rh immunization and IVIg for the treatment of affected neonates. Transfus Apher Sci. 2020;59(5):102950.
5. Pollack W, Gorman JG, Freda VJ, Ascari WQ, Allen AE, Baker WJ. Results of clinical trials of RhoGAM in women. Transfusion. 1968;8(3):151-3.
6. Koelewijn JM, de Haas M, Vrijkotte TG, Bonsel GJ, van der Schoot CE. One single dose of 200 microg of antenatal RhIG halves the risk of anti-D immunization and hemolytic disease of the fetus and newborn in the next pregnancy. Transfusion. 2008;48(8):1721-9.
7. Pegoraro V, Urbinati D, Visser GHA, Di Renzo GC, Zipursky A, Stotler BA, et al. Hemolytic disease of the fetus and newborn due to Rh(D) incompatibility: A preventable disease that still produces significant morbidity and mortality in children. PloS one. 2020;15(7):e0235807-e.
8. de Haas M, Thurik FF, Koelewijn JM, van der Schoot CE. Haemolytic disease of the fetus and newborn. Vox Sang. 2015;109(2):99-113.
9. Dukler D, Oepkes D, Seaward G, Windrim R, Ryan G. Noninvasive tests to predict fetal anemia: a study comparing Doppler and ultrasound parameters. Am J Obstet Gynecol. 2003;188(5):1310-4.
10. Oepkes D, Brand R, Vandenbussche FP, Meerman RH, Kanhai HH. The use of ultrasonography and Doppler in the prediction of fetal haemolytic anaemia: a multivariate analysis. Br J Obstet Gynaecol. 1994;101(8):680-4.
11. Oepkes D, Seaward PG, Vandenbussche FP, Windrim R, Kingdom J, Beyene J, et al. Doppler ultrasonography versus amniocentesis to predict fetal anemia. N Engl J Med. 2006;355(2):156-64.
12. Zimmerman R, Carpenter RJ, Jr., Durig P, Mari G. Longitudinal measurement of peak systolic velocity in the fetal middle cerebral artery for monitoring pregnancies complicated by red cell alloimmunisation: a prospective multicentre trial with intention-to-treat. Bjog. 2002;109(7):746-52.
13. Zwiers C, van Kamp I, Oepkes D, Lopriore E. Intrauterine transfusion and non-invasive treatment options for hemolytic disease of the fetus and newborn - review on current management and outcome. Expert Rev Hematol. 2017;10(4):337-44.
14. Snelgrove JW, D'Souza R, Seaward PGR, Windrim R, Kelly EN, Ryan G. Predicting Intrauterine Transfusion Interval and Perinatal Outcomes in Alloimmunized Pregnancies: Time-to-Event Survival Analysis. Fetal Diagn Ther. 2019;46(6):425-32.
15. Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffmann TC, Mulrow CD, et al. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. Bmj. 2021;372:n71.
16. Brooke BS, Schwartz TA, Pawlik TM. MOOSE Reporting Guidelines for Meta-analyses of Observational Studies. JAMA Surgery. 2021;156(8):787-8.
17. Centre for Reviews and Dissemination. Systematic Reviews: CRD's guidance for undertaking reviews in health care. York, UK: University of York; 2009.
18. Moola S, Munn Z, Tufanaru C, Aromartis E, Sears K, Sfetcu R, et al. Chapter 7: Systematic reviews of etiology and risk. In: Aromataris E, editor. JBI Manual for Evidence Synthesis2020.
19. Munn Z, Barker TH, Moola S, Tufanaru C, Stern C, McArthur A, et al. Methodological quality of case series studies: an introduction to the JBI critical appraisal tool. JBI Evidence Synthesis. 2020;18(10).
20. Wells G, Shea B, O'Connell D, Peterson J, Welch V, Losos M, et al. Newcastle-Ottawa quality assessment scale cohort studies. University of Ottawa; 2014.
21. Eysenbach G. Improving the quality of Web surveys: the Checklist for Reporting Results of Internet E-Surveys (CHERRIES). J Med Internet Res. 2004;6(3):e34-e.
22. Akdağ A, Erdeve O, Uraş N, Simşek Y, Dilmen U. Hydrops Fetalis due to Kell Alloimmunization: A Perinatal Approach to a Rare Case. Turk J Haematol. 2012;29(1):72-5.

23. Al-Dughaishi T, Al Harrasi Y, Al-Duhli M, Al-Rubkhi I, Al-Riyami N, Al Riyami A, et al. Red Cell Alloimmunization to Rhesus Antigen Among Pregnant Women Attending a Tertiary Care Hospital in Oman. Oman Med J. 2016;31(1):77-80.

24. Bek SG, Eren N, Uzay A, Bakirdogen S. Rh (D) alloimmunization treated by double filtration plasmapheresis. Transfus Apher Sci. 2019;58(1):83-6.

25. Bennardello F, Curciarello G. Survey on the prevention and incidence of haemolytic disease of the newborn in Italy. Blood Transfus. 2013;11(4):518-27.

26. Bi SH, Jiang LL, Dai LY, Zheng H, Zhang J, Wang LL, et al. Rh-incompatible hemolytic disease of the newborn in Hefei. World J Clin Cases. 2019;7(20):3202-7.

27. Brumbaugh JE, Morgan S, Beck JC, Zantek N, Kearney S, Bendel CM, et al. Blueberry muffin rash, hyperbilirubinemia, and hypoglycemia: A case of hemolytic disease of the fetus and newborn due to anti-Kpa. Journal of Perinatology. 2011;31(5):373-6.

28. Canlorbe G, Macé G, Cortey A, Cynober E, Castaigne V, Larsen M, et al. Management of very early fetal anemia resulting from red-cell alloimmunization before 20 weeks of gestation. Obstet Gynecol. 2011;118(6):1323-9.

29. Chatziantoniou V, Heeney N, Maggs T, Rozette C, Fountain C, Watts T, et al. A descriptive single-centre experience of the management and outcome of maternal alloantibodies in pregnancy. Transfus Med. 2017;27(4):275-85.

30. Colpo A, Tison T, Gervasi MT, Vio C, Vicarioto M, De Silvestro G, et al. Personalized treatment with immunoadsorption and intravenous immunoglobulin in a case of severe Rh alloimmunization during pregnancy unresponsive to plasma - exchange. Transfus Apher Sci. 2017;56(3):480-3.

31. Craparo FJ, Bonati F, Gementi P, Nicolini U. The effects of serial intravascular transfusions in ascitic/hydropic RhD-alloimmunized fetuses. Ultrasound Obstet Gynecol. 2005;25(2):144-8.

32. de Assunção RA, Liao AW, Brizot Mde L, Francisco RP, Zugaib M. Changes in fetal myocardial performance index following intravascular transfusion: preliminary report. J Matern Fetal Neonatal Med. 2016;29(16):2697-702.

33. Dubey A, Sonker A, Chaudhary R. Enhancement of Antibody Titre and Development of Additional Red Cell Alloantibodies Following Intrauterine Transfusion. Indian J Hematol Blood Transfus. 2016;32(1):92-4.

34. Fernández Alba JJ, León R, González-Macías C, Paz A, Prado F, Moreno LJ, et al. Treatment of D alloimmunization in pregnancy with plasmapheresis and intravenous immune globulin: case report. Transfus Apher Sci. 2014;51(1):70-2.

35. Fox C, Martin W, Somerset DA, Thompson PJ, Kilby MD. Early intraperitoneal transfusion and adjuvant maternal immunoglobulin therapy in the treatment of severe red cell alloimmunization prior to fetal intravascular transfusion. Fetal Diagn Ther. 2008;23(2):159-63.

36. Gottvall T, Filbey D. Alloimmunization in pregnancy during the years 1992-2005 in the central west region of Sweden. Acta Obstet Gynecol Scand. 2008;87(8):843-8.

37. Gudlaugsson B, Hjartardottir H, Svansdottir G, Gudmundsdottir G, Kjartansson S, Jonsson T, et al. Rhesus D alloimmunization in pregnancy from 1996 to 2015 in Iceland: a nation-wide population study prior to routine antenatal anti-D prophylaxis. Transfusion. 2020;60(1):175-83.

38. Haider M, Memon S, Tariq F, Fatima S, Hameed A. Rhesus Isoimmunization: Late-onset Hemolytic Disease of the Newborn Without Jaundice. Cureus. 2020;12(1):e6559.

39. Harper DC, Swingle HM, Weiner CP, Bonthius DJ, Aylward GP, Widness JA. Long-term neurodevelopmental outcome and brain volume after treatment for hydrops fetalis by in utero intravascular transfusion. Am J Obstet Gynecol. 2006;195(1):192-200.

40. Hassan MZ, Iberahim S, Abdul Rahman WSW, Zulkafli Z, Bahar R, Ramli M, et al. Severe anti-D haemolytic disease of fetal and newborn in rhesus D negative primigravida. Malays J Pathol. 2019;41(1):55-8.

41. Karagol BS, Zenciroglu A, Okumus N, Karadag N, Dursun A, Hakan N. Hemolytic disease of the newborn caused by irregular blood subgroup (Kell, C, c, E, and e) incompatibilities: report of 106 cases at a tertiary-care centre. Am J Perinatol. 2012;29(6):449-54.

42. Kriplani A, Malhotra Singh B, Mandal K. Fetal intravenous immunoglobulin therapy in rhesus hemolytic disease. Gynecol Obstet Invest. 2007;63(3):176-80.

43. Lakhwani S, Machado P, Pecos P, Coloma M, Rebollo S, Raya JM. Kell hemolytic disease of the fetus. Combination treatment with plasmapheresis and intrauterine blood transfusion. Transfus Apher Sci. 2011;45(1):9-11.

44. Levy-Zauberman Y, Mailloux A, Kane A, Castaigne V, Cortey A, Carbonne B. Massive fetomaternal hemorrhage secondary to intrauterine intravascular transfusion. Obstet Gynecol. 2011;118(2 Pt 2):439-42.

45. Lieberman L, Callum J, Cohen R, Cserti-Gazdewich C, Ladhani NNN, Buckstein J, et al. Impact of red blood cell alloimmunization on fetal and neonatal outcomes: A single center cohort study. Transfusion. 2020;60(11):2537-46.

46. Manoura A, Korakaki E, Hatzidaki E, Saitakis E, Maraka S, Papamastoraki I, et al. Use of recombinant erythropoietin for the management of severe hemolytic disease of the newborn of a K0 phenotype mother. Pediatr Hematol Oncol. 2007;24(1):69-73.

47. Matijevic R, Grgic O, Klobucar A, Miskovic B. Diagnosis and Management of Rh Alloimmunization. Fetal Diagnosis and Therapy. 2005;20(5):393-401.

48. Mayer B, Hinkson L, Hillebrand W, Henrich W, Salama A. Efficacy of Antenatal Intravenous Immunoglobulin Treatment in Pregnancies at High Risk due to Alloimmunization to Red Blood Cells. Transfusion Medicine and Hemotherapy. 2018;45(6):429-36.

49. Meraj B, Mobusher I, Waheed S, Waseem M, Rashid Y. Role of intrauterine blood transfusions in management of Rh-isoimmunized pregnancies. Pakistan J Medical Health Sci. 2015;9:318-21.

50. Navarro M, Negre S, Matoses ML, Golombek SG, Vento M. Necrotizing enterocolitis following the use of intravenous immunoglobulin for haemolytic disease of the newborn. Acta Paediatr. 2009;98(7):1214-7.

51. Nwogu LC, Moise KJ, Jr., Klein KL, Tint H, Castillo B, Bai Y. Successful management of severe red blood cell alloimmunization in pregnancy with a combination of therapeutic plasma exchange, intravenous immune globulin, and intrauterine transfusion. Transfusion. 2018;58(3):677-84.

52. Palfi M, Hildén JO, Matthiesen L, Selbing A, Berlin G. A case of severe Rh (D) alloimmunization treated by intensive plasma exchange and high-dose intravenous immunoglobulin. Transfus Apher Sci. 2006;35(2):131-6.

53. Phung TV, Houfflin-Debarge V, Ramdane N, Ghesquière L, Delsalle A, Coulon C, et al. Maternal red blood cell alloimmunization requiring intrauterine transfusion: a comparative study on management and outcome depending on the type of antibody. Transfusion. 2018;58(5):1199-205.

54. Rahimi-Levene N, Chezar J, Yahalom V. Red blood cell alloimmunization prevalence and hemolytic disease of the fetus and newborn in Israel: A retrospective study. Transfusion. 2020;60(11):2684-90.

55. Rahimi-Sharbaf F, Niromanesh S, Talebzadeh Z, Kaveh M, Nayary F. Rh alloimmunization and term delivery. Arch Iran Med. 2007;10(1):111-3.

56. Rath ME, Smits-Wintjens VE, Lindenburg IT, Brand A, van Kamp IL, Oepkes D, et al. Exchange transfusions and top-up transfusions in neonates with Kell haemolytic disease compared to Rh D haemolytic disease. Vox Sang. 2011;100(3):312-6.

57. Rath ME, Smits-Wintjens VE, Oepkes D, van Zwet EW, van Kamp IL, Brand A, et al. Thrombocytopenia at birth in neonates with red cell alloimmune haemolytic disease. Vox Sang. 2012;102(3):228-33.

58. Rath ME, Smits-Wintjens VE, Oepkes D, Walther FJ, Lopriore E. Iron status in infants with alloimmune haemolytic disease in the first three months of life. Vox Sang. 2013;105(4):328-33.

59. Rath ME, Smits-Wintjens VE, Lindenburg IT, Folman CC, Brand A, van Kamp IL, et al. Postnatal outcome in neonates with severe Rhesus c compared to rhesus D hemolytic disease. Transfusion. 2013;53(7):1580-5.

60. Ree IMC, de Haas M, Middelburg RA, Zwiers C, Oepkes D, van der Bom JG, et al. Predicting anaemia and transfusion dependency in severe alloimmune haemolytic disease of the fetus and newborn in the first 3 months after birth. Br J Haematol. 2019;186(4):565-73.

61. Ree IMC, Lopriore E, Zwiers C, Böhringer S, Janssen MWM, Oepkes D, et al. Suppression of compensatory erythropoiesis in hemolytic disease of the fetus and newborn due to intrauterine transfusions. Am J Obstet Gynecol. 2020;223(1):119.e1-.e10.

62. Ree IMC, de Grauw AM, Bekker V, de Haas M, Te Pas AB, Oepkes D, et al. Necrotizing enterocolitis in haemolytic disease of the newborn: a retrospective cohort study. Vox sanguinis. 2020;115(2):196-201.

63. Ruma MS, Moise KJ, Jr., Kim E, Murtha AP, Prutsman WJ, Hassan SS, et al. Combined plasmapheresis and intravenous immune globulin for the treatment of severe maternal red cell alloimmunization. Am J Obstet Gynecol. 2007;196(2):138.e1-6.

64. Sainio S, Nupponen I, Kuosmanen M, Aitokallio-Tallberg A, Ekholm E, Halmesmäki E, et al. Diagnosis and treatment of severe hemolytic disease of the fetus and newborn: a 10-year nationwide retrospective study. Acta Obstet Gynecol Scand. 2015;94(4):383-90.

65. Santos MC, Sá C, Gomes Jr SC, Camacho LA, Moreira ME. The efficacy of the use of intravenous human immunoglobulin in Brazilian newborns with rhesus hemolytic disease: a randomized double-blind trial. Transfusion. 2013;53(4):777-82.

66. Şavkli AÖ, Çetin BA, Acar Z, Özköse Z, Behram M, Çaypinar SS, et al. Perinatal outcomes of intrauterine transfusion for foetal anaemia due to red blood cell alloimmunisation. Journal of Obstetrics and Gynaecology. 2020;40(5):649-53.

67. Sikkel E, Klumper FJ, Oepkes D, Teunissen AK, Meerman RH, Le Cessie S, et al. Fetal cardiac contractility before and after intrauterine transfusion. Ultrasound Obstet Gynecol. 2005;26(6):611-7.

68. Simonazzi G, Bernabini D, Curti A, Bisulli M, Pilu G, Brill CB, et al. Fetal cerebellar damage in fetuses with severe anemia undergoing intrauterine transfusions. J Matern Fetal Neonatal Med. 2016;29(3):389-92.

69. Somerset DA, Moore A, Whittle MJ, Martin W, Kilby MD. An audit of outcome in intravascular transfusions using the intrahepatic portion of the fetal umbilical vein compared to cordocentesis. Fetal Diagn Ther. 2006;21(3):272-6.

70. Takcı S, Alarcon-Martinez T, Bozkaya D, Yiğit Ş, Korkmaz A, Yurdakök M. Cholestasis in infants with immune hydrops fetalis. Turk J Pediatr. 2013;55(6):616-9.

71. Tara F, Maleki A, Taheri N, Moein Darbari S. A case of D alloimmunization in pregnancy: successfully treated solely with therapeutic plasma exchange (TPE). J Blood Med. 2019;10:251-3.

72. Temel Yüksel İ, Acar D, Turhan U, Aslan Çetİn B, Köroğlu N, Şenol G, et al. Assessment of fetal right ventricular myocardial performance index changes following intrauterine transfusion. The Journal of Maternal-Fetal & Neonatal Medicine. 2021;34(18):3046-9.

73. Tiblad E, Kublickas M, Ajne G, Bui TH, Ek S, Karlsson A, et al. Procedure-related complications and perinatal outcome after intrauterine transfusions in red cell alloimmunization in Stockholm. Fetal Diagn Ther. 2011;30(4):266-73.

74. Urutherakumar V, Welsh A, Henry A. Short-term outcomes following intrauterine transfusions for fetal anaemia: A retrospective cohort study. Australian and New Zealand Journal of Obstetrics and Gynaecology. 2020;60(5):738-45.

75. van den Akker ES, Klumper FJ, Brand A, Kanhai HH, Oepkes D. Kell alloimmunization in pregnancy: associated with fetal thrombocytopenia? Vox Sang. 2008;95(1):66-9.

76. Walsh CA, Russell N, McAuliffe FM, Higgins S, Mahony R, Carroll S, et al. Relationship between maternal antibody type and antenatal course following intrauterine transfusion for red cell alloimmunisation. Eur J Obstet Gynecol Reprod Biol. 2013;171(2):235-9.

77. Xu W. A case of severe Rh (D) alloimmunization pregnant woman delivery an infant with limited treatment. Transfus Apher Sci. 2013;49(2):168-70.

78. Zwiers C, Lindenburg ITM, Klumper FJ, de Haas M, Oepkes D, Van Kamp IL. Complications of intrauterine intravascular blood transfusion: lessons learned after 1678 procedures. Ultrasound Obstet Gynecol. 2017;50(2):180-6.

79. Zwiers C, Oepkes D, Lopriore E, Klumper FJ, de Haas M, van Kamp IL. The near disappearance of fetal hydrops in relation to current state-of-the-art management of red cell alloimmunization. Prenat Diagn. 2018;38(12):943-50.

80. Zwiers C, van der Bom JG, van Kamp IL, van Geloven N, Lopriore E, Smoleniec J, et al. Postponing Early intrauterine Transfusion with Intravenous immunoglobulin Treatment; the PETIT study on severe hemolytic disease of the fetus and newborn. Am J Obstet Gynecol. 2018;219(3):291.e1-.e9.

81. Lindenburg IT, Smits-Wintjens VE, van Klink JM, Verduin E, van Kamp IL, Walther FJ, et al. Long-term neurodevelopmental outcome after intrauterine transfusion for hemolytic disease of the fetus/newborn: the LOTUS study. Am J Obstet Gynecol. 2012;206(2):141.e1-8.

82. Escobar GJ, Clark RH, Greene JD. Short-term outcomes of infants born at 35 and 36 weeks gestation: we need to ask more questions. Semin Perinatol. 2006;30(1):28-33.

83. Natile M, Ventura ML, Colombo M, Bernasconi D, Locatelli A, Plevani C, et al. Short-term respiratory outcomes in late preterm infants. Italian Journal of Pediatrics. 2014;40(1):52.

84. Kitsommart R, Janes M, Mahajan V, Rahman A, Seidlitz W, Wilson J, et al. Outcomes of late-preterm infants: a retrospective, single-center, Canadian study. Clin Pediatr (Phila). 2009;48(8):844-50.

85. Santos IS, Matijasevich A, Silveira MF, Sclowitz IK, Barros AJ, Victora CG, et al. Associated factors and consequences of late preterm births: results from the 2004 Pelotas birth cohort. Paediatr Perinat Epidemiol. 2008;22(4):350-9.

86. Heinonen K, Eriksson JG, Lahti J, Kajantie E, Pesonen AK, Tuovinen S, et al. Late preterm birth and neurocognitive performance in late adulthood: a birth cohort study. Pediatrics. 2015;135(4):e818-25.

87. Martínez-Nadal S, Bosch L. Cognitive and Learning Outcomes in Late Preterm Infants at School Age: A Systematic Review. Int J Environ Res Public Health. 2020;18(1).

88. Deka D, Dadhwal V, Sharma AK, Shende U, Agarwal S, Agarwal R, et al. Perinatal survival and procedure-related complications after intrauterine transfusion for red cell alloimmunization. Arch Gynecol Obstet. 2016;293(5):967-73.

89. van Klink JM, van Veen SJ, Smits-Wintjens VE, Lindenburg IT, Rijken M, Oepkes D, et al. Immunoglobulins in Neonates with Rhesus Hemolytic Disease of the Fetus and Newborn: Long-Term Outcome in a Randomized Trial. Fetal Diagn Ther. 2016;39(3):209-13.

90. van den Akker ES, de Haan TR, Lopriore E, Brand A, Kanhai HH, Oepkes D. Severe fetal thrombocytopenia in Rhesus D alloimmunized pregnancies. Am J Obstet Gynecol. 2008;199(4):387.e1-4.

91. Smits-Wintjens VE, Rath ME, van Zwet EW, Oepkes D, Brand A, Walther FJ, et al. Neonatal morbidity after exchange transfusion for red cell alloimmune hemolytic disease. Neonatology. 2013;103(2):141-7.

92. Smits-Wintjens VE, Walther FJ, Rath ME, Lindenburg IT, te Pas AB, Kramer CM, et al. Intravenous immunoglobulin in neonates with rhesus hemolytic disease: a randomized controlled trial. Pediatrics. 2011;127(4):680-6.

93. Ree IMC, Besuden CFJ, Wintjens V, Verweij J, Oepkes D, de Haas M, et al. Exchange transfusions in severe Rh-mediated alloimmune haemolytic disease of the foetus and newborn: a 20-year overview on the incidence, associated risks and outcome. Vox Sang. 2021;116(9):990-7.

94. Koelewijn JM, Vrijkotte TG, van der Schoot CE, Bonsel GJ, de Haas M. Effect of screening for red cell antibodies, other than anti-D, to detect hemolytic disease of the fetus and newborn: a population study in the Netherlands. Transfusion. 2008;48(5):941-52.

SUPPLEMENTAL FILES

Table S1. General Characteristics of Studies and Patient Groups

Study	Study Type	Sample Size	Groups	Group Size	Data Collection Period	Country
Akdag 2012[22]	Case report	1	Neonate, K-mediated HDFN	1	NR	Turkey
Al-Dughaishi 2016[23]	Retrospective cohort	1,251	Pregnant women, D-mediated HDFN Pregnant women, K-mediated HDFN	8 2	June 2011–June 2013	Oman
Bek 2019[24]	Case report	1	Pregnant women, D-mediated HDFN	1	NR	Turkey
Bennardello 2013[25]	Questionnaire	1,661	Deliveries (Rh[D]- women), D-mediated HDFN	111	NR	Italy
			Deliveries, K-mediated HDFN	8		
			Neonates, D-mediated HDFN	111		
Bi 2019[26]	Prospective cohort	18	Neonates, D-mediated HDFN	5	January 2017–June 2019	PRC
Brumbaugh 2011[27]	Case report	1	Neonate, K-mediated HDFN	1	NR	USA
Canlorbe 2011[28]	Retrospective cohort	18	Pregnancies <20 weeks' gestation treated with IUT	18	January 1, 1990–August 15, 2011	France
		25	IUTs performed/attempted <20 weeks' gestation	25		
			IUTs performed/attempted <20 weeks' gestation, D-mediated HDFN	15		
			IUTs performed/attempted <20 weeks' gestation, K-mediated HDFN	1		
Chatziantoniou 2017[29]	Retrospective cohort	130	Pregnancies, D-mediated HDFN	32	June 2006–June 2013	United Kingdom
			Neonates, D-mediated HDFN	22		
			Neonate, K-mediated HDFN	1		

Study	Study Type	Sample Size	Groups	Group Size	Data Collection Period	Country
Colpo 2017[30]	Case report	1	Pregnancy, D-mediated HDFN	1	NR	Italy
			Neonate, D-mediated HDFN	1		
Craparo 2005[31]	Prospective cohort	31	Pregnancies, D-mediated HDFN treated with IUT	31	1998–2002	Italy
De Assuncão 2016[32]	Prospective cohort	13	Pregnancies, D-mediated HDFN treated with IUT	13	NR	Brazil
			Neonates, D-mediated HDFN	13		
Dubey 2016[33]	Prospective cohort	29	Pregnancies, D-mediated HDFN treated with IUT	31	NR	India
Fernandez Alba 2014[34]	Case report	1	Pregnancy, D-mediated HDFN	1	NR	Spain
			Neonate, D-mediated HDFN	1		
Fox 2008[35]	Case series	6	Pregnancies (D- or K-mediated HDFN) in whom in a previous pregnancy the fetus had been hydropic and severely anemic Pregnancies <20 weeks' gestation associated with a high perinatal mortality	6	1997–2004	United Kingdom
			Pregnancies treated with IUT only	4		
			Pregnancies treated with maternal IVIG and IUT	2		
Gottvall 2008[36]	Retrospective cohort	78,145	Pregnancies, D-mediated HDFN	71	January 1992–December 2005	Sweden
			Neonates, D-mediated HDFN	71		
			Pregnancies, K-mediated HDFN	9		
Gudlaugsson 2020[37]	Retrospective cohort	132	Deliveries (Rh[D]- women), D-mediated HDFN	130	1996–2015	Iceland
			Fetuses, D-mediated HDFN	112		

Study	Study Type	Sample Size	Groups	Group Size	Data Collection Period	Country
Haider 2020[38]	Case report	1	Neonate	1	NA	Pakistan
Harper 2006[39]	Prospective cohort	18	Hydropic fetus, D-mediated HDFN (case #3)	1	July 1985–October 1995	USA
			Hydropic fetus, K-mediated HDFN (case #7)	1		
			Hydropic fetus, D-mediated HDFN (case #10)	1		
			Hydropic fetus, K-mediated HDFN (case #13)	1		
			Hydropic fetus, D-mediated HDFN (case #15)	1		
Hassan 2019[40]	Case report	1	Neonate, D-mediated HDFN	1	NA	Malaysia
Karagol 2012[41]	Retrospective cohort	106	Neonates, K-mediated HDFN	5	January 2005–December 2010	Turkey
Kriplani 2007[42]	Case series	4	Pregnancies, D-mediated HDFN	4	NR	India
			Neonates, D-mediated HDFN	4		
Lakhwani 2011[43]	Case report	1	Pregnancy, K-mediated HDFN	1	NR	Spain
			Neonate, K-mediated HDFN	1		
Levy-Zauberman 2011[44]	Case report	1	Pregnancy, D-mediated HDFN	1	NR	France
			Neonate, D-mediated HDFN	1		
Lieberman 2020[45]	Retrospective cohort	128	Mothers, D-mediated HDFN Mothers, K-mediated HDFN	16 2	November 2010–June 2017	Canada
			Neonates, D-mediated HDFN Neonates, K-mediated HDFN	18 2		
Manoura 2007[46]	Case report	1	Pregnancy, K-mediated HDFN	1	NR	Greece
			Neonate, K-mediated HDFN	1		
Matijevic 2005[47]	Retrospective cohort	23	Pregnancies, D-mediated HDFN Pregnancies, K-mediated HDFN	15 1	January 1997–January 2003	Croatia

Study	Study Type	Sample Size	Groups	Group Size	Data Collection Period	Country
Mayer 2018[48]	Case series	3	Pregnancies, D- or K-mediated HDFN	2	NR	Germany
			Neonates, D- or K-mediated HDFN	2		
Meraj 2015[49]	Retrospective cohort	8	Fetuses, D-mediated HDFN treated with IUT	8	January 2001–December 2001	Pakistan
			Hydropic fetuses, D-mediated HDFN treated with IUT	4		
			Non-hydropic fetuses, D-mediated HDFN treated with IUT	4		
			Neonates, D-mediated HDFN treated with IUT	5		
Navarro 2009[50]	Case series	3	Neonate, D-mediated HDFN (case #3)	1	NR	USA
Nwogu 2018[51]	Case series	5	Fetuses, D-mediated HDFN Fetus, D- and K-mediated HDFN Fetus, K-mediated HDFN	3 1 1	November 2011–December 2015	USA
Palfi 2006[52]	Case report	1	Pregnancy, D-mediated HDFN	1	NA	Sweden
			Neonate, D-mediated HDFN	1		
Phung 2018[53]	Retrospective cohort	106	Fetuses, isolated D-mediated HDFN treated with IUT	29	1999–2015	France
			Neonates, isolated D-mediated HDFN treated with IUT	27		
Raguz 2020[2]	Retrospective cohort	29,663 pregnancies, 545 pos antibodies, 175 anti-D, 21 Kell 310 newborns with HDFN, 59 anti-D newborns, 5 Kell newborns	Infants, D-mediated HDFN	59	2000–2019	Bosnia and Herzegovina
			Infants, K-mediated HDFN	5		

Study	Study Type	Sample Size	Groups	Group Size	Data Collection Period	Country
Rahimi-Levene 2020[54]	Retrospective cohort	90,948	Pregnant women, D-mediated HDFN	63	January 1, 2011–December 31, 2011	Israel
			Pregnant women, K-mediated HDFN	114		
Rahimi-Sharbaf 2007[55]	Case report	1	Pregnancy, D-mediated HDFN	1	NA	Iran
Rath 2011[56]	Retrospective cohort	191	(Near)-term neonates, D-mediated HDFN (Near)-term neonates, K-mediated HDFN	157 34	January 2000–December 2008	The Netherlands
			Neonate, D-mediated HDFN	1		
Rath 2012[57]	Retrospective cohort	362	Infants, D-mediated HDFN	268	January 2000–September 2010	The Netherlands
			Infants, K-mediated HDFN	51		
Rath 2013[58]	Observational cohort	35	Neonate, D-mediated HDFN	1	November 2010–March 2012	The Netherlands
			Neonates, HDFN and cholestasis	5		
Rath 2013[59]	Retrospective cohort	125	Neonates, D-mediated HDFN	103	January 2000–October 2011	The Netherlands

Study	Study Type	Sample Size	Groups	Group Size	Data Collection Period	Country
Ree 2019[60]	Observational cohort	298	Infants, D-mediated HDFN	224	January 2006–January 2018	The Netherlands
			Infants, D-mediated HDFN treated with IUT	148		
			Infants, D-mediated HDFN not treated with IUT	76		
			Infants, D-mediated HDFN treated with IUT and RBC transfusion	134		
			Infants, D-mediated HDFN treated with IUT and not with RBC transfusion	14		
			Infants, D-mediated HDFN not treated with IUT and with RBC transfusion	53		
			Infants, D-mediated HDFN not treated with IUT or RBC transfusion	23		
			Infants, K-mediated HDFN	39		
Ree 2020[61]	Observational cohort	235	Fetuses, D- or K-mediated HDFN	235	January 2005–December 2018	The Netherlands
			Fetuses, D-mediated HDFN	189		
			Fetuses, K-mediated HDFN	46		
			Neonates, D-mediated HDFN	189		
			Neonates, K-mediated HDFN	46		
Ree 2020[62]	Retrospective cohort	317	Infant, D-mediated HDFN and NEC (case #1)	1	January 2000–December 2016	The Netherlands
			Infant, D-mediated HDFN and NEC (case #2)	1		
			Infant, K-mediated HDFN and NEC (case #4)	1		

Study	Study Type	Sample Size	Groups	Group Size	Data Collection Period	Country
Ruma 2007[63]	Multicenter case series	9	Pregnancies, D- or K-mediated HDFN	9	1996–2005	USA
			Pregnancies, D-mediated HDFN	5		
			Pregnancies, K-mediated HDFN	4		
			Infants, D- or K-mediated HDFN	9		
Sainio 2015[64]	Retrospective cohort	104	Pregnancies, K-mediated HDFN treated with IUT	8	2003–2012	Finland
			Pregnancies, D-mediated HDFN treated with IUT	86		
		339	IUTs	339		
			Intrauterine deaths associated with IUTs	4		
Santos 2013[65]	RCT	92	Infants, D-mediated HDFN receiving IVIG	46	April 2006–June 2009	Brazil
			Infants, D-mediated HDFN receiving placebo	46		
			Infants, D-mediated HDFN receiving IVIG, no IUT	42		
			Infants, D-mediated HDFN receiving placebo, no IUT	40		
Şavkli 2020[66]	Retrospective cohort	110	IUTs IUTs in pregnancies where fetuses were hydropic at first IUT, D-mediated HDFN	51	January 2015–July 2018	Turkey
			IUTs in pregnancies where fetuses were not hydropic at first IUT, D-mediated HDFN	59		
		42	Pregnancies treated with IUT, D-mediated HDFN	42		

Study	Study Type	Sample Size	Groups	Group Size	Data Collection Period	Country
Sikkel 2005[67]	Prospective cohort	49 transfusions, 23 fetuses	IUTs in D-mediated HDFN group	41	March 2001–May 2002	The Netherlands
			IUTs in K-mediated HDFN group	3		
Simonazzi 2016[68]	Case series	4	Fetus, D-mediated HDFN (case #1)	1	NR	Italy
			Fetus, D-mediated HDFN (case #2)	1		
			Fetus, D-mediated HDFN (case #3)	1		
Somerset 2006[69]	Retrospective cohort	66	Anemic fetuses, D-mediated HDFN	57	March 1997–March 2004	USA
			Anemic fetuses, K-mediated HDFN	7		
Takci 2013[70]	Retrospective cohort	30	Hydropic infants, D-mediated HDFN	30	January 2001–June 2012	Turkey
			Hydropic infants, D-mediated HDFN, non-cholestasis group	12		
			Hydropic infants, D-mediated HDFN, cholestasis group	18		
Tara 2019[71]	Case report	1	Fetus, D-mediated HDFN	1	NR	Iran
Temel 2019[72]	Prospective cohort	17	Pregnant women, D-mediated HDFN	17	January 2018–June 2019	Turkey
			Neonates, D-mediated HDFN	17		

Study	Study Type	Sample Size	Groups	Group Size	Data Collection Period	Country
Tiblad 2011[73]	Retrospective cohort	84	Fetuses, D-mediated HDFN	67	June 1990–June 2010	Sweden
			Fetuses, K-mediated HDFN	9		
			Fetus, D-mediated HDFN (case #511)	1		
			Fetus, D-mediated HDFN (case #512)	1		
			Fetus, D-mediated HDFN (case #514)	1		
			Fetus, D-mediated HDFN (case #518; 1991 pregnancy)	1		
			Fetus, D-mediated HDFN (case #518; 1993 pregnancy)	1		
			Fetus, D-mediated HDFN (case #249)	1		
			Fetus, D-mediated HDFN (case #462)	1		
			Fetus, D-mediated HDFN (case #459)	1		
			Fetus, D-mediated HDFN (case #343)	1		
Urutherakumar 2020[74]	Retrospective cohort	39	Transfused fetus (case #4)	1	January 1, 2005–July 30, 2017	Australia
Van Den Akker 2008[75]	Retrospective cohort	42	Fetuses, K-mediated HDFN	42	January 1988–December 2005	The Netherlands
			Severely hydropic fetuses, K-mediated HDFN	15		
Walsh 2013[76]	Retrospective cohort	102	Fetuses, D-mediated HDFN	26	January 1, 1996–December 31, 2011	Ireland
			Fetuses, K-mediated HDFN	11		
Xu 2013[77]	Case report	1	Neonate	1	NA	PRC
Zwiers 2017[78]	Retrospective cohort	589	Neonate	1	January 1988–January 2015	The Netherlands

Study	Study Type	Sample Size	Groups	Group Size	Data Collection Period	Country
Zwiers 2018[79]	Retrospective cohort	645	Fetuses, D-mediated HDFN,	524	January 1, 1987–December 31, 2016	The Netherlands
			Fetuses, K-mediated HDFN	83		
			Fetuses, D-mediated HDFN, 1999 onward cohort	334		
			Fetuses, K-mediated HDFN, 1999 onward cohort	65		
Zwiers 2018[80]	Retrospective multicenter cohort	52	Women (D- or K-mediated HDFN) managed with IVIG, unadjusted data	24	January 2010–June 2016	The Netherlands
			Women (D- or K-mediated HDFN) managed without IVIG, unadjusted data	28		
			Women (D- or K-mediated HDFN) managed with IVIG, weighted data[a]	24		
			Women (D- or K-mediated HDFN) managed without IVIG, weighted data[a]	25		

D, Rh(D); HDFN, hemolytic disease of the fetus and newborn; IUT, intrauterine transfusion; IVIG, intravenous immunoglobulin; K, Kell; NA, not applicable; NEC, necrotizing enterocolitis; NR, not reported; PRC, People's Republic of China; RCT, randomized controlled trial; RBC, red blood cell; Rh, Rhesus; USA, United States of America.
For D- or K-mediated HDFN, data were captured only for cases with immunization types of D only, K only, or both D and K only.
[a]Data were corrected for the probability that women would be selected for IVIG treatment by their caregivers in the index pregnancy. Propensity scores were calculated using gestational age at onset of anemia in the previous pregnancy, interval between pregnancies, number of previous births, type of alloimmunization, maternal body mass index, and number of IUTs in the previous pregnancy.

Table S2. Research Questions Relevant to Mothers and Fetuses

Treatment landscape	What is the current treatment landscape of HDFN in mothers and fetuses?
	What are the commonly used treatments?
Clinical evidence of HDFN treatments	What are the maternal and fetal clinical outcomes?
	What is the efficacy of intravenous immunoglobulin in women with HDFN?
	What are the complications of treatment (eg, adverse events)?
	What is the prevalence of HDFN, and what is the prevalence of early-onset HDFN[a]?
Evidence gaps and recommendations	What are the evidence gaps in the HDFN literature?
	What recommendations can be proposed for future research?

HDFN, hemolytic disease of the fetus and newborn.
[a]HDFN requiring intervention before 24 weeks of pregnancy.

Table S3. Inclusion and Exclusion Criteria[a]

	Inclusion Criteria	Exclusion Criteria
Population	• Women and/or their fetus/infant/child experiencing or having experienced HDFN caused by Rh incompatibility • Antigen status: Rh(D) or Kell antigen	• Women and fetus/infant/child who did not experience HDFN with ABO incompatibility • Antigen status: c, e, E, Duffy (Fy), Kidd (Jk), MNS (S), or Gerbich antigen
Intervention	• Any or none	• Not applicable
Comparator	• Any or none	• Not applicable
Outcomes	• Treatment patterns • Clinical outcomes • Treatment efficacy	• Not applicable
Study design	• Observational studies (retrospective or prospective) including cohort, case-control, or cross-sectional studies • Trials (randomized or nonrandomized) • Modelling studies • Systematic reviews of cohort studies (for identification of primary studies only) • Case reports and case series	• Notes, editorials, or commentaries • Nonsystematic reviews
Other	• Journal articles • Human subjects • English language • Studies published between January 1, 2005, and March 10, 2021	• Indexed conference abstracts • Publication types not of interest • Animal or preclinical studies • Non-English language • Studies published before January 1, 2005

HDFN, hemolytic disease of the fetus and newborn; Rh, Rhesus.
[a]Population, intervention, comparison, outcomes, and study (PICOS) design was used as a framework to formulate eligibility criteria.[61]

Table S4a. Methodologic Quality of Selected Case Reports

JBI Critical Appraisal Checklist for Case Reports									
	Q1	**Q2**	**Q3**	**Q4**	**Q5**	**Q6**	**Q7**	**Q8**	**Total Score**
Akdağ et al	Yes	Yes	Yes	Yes	Yes	No	Yes	Yes	**7/8**
Bek et al	Yes	No	Yes	Yes	Yes	Yes	Yes	Yes	**7/8**
Brumbaugh et al	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	**8/8**
Colpo et al	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	**8/8**
Fernández Alba et al	Yes	Yes	Yes	Yes	Yes	No	Unc	Yes	**6/8**
Haider et al	Yes	No	Yes	Yes	Yes	Yes	Unc	Yes	**6/8**
Hassan et al	No	Yes	No	Yes	No	Yes	Yes	Yes	**5/8**
Lakhwani et al	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	**8/8**
Levy-Zauberman et al	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	**8/8**
Manoura et al	No	Unc	No	Yes	Yes	Unc	Unc	Yes	**3/8**
Palfi et al	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	**8/8**
Rahimi-Sharbaf et al	Yes	Yes	No	Yes	No	No	Unc	Yes	**4/8**
Tara et al	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	**8/8**
Xu et al	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	**8/8**

JBI, Joanna Briggs Institute; Unc, unclear.
Q1: Were patient's demographic characteristics clearly described?
Q2: Was the patient's history clearly described and presented as a timeline?
Q3: Was the current clinical condition of the patient on presentation clearly described?
Q4: Were diagnostic tests or assessment methods and the results clearly described?
Q5: Was the intervention(s) or treatment procedure(s) clearly described?
Q6: Was the post-intervention clinical condition clearly described?
Q7: Were adverse events (harms) or unanticipated events identified and described?
Q8: Does the case report provide takeaway lessons?

Table S4b. Methodologic Quality of Selected Case Series

JBI Critical Appraisal Checklist for Case Series											
	Q1	**Q2**	**Q3**	**Q4**	**Q5**	**Q6**	**Q7**	**Q8**	**Q9**	**Q10**	**Total Score**
Fox et al	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	NA	**9/9**
Kriplani et al	No	Yes	Yes	Yes	Unc	Yes	Yes	Unc	No	NA	**5/9**
Mayer et al	No	Yes	Yes	Unc	Unc	Yes	Yes	Unc	No	NA	**4/9**
Navarro et al	No	Yes	Yes	Unc	Unc	Yes	Yes	Yes	No	NA	**5/9**
Nwogu et al	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No	Yes	NA	**8/9**
Ruma et al	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	**10/10**
Simonazzi et al	No	Yes	Yes	Unc	Unc	Yes	Yes	Yes	No	NA	**5/9**

JBI, Joanna Briggs Institute; Unc, unclear.
NA = Not applicable
Q1: Were there clear criteria for inclusion in the case series?
Q2: Was the condition measured in a standard, reliable way for all participants included in the case series?
Q3: Were valid methods used for identification of the condition for all participants included in the case series?
Q4: Did the case series have consecutive inclusion of participants?
Q5: Did the case series have complete inclusion of participants?
Q6: Was there clear reporting of the demographics of the participants in the study?
Q7: Was there clear reporting of clinical information of the participants?
Q8: Were the outcomes or follow up results of cases clearly reported?
Q9: Was there clear reporting of the presenting site(s)/clinic(s) demographic information?
Q10: Was statistical analysis appropriate?

Table S4c. Methodologic Quality of Selected Retrospective Cohort Studies

Newcastle-Ottawa Scale (Retrospective Cohort Studies)										
	Selection				Comparability	Outcome				
	I1	I2	I3	I4	I5	I6	I7	I8	Total Score	Quality
Al-Dughaishi 2016	*	*	*	*		*			5	Poor
Åžavkli 2020	*	*	*	*	*	*	*	*	8	Good
Canlorbe 2011	*	*	*	*		*	*	*	7	Fair
Chatziantoniou 2017	*	*	*	*	*	*	*	*	8	Good
Gottvall 2008	*	*	*	*	*	*	*	*	8	Good
Gudlaugsson 2020	*	*	*	*	*	*	*	*	8	Good
Karagol 2012	*	*	*	*	*	*	*	*	8	Good
Lieberman 2020	*	*	*	*	*	*	*	*	8	Good
Matijevic 2005	*		*	*		*	*	*	6	Fair
Meraj 2015	*		*	*		*	*	*	6	Fair
Phung 2018	*	*	*	*		*	*	*	7	Fair
Rahimi-Levene 2020	*	*	*	*		*	*	*	7	Fair
Rath 2012	*	*	*	*	*	*	*	*	8	Good
Rath 2013a	*	*	*	*	*	*	*	*	8	Good
Rath 2013b	*	*	*	*	*	*	*	*	8	Good
Ree 2019	*	*	*	*	*	*	*	*	8	Good
Ree 2020a	*	*	*	*	*	*	*	*	8	Good
Ree 2020b	*	*	*	*	*	*	*	*	8	Good
Sainio 2015	*	*	*	*	*	*	*	*	8	Good
Somerset 2006	*	*	*	*	*	*	*	*	8	Good
Takci 2013	*	*	*	*		*	*	*	7	Fair
Tiblad 2011	*	*	*	*	*	*	*	*	8	Good
Urutherakumar 2020	*	*	*	*		*	*	*	7	Fair
Van Den Akker 2008a	*	*	*	*	*	*	*	*	8	Good
Walsh 2013	*	*	*	*	*	*	*	*	8	Good
Zwiers 2017	*	*	*	*	*	*	*	*	8	Good
Zwiers 2018a	*	*	*	*	*	*	*	*	8	Good
Rath 2011	*	*	*	*	*	*	*	*	8	Good
Raguz 2020	*	*	*	*		*	*	*	7	Fair
Zwiers 2018b	*	*	*	*	*	*	*	*	8	Good

I1: Representativeness of the exposed cohort
I2: Selection of the non-exposed cohort
I3: Ascertainment of exposure
I4: Demonstration that outcome of interest was not present at start of study
I5: Comparability of cohorts on the basis of the design or analysis controlled for confounders
I6: Assessment of outcome
I7: Was follow-up long enough for outcomes to occur
I8: Adequacy of follow-up of cohorts

Table S4d. Methodologic Quality of Selected Prospective Cohort Studies

	Newcastle-Ottawa Scale (Prospective Cohort Studies)									
	Selection				Comparability	Outcome				
	I1	I2	I3	I4	I5	I6	I7	I8	Total Score	Quality
Bi 2019	*		*	*	*	*	*	*	7	Good
Craparo 2005	*		*	*	*	*	*	*	7	Good
de Assunção 2016	*	*	*	*	*	*	*	*	8	Good
Dubey 2016	*	*	*	*	*	*	*	*	8	Good
Sikkel 2005	*	*	*	*	*	*	*	*	8	Good
Temel 2019	*	*	*	*	*	*	*	*	8	Good
Harper 2006	*	*	*	*	*	*	*	*	8	Good
Koelewijn 2008	*	*	*	*	*	*	*	*	8	Good

I1: Representativeness of the exposed cohort
I2: Selection of the non-exposed cohort
I3: Ascertainment of exposure
I4: Demonstration that outcome of interest was not present at start of study
I5: Comparability of cohorts on the basis of the design or analysis controlled for confounders
I6: Assessment of outcome
I7: Was follow-up long enough for outcomes to occur
I8: Adequacy of follow-up of cohorts

Table S4e. Methodologic Quality of Bennardello et al(25)

Item	Bennardello et al
Design	
Is the target population defined?	Yes, all Italian Transfusion Structures
IRB approval and informed consent process	
IRB approval	No report
Informed consent	No report
Data protection	No report
Development and pre-testing	
Development and testing	The SIMTI set up a Working Group to design a multiple choice questionnaire
Recruitment process and description of the sample having access to the questionnaire	
Open survey versus closed survey	Closed survey
Contact mode	Website, fax, or email
Advertising the survey	Internal communication within the organization

Item	Bennardello et al
Survey administration	
Web/e-mail	Online entry was possible
Context	SIMTI website, specifically for Italian Transfusion Structures
Mandatory/voluntary	Voluntary
Incentives	No incentives reported
Time/date	2011
Randomization of items or questionnaires	NA
Adaptive questioning	No report
Number of items	37 questions
Number of screens (pages)	5 sections
Completeness check	The mean percentage of questions answered was 76%
Review step	No report
Response rates	
Unique site visitor	The 280 Italian Transfusion Structures were invited via the SIMTI website
View rate (ratio of unique survey visitors/unique site visitors)	NA
Participation rate (ratio of unique visitors who agreed to participate/unique first survey page visitors)	176/280 overall (participation rate differed be region)
Completion rate (ratio of users who finished the survey/users who agreed to participate)	164/280
Preventing multiple entries from the same individual	
Cookies used	No report
IP check	No report
Log file analysis	No report
Registration	Yes, per Italian Transfusion Structure
Analysis	
Handling of incomplete questionnaires	Incomplete questionnaires were included for analysis
Questionnaires submitted with an atypical timestamp	No report
Statistical correction	NA

IRB, institutional review board; NA, not applicable; SIMTI, The Italian Society of Transfusion Medicine and Immunohaematology.

Table S5. Antenatal Treatments Reported in Studies With Representative Data

Citation	Population	D Only and/or K Only[a]		Treatment (%)[b]
IUT Only				
Santos 2013[45]	4/46 infants treated with IVIG[b,c] 6/46 infants given placebo[c]	D D	-- --	8.7* 13.0*
Dubey 2016[12]	17/29	D	--	58.6
Chatziantoniou 2017[8]	2/32	D	--	6.3*
Gottvall 2008[15]	0/9	--	K	0
Gudlaugsson 2020[16]	5/112	D	--	4.5*
Rath 2011[36]	104/157 28/34	D --	-- K	66 82
Rath 2013[39]	61/103	D	--	59
Ree 2019[40]	148/224 35/39	D --	-- K	66 90
Bennardello 2013[4]	8/111 1/8	D --	-- K	7.2 12.5
IVIG Only				
Mayer 2018[27]	1/1 1/1	D --	-- K	100 100
IUT+IVIG				
Ree 2020[42]	6/189 treated with IUT+IVIG[d]	D	--	3.2*
	2/46 treated with IUT+IVIG[c]	--	K	4.3*
Other				
Tara 2019[51]	1/1	D	--	TPE, 100
Gottvall 2008[15]	10/71	D	--	Maternal plasma exchange and/or high-dose IVIG, 14.1*
Nwogu 2018[30]	5/5	D and/or K		TPE, IVIG, and IUT, 100
Palfi 2006[31]	1/1	D	--	TPE, IVIG, and IUT,100
Colpo 2017[9]	1/1	D	--	TPE, immunoadsorption, IVIG, and IUT, 100
Lakhwani 2011[22]	1/1	--	K	Plasmapheresis and IUT, 100
Fernandez Alba 2014[13]	1/1	D	--	Plasmapheresis and IVIG, 100
Zwiers 2018[60]	8/24	D or K		Plasmapheresis, IVIG, and IUT, 33.3*
Ruma 2007[43]	9/9	D or K		Plasmapheresis, IVIG, and IUT, 100
Bek 2019[3]	1/1	D	--	Plasmapheresis, IVIG, and IUT, 100

D, Rh(D); HDFN, hemolytic disease of the fetus and newborn; IUT, intrauterine transfusion; IVIG, intravenous immunoglobulin; K, Kell; Rh, Rhesus; TPE, therapeutic plasma exchange.
[a]Data were captured only for cases with immunization types of D only, K only, or both D and K only.
[b]Values are as provided in reports, unless otherwise noted (*values that were calculated).
[c]Studied to evaluate the need for exchange transfusion among neonates randomly allocated to treatment with or without IVIG (both groups had intensive phototherapy).
[d]Studied for suppression of fetal erythropoiesis.

Table S6. Hydrops Fetalis, Severe Fetal Anemia, and Adverse Events/Complications Reported in Studies With Representative Data

Citation	Patient Group	N	D Only and/or K Only[a]		Events (n/N [%][b])
Hydrops Fetalis					
Craparo 2005[10]	Pregnancies treated with IUT	31	D	--	7/31 (23)
de Assuncão 2016[11]	Pregnancies treated with IUT	13	D	--	3/13 (23.1)
Sikkel 2005[47]	IUTs	41	D	--	4/41 (9.8)
	IUTs	3	--	K	2/3 (66.7)
Al-Dughaishi 2016[2]	Pregnant women	8	D and/or K		0/8 (0)
Canlorbe 2011[7]	IUTs performed <20 weeks' gestation	25	D or K		11/25 (44)*
Karagol 2012[20]	Neonates	5	--	K	0/5 (0)
Matijevic 2005[26]	Pregnancies	15 1	D --	-- K	3/15 (18.8)* 0/1 (0)
Meraj 2015[28]	Fetuses treated with IUT	8	D	--	4/8 (50)
	Fetuses treated with IUT, hydropic	4	D	--	4/4 (100)
	Fetuses treated with IUT, non-hydropic	4	D	--	0/4 (0)
Phung 2018[32]	Fetuses treated with IUT for isolated D-mediated HDFN	29	D	--	4/29 (13.8)
Rath 2013[39]	Neonates	103	D	--	2/103 (1.9)
Somerset 2006[49]	Pregnancies complicated by fetal anemia requiring IUT	57	D	--	15/57 (26.3)*
		7	--	K	2/7 (28.6)*
Sainio 2015[44]	Pregnancies	104	D or K		12/104 (11.5)
		8	--	K	3/8 (37.5)
	Intrauterine deaths associated with IUTs	3	D	--	1/3 (33.3)*
		1	--	K	1/1 (100)*
Şavkli 2020[46]	Pregnancies treated with IUT	42	D	--	14/42 (33.3)
	IUTs in pregnancies where fetuses were hydropic at first IUT	51	D	--	51/51 (100)
	IUTs in pregnancies where fetuses were not hydropic at first IUT	59	D	--	0/59 (0)
van den Akker 2008[55]	Fetuses	42	--	K	15/42 (36) (severe) 9/42 (21) (mild)*
	Severely hydropic fetuses	15	--	K	15/15 (100) (severe)
Walsh 2013[56]	Fetuses	26	D	--	0/26 (0)
		11	--	K	2/11 (17)
Zwiers 2018[59]	Fetuses, 1999 onward cohort	334	D	--	40/338 (12)
	Fetuses, 1999 onward cohort	65	--	K	21/65 (32)

Citation	Patient Group	N	D Only and/ or K Only[a]	Events (n/N [%][b])
Zwiers 2018[60]	Unadjusted data, IVIG group	24	D or K	1/24 (4)
	Unadjusted data, non-IVIG group	28	D or K	7/28 (24)
	Weighted data,[c] IVIG group	24	D or K	1/24 (4)
	Weighted data,[c] non-IVIG group	25	D or K	12/25 (46)
Kriplani 2007[21]	Pregnancies	4	D --	2/4 (50)*
Nwogu 2018[30]	Fetuses, D-mediated HDFN, antenatal treatment with IVIG and IUT	3	D --	0/3 (0)
	Fetuses, D- and K-mediated HDFN	1	D K	0/1 (0)
	Fetuses, K-mediated HDFN	1	-- K	0/1 (0)
Severe Fetal Anemia[d]				
Canlorbe 2011[7]	Pregnancies with IUTs performed/ attempted ≤20 weeks' gestation	18	D or K	18/18 (100)
Adverse Events/Complications				
Dubey 2016[12]	Pregnancies	29	D --	Adverse serological outcomes, 17/29 (58.8)
Temel Yüksel 2019[52]	Fetuses treated with IUT	17	D --	non-reassuring fetal heart rate tracing, 3/17 (17.6)
Canlorbe 2011[7]	IUTs performed <20 weeks' gestation	25	D or K	Failed intravascular transfusion, 2/25 (8)*; bradycardia, 1/25 (4)*
Matijevic 2005[26]	Pregnancies	15	D --	Fetal death, 2/15 (13.3)*
		1	-- K	Fetal death, 0/1 (0)
Sainio 2015[44]	Pregnancies	104 86	D or K	Fetal death, 4/104 (3.8) Fetal death, 3/86 (3.5)
			D --	
		8	-- K	Fetal death, 1/8 (1.3)
		104	D or K	Severe fetal bradycardia requiring immediate cesarean section, 11/104 (10.6)
		104	D or K	Infection, PROM, or spontaneous preterm delivery <32 weeks of gestation, 9/104 (8.7)
	IUTs	339	D or K	Fetal death, 4/339 (1.2)
		339	D or K	Severe fetal bradycardia requiring immediate

Citation	Patient Group	N	D Only and/ or K Only[a]		Events (n/N [%][b])
					cesarean section, 11/339 (3.2)
		339	D or K		Infection, PROM, or spontaneous preterm delivery <32 weeks of gestation, 9/339 (2.7)*
	Intrauterine deaths associated with IUTs	3	D	--	PROM, 1/3 (33.3)*; infection, 1/3 (33.3)*
		1	--	K	0/1 (0)
Şavkli 2020[46]	IUTs	110	D	--	13-110 (11.8) (PROM, 0/110; preterm labor; 2/110 (1.8)*; fetal distress, 5/110 (4.5)*; fetal death, 6/110 (5.5)*
Somerset 2006[49]	Anemic fetuses	57	D	--	Bradycardia, 7/57 (12.3)*; spontaneous rupture of membranes, 3/57 (5.3)*; chorioamnionitis, 1/57 (1.8)*; immediate delivery 3/57 (5.3)*
	Anemic fetuses	7	--	K	Bradycardia, 1/7 (14.3)*; spontaneous rupture of membranes, 0/7 (0); chorioamnionitis, 1/7 (14.3)*; immediate delivery, 0/7 (0)
Zwiers 2018[60]	Unadjusted data, women managed without IVIG	28	D or K		PROM/premature delivery, 1/28 (3.7)
Ruma 2007[43]	Pregnancies	9	D or K		4/9 (44) (IVIG associated: headache, 2/9 [22.2]*; chest congestion, 1/9 [11.1]*; mild flushing and pruritis, 1/9 [11.1]*; plasmapheresis associated: 0/9 [0])

D, Rh(D); HDFN, hemolytic disease of the fetus and newborn; IUT, intrauterine transfusion; IVIG, intravenous immunoglobulin; K, Kell; NICU, neonatal intensive care unit; PROM, premature rupture of membranes; Rh, Rhesus; SD, standard deviation.

[a]Data were captured only for cases with immunization types of D only, K only, or both D and K only.

[b]Values are as provided in reports, unless otherwise noted (*for values that were calculated).

[c]Data were corrected for the probability that women would be selected for IVIG treatment by their caregivers in the index pregnancy. Propensity scores were calculated using gestational age at onset of anemia in the previous pregnancy, interval between pregnancies, number of previous births, type of alloimmunization, maternal body mass index, and number of IUTs in the previous pregnancy.

[d]≥5 SDs from fetal hemoglobin reference value (15 g/dL). 1 SD=1 g/dL difference from the reference value.

Table S7. IVIG Treatment Response and Associated Mortality

Citation	Patient Group	N	D Only and/or K Only[a]		Treatment (n/N [%][b])	Treatment Response
IVIG Treatment Response						
Ree 2020[42]	Fetuses	189	D	--	IUT only, 183/189 (96.8)*; IVIG+IUT, 6/189 (3.2)*	62% decline in absolute reticulocyte count[a]
	Fetuses	46	--	K	IUT only, 44/46 (95.6)*; IVIG+IUT, 2/46 (4.4)*	32% decline in absolute reticulocyte count[a]
Zwiers 2018[60]	Unadjusted data, women managed with IVIG	24	D or K		IVIG+IUT, 24/24 (100); plasmapheresis, 8/24 (33.3)	Delay of the need for IUT was on average 15 gestational days later
	Weighted data,[c] women managed with IVIG	24	D or K		IVIG+IUT, 24/24 (100); plasmapheresis, 4/24 (15)	Delay of the need for IUT was on average 0 gestational days later
Bek 2019[3]	Pregnancy	1	D	--	DFPP and IUT+IVIG, 1/1 (100)	Successful delivery at 30 weeks' gestation
Colpo 2017[9]	Pregnancy	1	D	--	TPE and IA + IVIG, 1/1 (100)	Patient required IUT because of severe fetal anemia at 30 weeks' gestation
Fernandez Alba 2014[13]	Pregnancy	1	D	--	Plasmapheresis and IVIG, 1/1 (100)	No need for IUT until delivery
Mayer 2018[27]	Pregnancies	2	D or K		IVIG, 2/2 (100)	IUT was prevented in both cases
IVIG-associated Mortality						
Fox 2008[14]	Pregnancies with prior pregnancy where fetus was hydropic and severely anemic before 20 weeks' gestation associated with a high perinatal mortality	6	D or K		IVT±IVIG, 6/6 (100)	1/6 (16.7)*
		2	D	--	IVT only, 2/2 (100)	1/2 (50)*
		3	D	--	IVT+IVIG, 3/3 (100)	0/3 (0)
		1	--	K	IVT+IVIG, 1/1 (100)	0/1 (0)
Kriplani 2007[21]	Pregnancies	4	D	--	IUT+IVIG, 4/4 (100)	0/0 (0)
Ruma 2007[43]	Pregnancies, all	9	D or K		Plasmapheresis+IVIG+ IUT, 9/9 (100)	0/0 (0)

D, Rh(D); DFPP, double-filtration plasmapheresis; IA, immunoadsorption; IUT, intrauterine transfusion; IVIG, intravenous immunoglobulin; IVT, intravascular infusion; K, Kell; Rh, Rhesus; TPE, therapeutic plasma exchange.
[a]Data were captured only for cases with immunization types of D only, K only, or both D and K only.
[b]Values are as provided in reports, unless otherwise noted (*for values that were calculated).
[c]Data were corrected for the probability that women would be selected for IVIG treatment by their caregivers in the index pregnancy. Propensity scores were calculated using gestational age at onset of anemia in the previous pregnancy, interval between pregnancies, number of previous births, type of alloimmunization, maternal body mass index, and number of IUTs in the previous pregnancy.

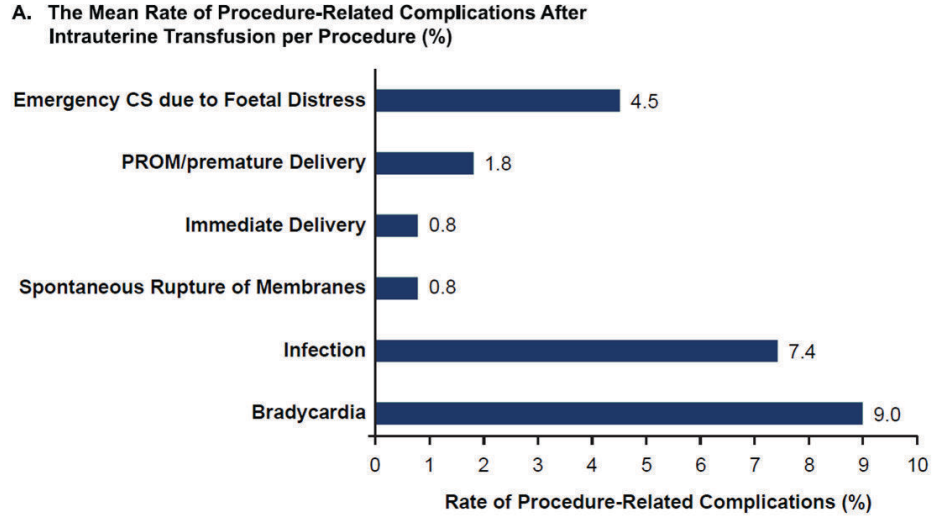
Table S8. Overall Fetal Mortality Associated With HDFN

Citation	Patient Group	N	D Only and/or K Only[a]		Treatment (n/N [%][b])	Mortality Rate (n/N [%])
Craparo 2005[10]	Pregnancies treated with IUT	31	D	--	IUT, 31/31 (100)	4/31 (12.9)*
Canlorbe 2011[7]	Pregnancies with IUT <20 weeks' gestation	18	D or K		IUT, 18/18 (100)	3/18 (16.7)*
Lieberman 2020[24]	Mothers	16	D	--	No treatment, 16/16 (100)	0/16 (0)
	Mothers	2	--	K	No treatment, 2/2 (100)	0/2 (0)
Matijevic 2005[26]	Pregnancies	23	D or K		IUIVT, 9/13 (39.1)* (9/13* who had cordocentesis); No IUIVT, 4/13 (60.9)* (4/13 who had cordocentesis + 10/23 who did not have cordocentesis)	3/23 (13.0)
Meraj 2015[28]	Fetuses treated with IUT	8	D	--	IUT, 8/8 (100)	1/8 (12.5)*
Phung 2018[32]	Fetuses, isolated D-mediated HDFN treated with IUT	29	D	--	IUT, 29/29 (100)	2/29 (6.9)
Sainio 2015[44]	Pregnancies	94	D or K		IUT, 94/94 (100)	4/94 (3.8)
		86	D	--	IUT, 86/86 (100)	3/86 (3.5)*
		8	--	K	IUT, 8/8 (100)	1/8 (12.5)
Şavkli 2020[46]	Pregnancies treated with IUTs	42	D	--	IUT, 42/42 (100)	6/42 (14.3)
Somerset 2006[49]	Anemic fetuses	57	D	--	IUT, 57/57 (100)	5/57 (8.8)*
	Anemic fetuses	7	--	K	IUT, 7/7 (100)	1/7 (14.3)*
Tiblad 2011[53]	Fetus (case #511)	1	D	--	IUT, 1/1 (100)	0/1 (0)
	Fetus (case #512)	1	D	--	IUT, 1/1 (100)	0/1 (0)
	Fetus (case #514)	1	D	--	IUT, 1/1 (100)	0/1 (0)
	Fetus (case #518; 1991 pregnancy)	1	D	--	IUT, 1/1 (100)	1/1 (100)
	Fetus (case #518; 1993 pregnancy)	1	D	--	IUT, 1/1 (100)	0/1 (0)
	Fetus (case #249)	1	D	--	IUT, 1/1 (100)	1/1 (100)
van den Akker 2008[55]	Severely hydropic fetuses	15	--	K	IUT, 15/15 (100)	2/15 (13)
Walsh 2013[56]	Fetus	11	--	K	IUT, 11/11 (100)	1/11 (9.1)
Zwiers 2017[58]	Fetus	1	D	--	IUT, 1/1 (100); interstitial laser, 1/1 (100)	1/1 (100)[d]
Zwiers 2018[60]	Unadjusted data, women managed with no IVIG	28	D or K		IUT, 28/28 (100)	4/28 (15)
	Weighted data,[c] women managed with no IVIG	25	D or K		IUT, 25/25 (100)	3/25 (12)

Citation	Patient Group	N	D Only and/or K Only[a]		Treatment (n/N [%][b])	Mortality Rate (n/N [%])
Fox 2008[14]	Pregnancies with whom in a previous pregnancy the fetus had been hydropic and severely anemic prior to 20 weeks' gestation associated with a high perinatal mortality	5	D or K		IVT±IVIG, 5/5 (100)	1/5 (20)
		2	D	--	IVT only, 2/2 (100)	1/2 (50)*
		2	D	--	IVT+IVIG, 2/2 (100)	0/2 (0)
		1	--	K	IVT+IVIG, 1/1 (100)	0/1 (0)
Kriplani 2007[21]	Pregnancies	4	D	--	IUT+IVIG, 4/4 (100)	0/4 (0)
Lakhwani 2011[22]	Pregnancy	1	--	K	Plasmapheresis+IUT, 1/1 (100)	0/1 (0)
Ruma 2007[43]	Pregnancies, all	9	D or K		Plasmapheresis+ IVIG+IUT, 9/9 (100)	0/9 (0)
Simonazzi 2016[48]	Pregnancy (case # 1)	1	D	--	IUT, 1/1 (100)	1/1 (100)

D, Rh(D); HDFN, hemolytic disease of the fetus and newborn; IUIVT, intrauterine intravascular transfusion; IUT, intrauterine transfusion; IVIG, intravenous immunoglobulin; IVT, intravascular infusion; K, Kell; Rh, Rhesus.
[a]Data were captured only for cases with immunization types of D only, K only, or both D and K only.
[b]Values are as provided in reports, unless otherwise noted (*for values that were calculated).
[c]Data were corrected for the probability that women would be selected for IVIG treatment by their caregivers in the index pregnancy. Propensity scores were calculated using gestational age at onset of anemia in the previous pregnancy, interval between pregnancies, number of previous births, type of alloimmunization, maternal body mass index, and number of IUTs in the previous pregnancy.
[d]During the study period, 39 cases of fetal or neonatal death occurred after IUT, of which 10 occurred from 2001 onward. However, number of fetal versus neonatal deaths were not reported separately.

SUPPLEMENTAL FIGURES



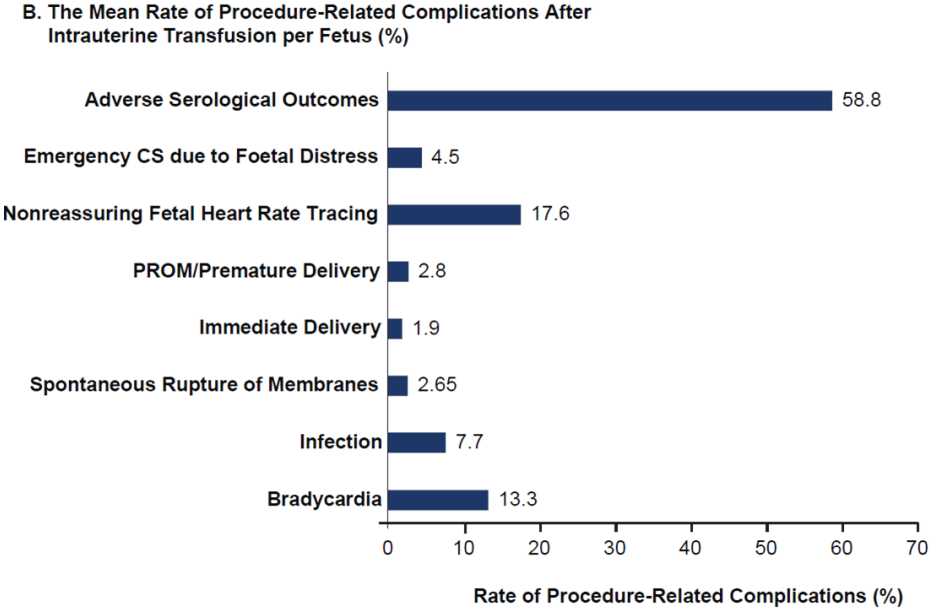


Figure S1. Mean Rate of Procedure-related Complications After IUT (Per Procedure [A] and Per Fetus [B]).[12,26,30,32,43,44,46,49,52,60]

CS, cesarean section; IUT, intrauterine transfusion; PROM, preterm rupture of membranes.
[a]Adverse serological outcome was defined as the development of additional maternal antibodies to anti-D and/or a ≥4-fold enhancement of antibody titer.[12]

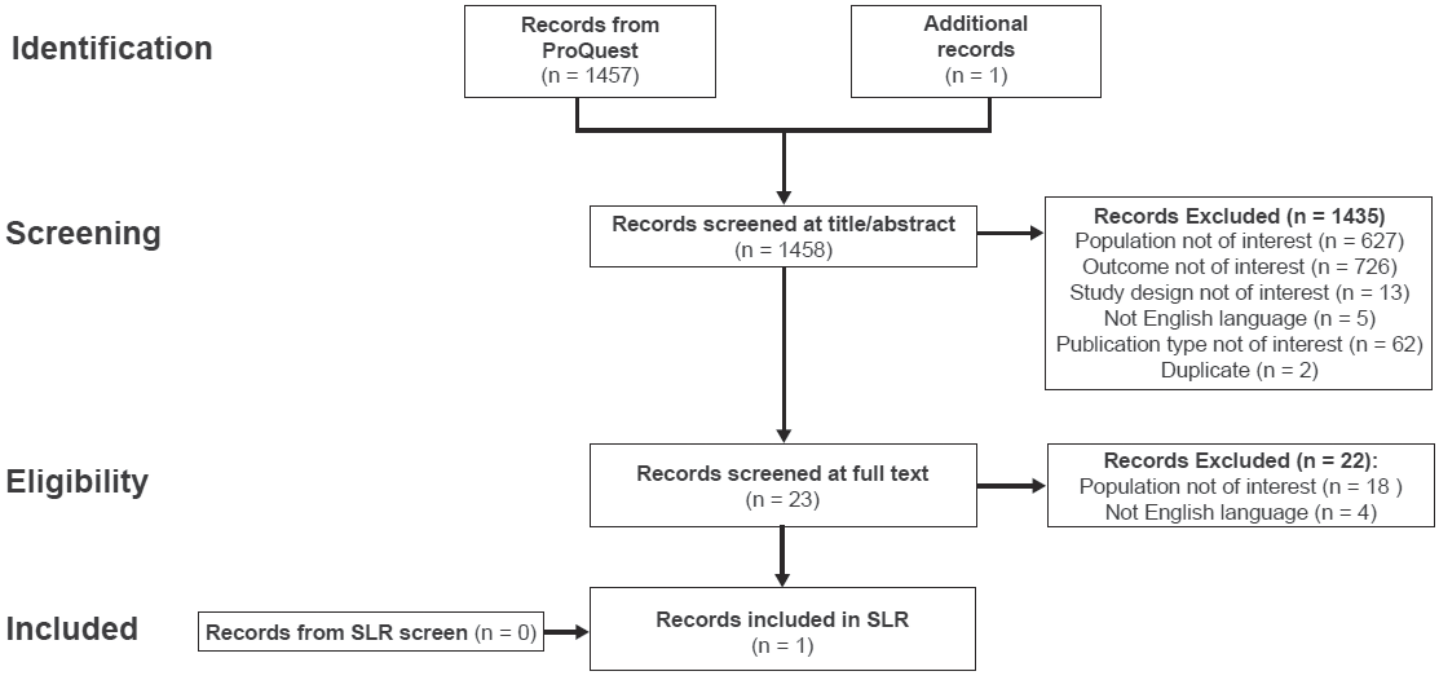


Figure S2. Flowchart of the Article Selection Process for the Humanistic and Economic Burden.
SLR, systematic literature review.
*From authors' personal library.
†From eligible SLRs of cohort studies.

Appendix S1. Search Strategy

#	Description	Search String	Hits
S1	Disease state	(EMB.EXACT.EXPLODE("newborn hemolytic disease") OR MESH.EXACT.EXPLODE("Erythroblastosis, Fetal")) AND (TI,AB(HDFN OR ((hemolytic OR haemolytic) NEAR/3 (fetus OR newborn OR foetus)))) OR TI,AB("hemolytic disease of the fetus and newborn" OR "haemolytic disease of the fetus and newborn" OR "hemolytic disease of the foetus and newborn" OR "haemolytic disease of the foetus and newborn" OR "hemolytic disease of the newborn" OR "haemolytic disease of the newborn" OR HDFN) OR EMB.EXACT.EXPLODE("alloimmunization") OR ("maternal alloimmunization") OR ("red cell alloimmunization")	12,499[a]
S2	Intervention/treatment setting	EMB.EXACT("intrauterine blood transfusion") OR EMB.EXACT("fetal therapy") OR EMB.EXACT.EXPLODE("neonatal intensive care unit") OR MESH.EXACT.EXPLODE("Fetal Therapies") OR MESH.EXACT("Intensive Care Units, Neonatal") OR EMB.EXACT("immunoglobulin") OR MESH.EXACT("Immunoglobulins, Intravenous") OR EMB.EXACT.EXPLODE("plasma exchange") OR MESH.EXACT.EXPLODE("Plasma Exchange") OR ("therapeutic plasma exchange") OR ("neonatal transfusion") OR EMB.EXACT.EXPLODE("exchange blood transfusion") OR ("neonatal exchange transfusion")	233,001[a]
S3	Clinical outcomes	MESH.EXACT("Mortality") OR EMB.EXACT("mortality") OR EMB.EXACT("infant mortality") OR MESH.EXACT.EXPLODE("Infant Mortality") OR MESH.EXACT(Morbidity) OR EMB.EXACT(morbidity) OR MESH.EXACT.EXPLODE("Anemia, Neonatal") OR MESH.EXACT.EXPLODE("Hyperbilirubinemia") OR MESH.EXACT("Fetal Blood") OR MESH.EXACT("Perinatal Death") OR MESH.EXACT("Fetal Death") OR EMB.EXACT("newborn jaundice") OR EMB.EXACT("neonatal hyperbilirubinemia") OR EMB.EXACT("anemia") OR EMB.EXACT("hyperbilirubinemia") OR EMB.EXACT.EXPLODE("perinatal morbidity") OR EMB.EXACT("newborn death") OR EMB.EXACT("fetus death") OR EMB.EXACT("rhesus incompatibility") OR (late NEAR/3 anemia) OR TI,AB(complication*) OR TI,AB(adverse /NEAR1 (effect* OR reaction* OR event*)) OR MESH.EXACT("Hydrops Fetalis") OR EMB.EXACT.EXPLODE("edema") OR EMB.EXACT.EXPLODE("fetus hydrops") OR MESH.EXACT.EXPLODE("Premature Birth") OR EMB.EXACT.EXPLODE("prematurity") OR (emergency NEAR/2 childbirth OR delivery OR cesarean section) OR MESH.EXACT.EXPLODE("Respiration, Artificial") OR EMB.EXACT("artificial ventilation") OR MESH.EXACT("Heart Failure") OR EMB.EXACT("heart failure")	6,724,924[a]
S4	Combined disease state and outcomes of interest	S1 AND S2	1,903[b]
S5	Combined disease state and outcomes of interest	S1 AND S3	3,619[b]
S6	Total hits	S4 OR S5	5,069[a]
S7	Final hits published between January 1, 2005, and March 10, 2021	S6 AND publication date (>2004)	2,518[b]

[a]Duplicates were removed from the search but included in the result count.
[b]Duplicates were removed from the search and from the result count.

Appendix S2. Search Strategy for the Humanistic and Economic Burden

#	Description	Search String	Hits
S1	Disease state	(EMB.EXACT.EXPLODE("newborn hemolytic disease") OR MESH.EXACT.EXPLODE("Erythroblastosis, Fetal")) AND (TI,AB(HDFN OR ((hemolytic OR haemolytic) NEAR/3 (fetus OR newborn OR foetus)))) OR TI,AB("hemolytic disease of the fetus and newborn" OR "haemolytic disease of the fetus and newborn" OR "hemolytic disease of the foetus and newborn" OR "haemolytic disease of the foetus and newborn" OR "hemolytic disease of the newborn" OR "haemolytic disease of the newborn" OR HDFN) OR EMB. EXACT.EXPLODE("alloimmunization") OR ("maternal alloimmunization") OR ("red cell alloimmunization")	12499*
S2	Intervention/ Tx setting	EMB.EXACT("intrauterine blood transfusion") OR EMB. EXACT("fetal therapy") OR EMB.EXACT.EXPLODE("neonatal intensive care unit") OR MESH.EXACT.EXPLODE("Fetal Therapies") OR MESH.EXACT("Intensive Care Units, Neonatal") OR EMB.EXACT("immunoglobulin") OR MESH.EXACT("Immunoglobulins, Intravenous") OR EMB. EXACT.EXPLODE("plasma exchange") OR MESH.EXACT. EXPLODE("Plasma Exchange") OR ("therapeutic plasma exchange") OR ("neonatal transfusion") OR EMB.EXACT. EXPLODE("exchange blood transfusion") OR ("neonatal exchange transfusion")	233001*
S3	Humanistic Outcomes	EMB.EXACT("International Classification of Functioning, Disability and Health" OR "quality of life" OR "short form 36" OR "patient reported outcome" OR "patient preference" OR "questionnaire" OR "quality adjusted life years") OR EMB.EXACT.EXPLODE("health status indicator") OR MESH.EXACT("International Classification of Functioning, Disability and Health" OR "Quality of Life" OR "Value of Life" OR "Patient Reported Outcome Measures" OR "Patient Preference" OR Questionnaires OR "Quality-Adjusted Life Years") OR MESH.EXACT.EXPLODE("Health Status Indicators") OR TI,AB(burden OR (impact NEAR/3 (caregiver OR family OR families OR society OR societal or patient OR person)) OR "unmet need" OR "disability adjusted" OR DALY* OR "dartmouth coop" OR "Duke health profile" OR EQ OR (EURO NEAR/2 (QUAL OR QOL)) OR ((daily OR day) NEAR/3 activit*) OR "functional status" OR FSQ OR (function* NEAR/5 (reduc* OR impair* OR decrease* OR impact*)) OR "quality of life" OR QOL OR hrqol OR hrql OR hql OR hqol OR "hr qol" OR "h qol" OR "life quality" OR (health NEAR/3 (status OR indicator*)) OR "Nottingham health" OR NHP OR PQOL OR "perceived quality" OR QLS OR "quality of life scale" OR wellbeing OR "well being" OR QWB OR rosser OR SF OR "short form" OR "shortform*" OR "sickness impact" OR SIP OR "patient reported" OR "self reported" OR QALY* OR QALD* OR QALE* OR QTIME* OR (utilit* NEAR/3 (valu* OR measur* OR health OR life OR estimat* OR elicit* OR disease OR score* OR weight OR instrument OR instruments OR index)) OR "quality adjusted" OR "life year*" OR "health year*" OR disutilit* OR "willingness to pay" OR WTP OR (preference* NEAR/3 (valu* OR measur* OR health OR life OR estimat* OR elicit* OR disease OR score* OR instrument OR instruments OR index)) OR "healthy utility index" OR hui OR "standard gamble" OR "time trade off" OR "time tradeoff" OR TTO OR "health assessment questionnaire" OR "health assessment questionnaires" OR HAQ OR (humanistic AND burden))	4018894*

#	Description	Search String	Hits
S4	Economic Outcomes	MESH.EXACT("Cost of Illness" OR "Economics, Hospital" OR "Economics, Nursing" OR "Economics, Pharmaceutical" OR "Fees and Charges" OR "Economics, Dental" OR "Employer Health Costs" OR "Efficiency" OR "Presenteeism" OR "Absenteeism") OR MESH.EXACT.EXPLODE("Health Care Costs" OR "Health Expenditures" OR "Economics, Medical" OR "Salaries and Fringe Benefits") OR EMB.EXACT("cost of illness" OR "drug cost" OR "productivity" OR "medical leave" OR "presenteeism" OR "absenteeism") OR EMB.EXACT.EXPLODE("health care cost" OR "salary and fringe benefit") OR TI,AB(presenteeism OR absenteeism OR (cost* NEAR/3 (medical OR direct OR indirect OR drug OR pharmaceutical OR hospital OR emergency OR outpatient OR inpatient OR ambulatory OR "primary care" OR practitioner OR device OR informal OR economic OR societal OR intangible OR caregiver OR physician OR specialist OR healthcare OR "health care" OR annual* OR clinic)) OR ((burden OR impact) NEAR/3 (cost OR costs OR economic OR economics OR caregiver OR caregivers OR family OR families OR society OR societal OR employee OR employer)) OR (los* AND work AND day*) OR "sick day" OR "sick leave" OR "sickness absence" OR "work absence" OR "work incapacity" OR "work leave" OR "disability absence" OR ((resource OR healthcare OR "health care") NEAR/5 (use OR utilization OR utilisation)) OR ((visit* OR admission* OR readmission* OR stay* OR day*) NEAR/3 (physician OR emergency OR specialist OR outpatient OR inpatient OR "primary care" OR practitioner OR hospital OR clinic)) OR hospitalization OR hospitalisation OR "length of stay" OR "LOS" OR (burden AND financial))	2205185*
S5	Combined disease state and outcomes of interest	S1 AND S2	1903°
S6		S1 AND S3	329°
S7		S1 AND S4	344°
S8	Total Hits	S5 OR S6 OR S7	2387°
S9	Final hits published on or after 2005	S8 and PD(>2004)	1441°

* Duplicates are removed from the search but included in the result count.
° Duplicates are removed from the search and from the result count.