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Transfusion of fresher versus older red blood cells for all conditions

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ABSTRACT

Background

Red blood cell transfusion is a common treatment for anaemia in many clinical conditions. One current concern is uncertainty as to the clinical consequences (notably efficacy and safety) of transfusing red blood cell units that have been stored for different durations of time before a transfusion. If evidence from randomised controlled trials were to indicate that clinical outcomes are affected by storage age, the implications for inventory management and clinical practice would be significant.

Objectives

To assess the effects of using fresher versus older red blood cells in people requiring a red blood cell transfusion.

Search methods

We ran the search on 29th September 2014. We searched the Cochrane Central Register of Controlled Trials (CENTRAL), MEDLINE (OvidSP), Embase (OvidSP), CINAHL (EBSCO), PubMed (for e-publications), three other databases and trial registers.

Selection criteria

We included randomised controlled trials comparing fresher red blood cell transfusion versus active transfusion of older red blood cells, and comparing fresher red blood cell transfusion versus current standard practice. All definitions of 'fresher' and 'older'/standard practice' red blood cells were included.

Data collection and analysis

Two review authors independently assessed trial quality and extracted from the trial report data on adverse red blood cell transfusion reactions, when reported.

Main results

We included 16 trials (1864 participants) in the review. Eight trials (279 participants) compared transfusion of fresher red blood cells versus transfusion of older stored red blood cells ('fresher' vs 'older'). Eight trials (1585 participants) compared the transfusion of fresher red blood cells versus current standard practice ('fresher' vs 'standard practice'). Five trials enrolled neonates, one trial enrolled children and 12 trials enrolled adults. Overall sample sizes were small: only two trials randomly assigned more than 100 participants.

We performed no meta-analyses for a variety of reasons: no uniform definition of 'fresher' or 'older' red blood cell storage; overlap in the distribution of the age of red blood cells; and heterogeneity in measurements and reporting of outcomes of interest to this review. We tabulated and reported results by individual trial. Overall risk of bias was low or unclear, with four incidences of high risk of bias: in allocation concealment (three trials) and in incomplete outcome data (one trial).

No trial measured all of the outcomes of interest in this review. Four trials comparing 'fresher' with 'older' red blood cells reported the primary outcome: mortality within seven days (one study; 74 participants) and at 30 days (three trials; 62 participants). Six trials comparing 'fresher' with 'standard practice' red blood cells reported the primary outcome: mortality within seven days (three studies; 159 participants) and at 30 days (three trials; 1018 participants). All 10 trials reported no clear differences in mortality at either time point between intervention arms.

Three trials comparing 'fresher' with 'standard practice' red blood cells reported red blood cell transfusion-associated adverse events. No adverse reactions were reported in two trials, and one incidence of cytomegalovirus (CMV) infection was described in the 'standard practice' arm in one trial.

Overall the trials reported no clear difference between either of the intervention comparisons in long-term mortality (three trials; 478 participants); clinically accepted measures of multiple organ dysfunction (two trials; 399 participants); incidence of in-hospital infection (two trials; 429 participants); duration of mechanical ventilation (three trials; 95 participants); and number of participants requiring respiratory organ support (five trials; 528 participants) or renal support (one trial; 57 participants). The outcome 'physiological markers of oxygen consumption or alterations in microcirculation' was reported by 11 studies, but the measures used were highly varied, and no formal statistical analysis was undertaken.

Authors' conclusions

Several factors precluded firm conclusions about the clinical outcomes of transfusing red blood cell units that have been stored for different periods of time before transfusion, including differences in clinical population and setting, diversity in the interventions used, methodological limitations and differences in how outcomes were measured and reported.

No clear differences in the primary outcome - death - were noted between 'fresher' and 'older' or 'standard practice' red blood cells in trials that reported this outcome. Findings of a large number of ongoing trials will be incorporated into this review when they are published.

Updates of this review will explore the degree of overlap in trials between 'fresher', 'older' and 'standard practice' storage ages of red blood cells and will consider whether the size of any observed effects is dependent on recipient factors such as clinical background, patient age or clinical presentation.

PLAIN LANGUAGE SUMMARY

Transfusion of fresher versus older red blood cells for all conditions

After donation, red blood cells can be stored for up to 42 days before they are transfused to anyone needing a red blood cell transfusion. One current concern is uncertainty as to the clinical consequences (notably efficacy and safety) of transfusing red blood cell units that have been stored for different durations before a transfusion. We sought to explore this uncertainty in a systematic review comparing 'fresher' versus 'older' or 'standard practice' red blood cells given to anyone needing a red blood cell transfusion.

Sixteen trials (1864 participants) were identified. Eight trials (279 participants) compared transfusion of 'fresher' versus 'older' red blood cells. Eight trials (1585 participants) compared transfusion of 'fresher' versus 'current standard practice' red blood cells. Within these groups, trials were divided into those involving very low birth weight (VLBW) infants and those involving adult participants, because of the different physiology of these participants.

An important finding was wide differences in the definitions of 'fresher', 'older', and 'standard practice' red blood cells across trials. Large overlap between age ranges prevented data analysis across trials. Instead, outcome results are reported descriptively and are presented in tables.

Outcomes of interest in the review included death (whilst in-hospital, up to 30 days after transfusion and longer term), length of hospital stay, incidence of new infection and measures that assessed how well the body was functioning. Outcomes that assessed how well the body was functioning were measured in many different ways by trial authors. No single trial measured all of the outcomes of interest in this review. Adverse events following a red blood cell transfusion were measured in three trials comparing 'fresher' versus 'standard practice' red blood cells. Overall, in all trials, no clear difference was observed between the 'fresher' red blood cell transfusion and the 'older' or 'standard practice' red blood cell transfusion for any outcome, including the review's primary outcome of death.

Review authors were unable to draw firm conclusions on the clinical consequences of transfusing red blood cells that have been stored for different durations before a transfusion, because each study measured duration of storage differently. Further research is needed to explore the extent of overlap in trials between 'fresher', 'older' and 'standard practice' storage groups.

BACKGROUND

blood cell transfusion is the only viable treatment modality capable of restoring tissue oxygenation.

Description of the condition

The haemoglobin (Hb) contained within red blood cells is essential for oxygen transportation. Anaemia, defined by the World Health Organization (WHO) as a Hb concentration < 13 g/dL in men and < 12 g/dL in women, describes a clinical state in which this physiological process is disturbed and tissue hypoxia may occur (Beutler 2006). Anaemia has no single cause; rather it is the consequence of a variety of aetiological factors. In high-income countries, the overall prevalence of anaemia is estimated to be 10% (McLean 2009). However, this figure varies significantly with demographic profiles and patterns of co-morbid diagnoses (McLean 2009; Tettamanti 2010). Children and older adults are most commonly affected by anaemia. For example, almost 90% of preterm infants with birth weight < 1.0 kg are anaemic (Martin 2010). In later life, rates rise again, largely because of the increasing incidence of co-morbid diagnoses.

The aetiology of anaemia can be broadly divided into disease processes that impair red blood cell production and those in which the life span or distribution of red blood cells is altered. In the former group, disorders such as acquired and iatrogenic marrow dysfunction and nutritional deficiency and cytokine-driven processes such as anaemia of chronic disease are commonplace. In the latter group, examples include disease processes such as pathological bleeding and immune haemolysis.

When possible, reversing the primary cause of anaemia remains the treatment of choice; however this cannot always be achieved. Furthermore, when severe anaemia results in life-threatening organ dysfunction, rapid correction is required. In these instances, red

Description of the intervention

Red blood cell transfusion

Red blood cell transfusion has been a commonplace treatment for anaemia for well over three decades (Alter 2008). It is a very widely practiced intervention. In the UK around two million red blood cell units are issued for transfusion annually (Stainsby 2006). This equates to transfusion of approximately 45 units per 1000 population per year - a figure not dissimilar to the rest of the developed world (Cobain 2007). For such a ubiquitous intervention, it is surprising that a recent systematic overview concluded that rigorous clinical trial data are lacking to support the benefits of many currently employed transfusion practices (Wilkinson 2011). As a generalisation, evidence obtained from randomised controlled trials (Carson 2012) indicates little or no benefit from red cell transfusion at higher haemoglobin concentration thresholds (commonly termed 'liberal' policies for red cell transfusion). Red cell transfusions are also associated with some well-described risks (Stainsby 2006). As biological products, hazards such as bacterial contamination and allergic reactions are well recognised. Other risks posed by red blood cell transfusion, whilst less tangible, are potentially far more common. Key amongst these is the long debated risk posed by red blood cells with a prolonged storage age (see below) (Schrier 1979). Therefore, practice guidelines for red cell transfusion in many clinical settings now promote more restrictive policies for red cell transfusion (Carson 2013). But despite this, red cell transfusion remains a very common intervention. For example, as

many as 60% of patients admitted to critical care units develop anaemia (Vincent 2002; Corwin 2004), but only 10% to 15% of patients have a history of chronic anaemia before admission to the intensive care unit (ICU). Unless modified by red cell transfusion, haemoglobin values typically are decreased by about 0.5 g/dL/d during critical illness for reasons including illness, co-morbidities, bleeding and phlebotomy. As a result, between 20% and 50% of critically ill patients receive a red cell transfusion, especially those with multiple organ failure. About 8% to 10% of the UK blood supply is transfused to patients in an ICU.

Red blood cell units and their storage

A major concern about red cell transfusion is uncertainty regarding the clinical consequences of transfusing red cell units that have been stored for longer periods before transfusion. The hypothesis of harm related to transfusion of a product stored longer was reignited by the authors of the Transfusion Requirements In Critical Care (TRICC) trial (Hebert 1999). This landmark randomised controlled trial compared liberal and restrictive transfusion practices in critically ill people. Investigators showed that restricting transfusions to maintain Hb concentration at 7 to 9 g/dL was safe and in some subgroups of patients superior to more liberal red blood cell use. Crucially, study authors suggested that the common practice of storing red blood cell units for prolonged periods may be one factor that explains the unexpected adverse effects of liberal transfusion.

This suggestion is biologically plausible in view of the growing body of evidence demonstrating changes in many cellular and physiological properties of red cells. These *in vitro* changes, which occur during red blood cell storage, are commonly known as the 'storage lesion' (D'Alessandro 2010; Glynn 2010). The storage lesion includes biochemical, metabolic and mechanical changes to the red cell, all of which may impair oxygen delivery. The term also encompasses changes that occur in the red cell storage medium, which theoretically could mediate inflammatory or oxidative tissue damage (Sharifi 2000; Kucukakin 2011). The most commonly described biochemical and metabolic components of the storage lesion are impaired nitric oxide metabolism (Stapley 2012), depletion of cellular 2,3-diphosphoglycerate (Vora 1989) and dysfunction of the membrane sodium-potassium pump (D'Alessandro 2010). Nitric oxide depletion induces vasoconstriction, in turn impairing blood flow and oxygenation (Stapley 2012). Altered 2,3-diphosphoglycerate reduces the oxygen affinity of haemoglobin (Sohmer 1979). Membrane sodium-potassium pump dysfunction results in harmful potassium leakage from the red cell into extracellular fluids (Hess 2010). Mechanical changes to the red cell membrane impair fluidity and red cell flow (Hess 2010). Similar to nitric oxide depletion, this may reduce transit of the red cell through the microscopic vasculature of organs such as lung and kidney (Roback 2011a). Again such changes may impair oxygen uptake and delivery. Storage medium changes include the generation of

inflammatory mediators such as soluble CD40 ligand, interleukin-6 (IL-6) and interleukin-8 (IL-8) (Khan 2006; Kucukakin 2011). Potential oxidative damage may also arise from superoxide generation in the storage media (Kucukakin 2011).

Extended red blood cell storage, as described above, is fundamental to effective blood stock management. In the UK, as a consequence of stock rotation processes, the average age of a red blood cell unit at the time of transfusion is 18 to 21 days (NHSBT 2012). Such a figure is very similar to that found throughout Europe and North America (Bennett-Guerrero 2009; Lacroix 2011; Heddle 2012). Changes in the storage lesion, as described above, may be well established by this time. It is biologically plausible, therefore, that critically ill patients may currently be denied the benefits of 'fresher' red cells, and exposed to the additional clinical risks posed by older red blood cell units. Limited clinical data would support this notion. Cohort studies have described associations between red blood cell storage age and a wide range of clinically important adverse outcomes (including infection, organ failure, increased hospital stay and death) (Vamvakas 1999; Mynster 2000; Leal-Noval 2003; Basran 2006; Koch 2008). However, these effects are not universally described (Vamvakas 2000; van de Watering 2006); although this important message is provided in the literature, many study authors point to the presence of significant confounding factors in the evidence (Steiner 2009). In particular, the strong linkage between the total volume transfused (which itself is strongly associated with the presence of co-morbidities, severity of illness and worse prognosis) and the average age of red blood cell units issued makes inferring causality very difficult (Vamvakas 2010).

How the intervention might work

The goal of administering red cell transfusion is improved tissue oxygenation achieved by increased red cell mass. It is commonly presumed that lower haemoglobin concentrations represent an accurate measure of diminished oxygen-carrying capacity, which can be corrected in part by red cell transfusion. Processing methods for red blood cell collection and storage for transfusion have been studied for many years (Alter 2008). After blood collection, whole blood is centrifuged, plasma is depleted and red cells are re-suspended in an optimal additive solution for storage within specially designed bags. This process, in conjunction with effective refrigeration, has allowed the duration of red cell concentrate storage to be significantly extended (D'Alessandro 2010). Many countries routinely store red cell concentrates for up to 42 days. This period is defined by the arbitrary requirement that, after storage, more than 75% of red blood cells should survive in the recipient's circulation at 24 hours (Roback 2011b). An extended shelf life facilitates stock management and is fundamental to effective blood banking. It is standard practice amongst both blood providers and blood banks to issue the oldest stock first in preference to newer stock (Stanger 2012). This is necessary to ensure that the demand

for this unique product can be met whilst wastage of what is a precious and financially costly resource is minimised (Stanger 2012).

Why it is important to do this review

This review is required to coalesce and appraise wider randomised evidence on the impact of storage age on the efficacy and safety of red cell transfusion. Although numerous reviews already published have addressed this question (including Lelubre 2009; Zimrin 2009; Vamvakas 2010), these reviews are based on observational study data and do not include recently published and ongoing trials in this area (Steiner 2010; Lacroix 2011; Fergusson 2012). Furthermore, mathematical modelling suggests, individually, that the ongoing Steiner 2010 and Lacroix 2011 studies may lack enough power to prove conclusive (Pereira 2013). Therefore, quantitative analysis will be needed to pool results across all relevant randomised studies.

If studies were to indicate that clinical outcomes are affected by storage age, the implications for inventory management and clinical practice would be significant. Clinicians would rightly expect fresher, safer and more efficacious red blood cell units. Implementation of such a strategy would be likely to place considerable additional strain on blood providers and blood banks. It may also result in increased wastage, higher financial costs and potentially threatened blood supplies (Glynn 2010). The urgent need to reconcile this issue, because of its potential harms to patients and its massive logistical implications, is keenly felt by clinicians, blood services and policy makers alike. Unfortunately, as with so many questions about the efficacy, safety and utility of red blood cell transfusion, the evidence needed to settle this debate is currently not clear.

OBJECTIVES

To assess the effects of using fresher versus older red blood cells in people requiring a red blood cell transfusion.

METHODS

Criteria for considering studies for this review

Types of studies

Randomised controlled trials (RCTs) comparing transfusion of fresher red blood cells with that of older red blood cells.

Types of participants

People of any age (neonates, children and adults) requiring transfusion for investigator-diagnosed and -defined anaemia of any aetiology.

Types of interventions

No consensus has been reached about what defines 'fresher' or 'older' red blood cell units. Arbitrarily defining 'fresher' and 'older' red blood cell units for the purposes of this review is scientifically unsound, invites legitimate criticism of the clinical validity of the review's results and risks exclusion of a large proportion of available data. Consequently, we included all definitions of fresher and older red cells. Studies comparing the following groups were eligible for inclusion.

- Fresher red blood cell transfusion versus active transfusion of older red blood cells.
- Fresher red blood cell transfusion versus current standard practice. Here, the age of the older red blood cells is dictated by standard inventory management practice.
- Fresher red blood cell transfusion (given as a historical standard of practice in neonatal patients) versus active transfusion of older red blood cells.

Types of outcome measures

Primary outcomes

- Mortality measured at two time points: immediate (occurring within seven days in hospital) and short term (up to 30 days).

Secondary outcomes

- Long-term mortality (analysed at appropriately similar time points (e.g. using medium- and long-term mortality as appropriate)).
- Clinically accepted measures of multiple organ dysfunction (e.g. Multiple Organ Dysfunction Score (MODS), Sequential Organ Failure Assessment (SOFA) score) and numbers of dysfunctional organs reported per participant.
- Incidence of any in-hospital infection.
- Duration of respiratory support (invasive and non-invasive ventilation), haemodynamic support (inotropic) and renal support (haemofiltration).
- Length of hospital and ICU stay.
- Adverse transfusion reactions.
- Physiological markers of oxygen consumption or alterations in microcirculation, as measured by included, eligible trials.
- Assessment of economic or blood stock inventory outcomes, as measured by included, eligible trials.

We were interested in data addressing any of the above outcomes and at any reported time points.

Information size for the primary outcome of the review was taken from the calculated sample size for the Age of Blood Evaluation (ABLE) randomised controlled trial ([Lacroix 2011](#)). The clinical population for ABLE consisted of adult critical care patients, and for the trial, a total sample size of 2510 participants was defined to assess an absolute reduction in mortality by 5% from a baseline mortality of 25%.

Search methods for identification of studies

In order to reduce publication and retrieval bias we did not restrict our search by language, date or publication status.

Electronic searches

We searched the following electronic databases:

- Cochrane Central Register of Controlled Trials (CENTRAL) (2014, Issue 8).
- MEDLINE (OvidSP) (1948 to 29 September 2014).
- Embase (OvidSP) (1974 to 29 September 2014).
- Cumulative Index to Nursing and Allied Health Literature (CINAHL) (EBSCOHost) (1982 to 29 September 2014).
- PubMed (current e-publications only, 29 September 2014).
- [Transfusion Evidence Library](#) (1980 to 29 September 2014).
- Latin American Caribbean Health Sciences Literature (LILACS) (1982 to 29 September 2014).
- Institute for Scientific Information (ISI) Web of Science: Conference Proceedings Citation Index-Science (CPCI-S) (1990 to 29 September 2014).

We searched the following databases for ongoing trials (all years):

- International Standard Randomised Controlled Trial Number Register ([ISRCTN Register](#)) (29 September 2014).
- EU Clinical Trials Register (29 September 2014).
- [ClinicalTrials.gov](#) (29 September 2014).
- [World Health Organization International Clinical Trials Registry Search Platform](#) (ICTRP) (29 September 2014).
- University Hospital Medical Information Network Clinical Trials Registry (UMIN-CTR) Japanese Clinical Trials Registry and Hong Kong Clinical Trials Registry (29 September 2014).

All search strategies are reported in [Appendix 1](#). We combined searches in MEDLINE, Embase and CINAHL with adaptations of the Cochrane RCT search filter, as detailed in Chapter 6 of the *Cochrane Handbook for Systematic Reviews of Interventions* ([Lefebvre 2011](#)).

Searching other resources

We checked references of all identified trials, relevant review articles and current treatment guidelines for further literature. We limited these searches to 'first-generation' reference lists.

Data collection and analysis

Selection of studies

One review author (CD) screened all titles and abstracts of papers identified through electronic searches; only clearly irrelevant references were be excluded at this stage. We subsequently retrieved full texts for the remaining references; three review authors (SB, SS and RG (see [Acknowledgements](#))) independently assessed them for inclusion using a study-specific eligibility form. When necessary, disagreements were resolved by an adjudicator (MT).

Data extraction and management

Four review authors (RG, SB, KW and SS) independently undertook data extraction using a piloted study-specific data extraction form. Disagreements were resolved by consensus between the review authors. One review author (SB) entered the data into the statistical software of The Cochrane Collaboration, [Review Manager 2013](#).

We undertook data extraction in accordance with guidance detailed in the *Cochrane Handbook for Systematic Reviews of Interventions* ([Higgins 2011b](#)). With regard to the intervention, we expected heterogeneity within the definition of fresh versus older red blood cell units. We anticipated potential issues with protocol non-adherence. Such problems influence the success with which the experimental arms were exposed to distinct, non-overlapping ages of red blood cell units. Consequently, we specifically extracted intervention data upon:

- age of red blood cell units transfused in the intervention group and in the control group; and
- total volume of red blood cell exposure in both intervention groups.

As the result of possible heterogeneity of red cell product specifications, we specifically extracted data on the use of:

- irradiated blood;
- whole blood; and
- red cell leucoreduction.

To minimise biases that could arise in the 'subcategorisation' of studies by their definition of 'fresher' and 'older' red blood cells, review authors undertaking data extraction (RG, SB, KW and SS) were not involved in categorising studies by their definition of 'fresher' and 'older' red cells for interpretation and analysis of outcome data. MT undertook this categorisation of studies.

Assessment of risk of bias in included studies

Four review authors (RG, SB, KW and SS) independently assessed risk of bias for each trial using the criteria outlined in the *Cochrane Handbook for Systematic Reviews of Interventions* (Higgins 2011c). We resolved disagreements by discussion. For each of the included trials, we assessed risk of bias (as low, high or unclear) for the following domains.

- Generation of random sequence (selection bias).
- Concealment of treatment allocation (selection bias).
- Blinding of participants and personnel (person(s) delivering treatment) to treatment allocation (performance bias).
- Blinding of outcome assessors to treatment allocation (detection bias).
- Completeness of the outcome data (including checks for possible attrition bias through withdrawals, loss to follow-up and protocol violations).
- Selective reporting of outcomes (reporting bias).
- Other sources of bias (other bias). An assessment will be made as to whether each trial was free of problems, not identified by those listed above, that could put it at risk of bias.

In this initial version of the review, we reported findings from the assessment of risk of bias as a narrative description.

Measures of treatment effect

We calculated risk ratios (RRs) for dichotomous outcomes. We expressed treatment effects for continuous data outcomes as mean differences (MDs) and used a 95% confidence interval (CI) in all instances.

Regarding the incidence of new hospital infections, we were going to estimate incidence ratio as the ratio of new observed cases over expected numbers of cases for exposed patients. However homogenous data are insufficient to permit meta-analysis; hence we did not calculate incidence rates. Instead we reported incidences of and calculated risk ratios for different infections within each study.

We reported but did not formally analyse all reported data on length of hospital stay, length of intensive care stay and duration of mechanical ventilation. Trials presented data on length of hospital stay and length of intensive care unit stay as median (with interquartile range) values, which are appropriate and robust ways to report these data, as these outcomes usually are not normally distributed; or as means (with standard deviations), which is usually an inappropriate way to report these particular outcomes (as length of stay more often than not is a skewed distribution). We have presented data for duration of mechanical ventilation as means (with standard deviations) and as numbers of participants requiring mechanical ventilation. When presented as means (with standard deviations), the same principles apply as for length of stay (i.e. that this is usually an inappropriate way to report an outcome of duration because more often than not it is skewed). We tabu-

lated data for these outcomes and have reported them narratively within the text.

In the absence of appropriate skills in our facility to fully analyse economic and blood stock inventory outcomes at this time, we have presented these data in a narrative format. One review author (SB) entered all data into [Review Manager 2013](#); thereafter a second review author (MT) checked the data.

Unit of analysis issues

No unit of analysis issues arose while this review was performed; we identified no eligible cross-over or cluster-randomised trials. The level of measurement was the patient.

Dealing with missing data

We had no overall concern regarding missing data; hence we did not contact the author(s) of individual trial(s). When denominator data allowed, we converted reported percentages into actual numbers of participants for two studies, so we could calculate outcome risk ratios for these studies. We noted levels of attrition for all included trials.

Assessment of heterogeneity

We did not undertake assessments of statistical heterogeneity within this systematic review, as no pooled analysis was performed. We explored instead clinical heterogeneity within studies, and this approach has precluded any formal pooling of data. The clinical heterogeneity that we identified included age of the participants (neonates vs adult participants); underlying clinical diagnosis of participants across trials; what constituted the red blood cell product used for the intervention (whereby we noted a range of age of red blood cells within each of the intervention arms, and in many instances, the range of the two intervention groups overlapped, as can be seen in [Table 1](#)) and heterogeneity in the techniques used to measure outcomes (see [Table 2](#), [Table 3](#) and [Table 4](#)). We found no clinical heterogeneity due to trial design or risk of bias.

Assessment of reporting biases

We did not assess publication bias using funnel plots, as fewer than 10 studies offered data.

Data synthesis

Comparisons made within the primary studies were diverse because of the absence of a uniform definition of fresher and older red blood cells. After data extraction, we assessed whether included studies were suitable, in terms of the definitions of fresher and older red blood cells employed, for inclusion in a single meta-analysis. Unfortunately, we found them to be not suitable as the result of differences in what constituted the red blood cell products

used for the intervention (whereby we noted a range of age of red blood cells within each of the intervention arms, as can be seen in [Table 1](#)) and other clinical and outcome measurement differences between trials (see also [Assessment of heterogeneity](#)).

Subgroup analysis and investigation of heterogeneity

If data had been sufficient, we would have undertaken subgroup analyses based on age groups (e.g. neonates) and transfusion indications (long-term transfusion dependence vs support during an acute illness) to look for significant differences in treatment effects. Although meta-analysis was not possible, we have reported our observations as a narrative, and have provided reporting by age group.

Sensitivity analysis

If data had been sufficient, we would have undertaken sensitivity analyses to explore aspects of methodology. These would have been done to explore the effects of removing trials at high or unclear risk of the following domains of bias: selection bias (reflecting lack of confirmation of random sequence generation and allocation concealment); detection bias (reflecting lack of assessor blinding); and attrition bias (as from high levels of missing data).

RESULTS

Description of studies

See [Characteristics of included studies](#) and [Characteristics of excluded studies](#).

Results of the search

Through the search, we identified 4089 references, of which we excluded 2323 in the first screening because they were duplicates or were clearly irrelevant to the scope of this review. Of the remaining 1767 references, which we screened by title and abstract, we excluded 1721 references that did not meet the inclusion criteria for participants, interventions or both, or were not randomised controlled trials ($n = 156$).

We obtained the full text of the remaining 44 references. Of these 44 references, we deemed 16 trials (reported in 27 references) to be eligible for inclusion ([Liu 1994](#); [Strauss 1996](#); [Strauss 2000](#); [Schulman 2002](#); [Walsh 2004](#); [Fernandes 2005](#); [Hebert 2005](#); [Bennett-Guerrero 2009](#) [1]; [Bennett-Guerrero 2009](#) [2]; [Aubron 2012](#); [Fergusson 2012](#); [Heddle 2012](#); [Kor 2012](#); [Dhabangi 2013](#); [Neuman 2013](#); [Yuruk 2013](#)); three trials (reported in five references) are trials awaiting assessment ([Steiner 2010](#); [Lacroix 2011](#); [NCT00458783](#)), and six are ongoing trials ([Kaukonen 2013](#); [NCT01586923](#); [NCT01976234](#); [NCT01977547](#); [NCT02050230](#); [NCT02087514](#)). We excluded five studies (reported in six full-text papers) because they did not meet the eligibility criteria of this review ([Wasser 1989](#); [Eshleman 1994](#); [Strauss 1994](#); [Seitelbach 2011](#); [Lebiedz 2012](#)) (see [Characteristics of excluded studies](#)). We have reported full details in the PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) flow diagram ([Figure 1](#)).

Figure 1. Study flow diagram.

Included studies

Participants in 15 trials were in-hospital patients (Liu 1994; Strauss 1996; Strauss 2000; Schulman 2002; Walsh 2004; Fernandes 2005; Hebert 2005; Bennett-Guerrero 2009 [1]; Bennett-Guerrero 2009 [2]; Aubron 2012; Fergusson 2012; Heddle 2012; Kor 2012; Dhabangi 2013; Neuman 2013). Participants in one trial were outpatients of the haematology department (Yuruk 2013).

One manuscript reported data on two separate trials, which will be reported throughout as Bennett-Guerrero 2009 [1] and Bennett-Guerrero 2009 [2]. One trial (Schulman 2002) was included in the content of a letter. Two trials were reported as conference abstracts (Schulman 2002; Neuman 2013).

Six of the 16 trials were feasibility trials for planned larger studies: Aubron 2012 for the (ongoing) TRANSFUSE trial; Dhabangi 2013 for the (ongoing) TOTAL trial; Hebert 2005 for the (trials awaiting assessment) ABLE trial (Heddle 2012) and Bennett-Guerrero 2009 [1] and Bennett-Guerrero 2009 [2] for the INFORM trial. See [Characteristics of included studies](#) for details of these trials.

Sample sizes

A total of 1864 participants were randomly assigned to the 16 trials. The numbers of participants randomly assigned to each trial ranged from 17 (Schulman 2002) to 910 (Heddle 2012), with only two trials randomly assigning more than 100 participants: Heddle 2012 (n = 910) and Fergusson 2012 (n = 377). The total number of participants included per outcome is detailed in the [Effects of interventions](#) section and ranged from 23 to 1177.

Setting

Three trials were multi-centre studies with participants from across Canada (Hebert 2005; Fergusson 2012) or Australia (Aubron 2012). The 13 single-centre trials were from the USA (Liu 1994; Strauss 1996; Strauss 2000; Schulman 2002; Bennett-Guerrero 2009 [1]; Bennett-Guerrero 2009 [2]; Kor 2012; Neuman 2013), Brazil (Fernandes 2005), Canada (Heddle 2012), Scotland (Walsh 2004), The Netherlands (Yuruk 2013) and Uganda (Dhabangi 2013).

Participants

Eligible trials included neonatal, paediatric or adult participants. Five trials randomly assigned very low birth weight premature neonate participants (Liu 1994; Strauss 1996; Strauss 2000; Fernandes 2005; Fergusson 2012), one trial focused on paediatric participants with malarial anaemia (Dhabangi 2013) and 10 trials

included adult participants (Schulman 2002; Walsh 2004; Hebert 2005; Bennett-Guerrero 2009 [1]; Bennett-Guerrero 2009 [2]; Aubron 2012; Heddle 2012; Kor 2012; Neuman 2013; Yuruk 2013). In trials including adult participants, the reason for hospital stay was the need to undergo cardiac surgery (Hebert 2005; Bennett-Guerrero 2009 [1]; Bennett-Guerrero 2009 [2]), as well as the need for anaemic haematology treatment (Yuruk 2013), treatment for critical illness (Walsh 2004), requirement for treatment in an ICU (Aubron 2012), admission to a level 1 trauma centre (Schulman 2002), the need for intubation and mechanical ventilation (Kor 2012) and the “requirement for a red blood cell transfusion” (no further details given) (Heddle 2012; Neuman 2013).

Interventions

The definition (i.e. the trial protocol or Methods section that defined the age of the red blood cells) and the actual age of red blood cells transfused to participants differed markedly across intervention arms and between trials (see [Table 1](#)). This was the case for both trials comparing fresher red blood cells with older red blood cells and those comparing fresher red blood cells with current standard practice.

In eight trials, the intervention arms compared fresher red blood cell transfusion versus active transfusion of older red blood cells (Liu 1994; Schulman 2002; Walsh 2004; Fernandes 2005; Bennett-Guerrero 2009 [1]; Dhabangi 2013; Neuman 2013; Yuruk 2013). The defined age of the ‘actively transfused older red blood cells’ ranged from ‘20 days or older’ (Schulman 2002; Walsh 2004) to ‘a maximum of 35 days old’ (Liu 1994); whilst the actual age ranged from a mean (SD) of 9.0 (8.9) days (Fernandes 2005) to a mean (SD) of 29.8 (5.7) days (Neuman 2013).

In eight trials, the intervention arms compared fresher red blood cell transfusion versus current standard practice (Strauss 1996; Strauss 2000; Hebert 2005; Bennett-Guerrero 2009 [2]; Aubron 2012; Fergusson 2012; Heddle 2012; Kor 2012). The defined age of the ‘current standard practice’ red blood cells included ‘from a range of 2 to 42 days old’ (Fergusson 2012), ‘oldest in the inventory’ (Bennett-Guerrero 2009 [2]; Heddle 2012) and less than 42 days old (Strauss 1996; Strauss 2000). The actual age of the ‘actively transfused older red blood cells’ ranged from less than 7 days old (Strauss 1996; Strauss 2000) to up to between 35 and 42 days old (Strauss 1996; Strauss 2000).

Across all the trials, the defined age of ‘fresh’ red blood cells ranged from ‘freshest available’ (Aubron 2012) to less than 21 days old (Bennett-Guerrero 2009 [2]). The actual age of the ‘fresh’ red blood cells transfused to participants ranged from a mean (SD) of 1.6 (0.6) days in Fernandes 2005 to a mean (SD) of 12.1 (3.8) days in Aubron 2012.

The additive solutions used to preserve red blood cells ahead of transfusion and whether red blood cells were leucoreduced and/or irradiated also differed between trials (see [Table 1](#)).

One trial ([Hebert 2005](#)) reported instances of non-compliance with allocated treatment. The other 15 trials did not mention compliance with treatment allocation in their manuscript or reported no non-compliance with treatment allocation.

We identified no trials that compared the third of our stated interventions, which was specific to a neonatal population: fresher red blood cell transfusion (given as a historical standard of practice in neonatal patients) versus active transfusion of older red blood cells.

Outcomes

No trial measured all outcomes of interest in this review.

Follow-up periods for primary outcomes measured by the included trials ranged from 'within four hours of a red blood cell transfusion' to 90 days after randomisation. Details of the primary outcomes measured can be found in the [Characteristics of included studies](#) table. All studies reported measuring secondary outcomes, as detailed in the [Characteristics of included studies](#) table.

Excluded studies

For the full list of excluded studies, see [Characteristics of excluded studies](#).

We excluded six studies from this review following assessment of the full text. We excluded two studies because the unit of randomisation was the intervention ([Eshleman 1994](#); [Seitelbach 2011](#)), another because it was an observational, non-randomised study ([Lebiedz 2012](#)), one study as the result of ineligible interventions ([Wasser 1989](#)) and one because it is a composite of the data subsequently presented in two full-text papers ([Strauss 1996](#); [Strauss 2000](#)), but data presented in the excluded paper ([Strauss 1994](#)) are not reported separately for each of the different intervention arms.

Ongoing studies and studies awaiting classification

We identified six ongoing trials ([Kaukonen 2013](#); [NCT01586923](#); [NCT01976234](#); [NCT01977547](#); [NCT02050230](#); [NCT02087514](#); see [Characteristics of ongoing studies](#) table).

We assessed three studies as awaiting assessment ([Steiner 2010](#); [Lacroix 2011](#); [NCT00458783](#); see [Characteristics of studies awaiting classification](#) table). Two of these trials are completed ([Steiner 2010](#); [Lacroix 2011](#)). The data have been analysed and will be reported in scientific journals in 2015. We will include both in a subsequent update to this review. The third trial is identified as completed on [ClinicalTrials.gov](#) (accessed 9 October 2014), but no further details are reported.

Risk of bias in included studies

Allocation

Random sequence generation

We assessed random sequence generation as presenting low risk of bias in nine studies because it was undertaken using a Web-based system ([Hebert 2005](#); [Bennett-Guerrero 2009 \[1\]](#); [Bennett-Guerrero 2009 \[2\]](#); [Aubron 2012](#); [Heddle 2012](#); [Kor 2012](#)), an interactive voice response system (and thereafter referral to a manual of unique random numbers generated by an independent statistician before study activation to determine trial arm allocation) ([Fergusson 2012](#)), a random numbers table ([Liu 1994](#)) and random length block randomisation by an external, independent research unit ([Walsh 2004](#)).

We assessed random sequence generation as presenting unclear risk of bias in seven studies because information on which to base an assessment of bias risk was insufficient ([Strauss 1996](#); [Strauss 2000](#); [Schulman 2002](#); [Fernandes 2005](#); [Dhabangi 2013](#); [Neuman 2013](#); [Yuruk 2013](#)). We assessed no trial as having high risk of bias (see [Figure 2](#); [Figure 3](#)).

Figure 2. Risk of bias graph: review authors' judgements about each risk of bias item presented as percentages across all included studies. Sixteen studies are included in this review.

Figure 3. Risk of bias summary: review authors' judgements about each risk of bias item for each included study.

	Random sequence generation (selection bias)	Allocation concealment (selection bias)	Blinding of participants (performance bias)	Blinding of personnel (performance bias)	Blinding of outcome assessment (detection bias)	Incomplete outcome data (attrition bias)	Selective reporting (reporting bias)	Other bias
Aubron 2012	+	+	+	+	?	+	+	+
Bennett-Guerrero 2009 [1]	+	+	+	+	?	+	+	+
Bennett-Guerrero 2009 [2]	+	+	+	+	?	+	+	+
Dhabangi 2013	?	?	?	?	?	+	+	+
Fergusson 2012	+	+	+	+	+	+	+	+
Fernandes 2005	?	?	?	?	?	+	+	+
Hebert 2005	+	+	+	?	?	-	+	+
Heddle 2012	+	-	+	+	+	+	+	+
Kor 2012	+	-	+	+	?	+	+	+
Liu 1994	+	?	?	?	?	+	+	+
Neuman 2013	?	?	?	?	?	?	?	+
Schulman 2002	?	?	?	?	?	?	?	+
Strauss 1996	?	?	+	+	?	+	?	+
Strauss 2000	?	?	+	+	?	+	?	+
Walsh 2004	+	-	+	+	?	+	+	+
Yuruk 2013	?	?	?	?	?	+	+	+

Allocation concealment

We assessed allocation concealment as bringing low risk of bias in five studies because participants' specific identification numbers were used with only the transfusion service scientist unblinded to treatment allocation (Aubron 2012); a computerised randomisation schedule was maintained by non-trial personnel (Bennett-Guerrero 2009 [1]; Bennett-Guerrero 2009 [2]); generation of random sequence occurred in a way that was out of the treating clinician's control (Fergusson 2012); and allocation details were stored in sequentially numbered, sealed opaque envelopes (Hebert 2005).

We assessed allocation concealment as causing high risk of bias in three studies (Walsh 2004; Heddle 2012; Kor 2012) because randomisation was performed using an unconcealed paper-based sequence held in the blood bank (Heddle 2012), or because the envelopes containing details of treatment allocation were not sequentially numbered (Kor 2012) or were not sequentially numbered and opaque (Walsh 2004).

We assessed allocation concealment as presenting unclear risk of bias in eight studies because information on which to base an assessment of bias risk was insufficient (Liu 1994; Strauss 1996; Strauss 2000; Schulman 2002; Fernandes 2005; Dhabangi 2013; Neuman 2013; Yuruk 2013).

Blinding

We report details regarding blinding to treatment allocation separately for participants, study personnel and outcome assessors.

Blinding of participants

We assessed blinding of participants as low risk of bias in 10 studies. In seven studies, participants were receiving intensive care treatment (Walsh 2004; Hebert 2005; Aubron 2012; Kor 2012) or were neonates whose parents were blinded to treatment allocation (Strauss 1996; Strauss 2000; Fergusson 2012); in Bennett-Guerrero 2009 [1] and Bennett-Guerrero 2009 [2], red blood cell units were provided by the hospital's transfusion service in a blinded manner from induction of general anaesthesia to postoperative day 7, such that participants were blinded to study group assignment. In Heddle 2012, participants were unblinded to treatment allocation, but as no outcomes were subjectively measured, knowledge of treatment allocation was not believed to constitute risk of bias in this trial.

We assessed blinding of participants as presenting unclear risk of bias in six studies (Liu 1994; Schulman 2002; Fernandes 2005; Dhabangi 2013; Neuman 2013; Yuruk 2013) because information reported to permit assessment of risk of bias was insufficient. We assessed no trial as having high risk of bias.

Blinding of study personnel

We assessed blinding of study personnel to treatment allocation as presenting low risk of bias in nine trials. In eight of these trials, study personnel were reported to be blinded to treatment allocation and it is unlikely that blinding could be broken (Strauss 1996; Strauss 2000; Walsh 2004; Bennett-Guerrero 2009 [1]; Bennett-Guerrero 2009 [2]; Aubron 2012; Fergusson 2012; Kor 2012). In Heddle 2012, study personnel were unblinded to treatment allocation, but as no outcomes were subjectively measured, knowledge of treatment allocation was not believed to constitute risk of bias in this trial.

We assessed blinding of study personnel as causing unclear risk of bias in seven studies (Liu 1994; Schulman 2002; Fernandes 2005; Hebert 2005; Dhabangi 2013; Neuman 2013; Yuruk 2013) because information reported to permit assessment of risk of bias was insufficient. We assessed no trial as having high risk of bias.

Blinding of outcome assessors

Two trials reported blinding of outcome assessors (Fergusson 2012; Heddle 2012): both were deemed to be at low risk of detection bias. In Fergusson 2012, composite outcomes were adjudicated independently by neonatologists blinded to study group allocation. In Heddle 2012, outcome assessors were not blinded, but as outcomes were objective, we believe that such knowledge would not have had an impact on outcome assessment.

We assessed blinding of outcome assessors as presenting unclear risk of bias in 14 studies (Liu 1994; Strauss 1996; Strauss 2000; Schulman 2002; Walsh 2004; Fernandes 2005; Hebert 2005; Bennett-Guerrero 2009 [1]; Bennett-Guerrero 2009 [2]; Aubron 2012; Kor 2012; Dhabangi 2013; Neuman 2013; Yuruk 2013) because information reported on how outcomes were assessed was insufficient. We assessed no trial as having high risk of bias.

Incomplete outcome data

In all, 14 trials were at low risk of attrition bias, as all participants who were randomly assigned and received a red blood cell transfusion were included in the analysis of outcome data, and no participants were lost during follow-up (Liu 1994; Strauss 1996; Strauss 2000; Walsh 2004; Fernandes 2005; Bennett-Guerrero 2009 [1]; Bennett-Guerrero 2009 [2]; Aubron 2012; Fergusson 2012; Heddle 2012; Kor 2012; Dhabangi 2013; Neuman 2013; Yuruk 2013). Information needed to assess attrition bias was insufficient in Schulman 2002.

One trial was at high risk of attrition bias (Hebert 2005). We made this assessment because outcome data for participants in analysis of 30-day and 90-day follow-up mortality outcomes were incomplete.

Selective reporting

In 13 trials, investigators reported all prespecified outcomes in the Results section, and we deemed them to be at low risk of reporting bias (Liu 1994; Walsh 2004; Fernandes 2005; Hebert 2005; Bennett-Guerrero 2009 [1]; Bennett-Guerrero 2009 [2]; Aubron 2012; Fergusson 2012; Heddle 2012; Kor 2012; Dhabangi 2013; Neuman 2013; Yuruk 2013). Three trials did not define in their Methods the outcomes they were interested in measuring; therefore, it is impossible to identify whether reporting bias was present in these trials (Strauss 1996; Strauss 2000; Schulman 2002), and we have rated them as having unclear risk of bias. We assessed no trial as having high risk of bias.

Other potential sources of bias

We have no concerns about other potential sources of bias.

Effects of interventions

We have reported outcome data in this review by intervention and then by outcome. We further subdivided these sections into trials of neonate participants, those including paediatric participants and those with adult participants.

When denominator data allowed, we converted reported percentages into actual numbers of participants in two trials (Bennett-Guerrero 2009 [1]; Bennett-Guerrero 2009 [2]) to enable us to calculate outcome risk ratios for these trials.

Fresher red blood cell transfusion versus active transfusion of older red blood cells

Eight trials (with 279 participants) reported data for this comparison (Liu 1994; Schulman 2002; Walsh 2004; Fernandes 2005; Bennett-Guerrero 2009 [1]; Dhabangi 2013; Neuman 2013; Yuruk 2013). Two trials included neonate participants (Liu 1994; Fernandes 2005), one trial included paediatric participants (Dhabangi 2013) and five trials included adult participants (Schulman 2002; Walsh 2004; Bennett-Guerrero 2009 [1]; Neuman 2013; Yuruk 2013).

We used the terms 'fresher' when referring to the fresher red blood cell transfusion intervention arm and 'older' when referring to the actively transfused older red blood cells intervention arm. Diversity in the clinical setting; age of participants (see [Characteristics of included studies](#)); variability in what constituted 'fresher', 'older' and 'current standard practice' red blood cells within the interventions of each of the trials; (see [Table 1](#)) and differences in how outcomes were measured have meant that we had no opportunity to pool data for any of our primary and secondary outcomes. Therefore throughout this section, data are reported per trial.

Primary outcome

Mortality: immediate (occurring within seven days in hospital) and short term (up to 30 days)

Trials with neonate participants

No trial in this participant population reported data for this outcome.

Trials with paediatric participants

Dhabangi 2013 reported data on mortality within seven days in hospital. Investigators reported no difference in the number of deaths between 'fresher' and 'older' intervention arms (risk ratio (RR) 3.00, 95% confidence interval (CI) 0.13 to 71.34; 74 participants). Researchers reported only one death, which occurred within three hours of a red blood cell transfusion in a child randomly assigned to receive fresh red blood cells.

Trials with adult participants

Bennett-Guerrero 2009 [1], Schulman 2002 and Walsh 2004 reported data on mortality up to 30 days. We did not undertake a meta-analysis because participants had different clinical diagnoses (in Bennett-Guerrero 2009 [1], cardiac surgery; in Schulman 2002, trauma and in Walsh 2004, critically ill participants; see [Characteristics of included studies](#) table).

Researchers reported no difference in the number of deaths at up to 30 days between 'fresher' and 'older' intervention arms in two studies (RR 2.77, 95% CI 0.12 to 61.65; 23 participants in Bennett-Guerrero 2009 [1]; RR 2.25, 95% CI 0.55 to 9.17; 17 participants in Schulman 2002). In Walsh 2004, investigators reported no deaths.

Secondary outcomes

Long-term mortality

Trials with neonate participants

Fernandes 2005 reported mortality at up to 40 weeks of gestational age and observed no difference in the number of deaths reported between 'fresher' and 'older' intervention arms (RR 0.90, 95% CI 0.41 to 1.85; 52 participants).

Trials with paediatric participants

The only trial in this participant population (Dhabangi 2013) did not report data for this outcome.

Trials with adult participants

No trial in this participant population reported data for this outcome.

Clinically accepted measures of multiple organ dysfunction (e.g. Multiple Organ Dysfunction Score (MODS), Sequential Organ Failure Assessment (SOFA) score) and numbers of dysfunctional organs reported per participant

Trials with neonate participants

No trial in this participant population reported data for this outcome.

Trials with paediatric participants

The only trial in this participant population (Dhabangi 2013) did not report data for this outcome.

Trials with adult participants

Walsh 2004 reported data for this outcome and measured organ failure score on the day of the study. They noted no differences in mean organ failure scores between 'fresher' and 'older' intervention arms (mean difference (MD) 0.60, 95% CI -1.71 to 2.91; 22 participants).

Incidence of in-hospital infection

Trials with neonate participants

Fernandes 2005 reported data for this outcome as the number of neonates developing clinical sepsis and necrotising enterocolitis. They observed no differences between 'fresher' and 'older' in incidence of clinical sepsis (RR 1.25, 95% CI 1.00 to 1.56; 52 participants) and necrotising enterocolitis (RR 1.50, 95% CI 0.48 to 4.70; 52 participants). The number of neonates developing clinical sepsis was high at 96% in those receiving 'fresher' red blood cells, and at 77% in those receiving 'older' red blood cells. The number of neonates developing necrotising enterocolitis was considerably smaller at 23% among those receiving 'fresher' red blood cells and 15% in those receiving 'older' red blood cells.

Trials with paediatric participants

The only trial in this participant population (Dhabangi 2013) did not report data for this outcome.

Trials with adult participants

No trial in this participant population reported data for this outcome.

Duration of respiratory support (invasive and non-invasive ventilation), haemodynamic support (inotropic) and renal support (haemofiltration)

Duration of respiratory support was reported as duration of mechanical ventilation and as the number of participants requiring mechanical ventilation. In terms of reporting of the 'duration of mechanical ventilation', this type of reporting falls under the same consideration as per length of stay, in that it is usually a skewed distribution and hence is better reported as a median value (and an interquartile range); none of the included trials did this. When trials reported data on the duration of mechanical ventilation as means and standard deviations, we did not analyse the data but reported data narratively and tabulated (see Table 2).

Trials with neonate participants

Fernandes 2005 reported respiratory data for this outcome by reporting the mean duration of mechanical ventilation. Participants receiving 'fresher' red blood cells required fewer days of mechanical ventilation when compared with participants receiving 'older' red blood cells (see Table 2). No trial reported data on haemodynamic or renal support.

Trials with paediatric participants

The only trial in this participant population (Dhabangi 2013) did not report data for this outcome.

Trials with adult participants

Bennett-Guerrero 2009 [1] reported respiratory data for this outcome by reporting the mean duration of mechanical ventilation and by measuring the number of participants on any vasopressor longer than 48 hours after surgery. Investigators observed no differences between 'fresher' and 'older' intervention arms in the numbers of participants on any vasopressor longer than 48 hours after surgery (RR 0.61, 95% CI 0.12 to 3.00; 23 participants). Adult participants receiving 'old' red blood cells required fewer

hours of mechanical ventilation when compared with adult participants receiving 'fresher' red blood cells (see [Table 2](#)). No trial reported data on haemodynamic or renal support.

Length of hospital and ICU stay

Trials with neonate participants

No trial reported median (and interquartile range) data for this outcome. [Fernandes 2005](#) reported mean (with standard deviation) length of hospital stay. We are not reporting these data: Actual data are provided in [Table 3](#).

Trials with paediatric participants

The only trial in this participant population ([Dhabangi 2013](#)) did not report data for this outcome.

Trials with adult participants

No trial reported median (and interquartile range) data for this outcome. [Bennett-Guerrero 2009 \[1\]](#) reported mean (with standard deviations) length of hospital stay. We did not analyse these data: actual data are presented in [Table 3](#).

Adverse transfusion reactions

No trials reported data for this outcome.

Physiological markers of oxygen consumption or alterations in microcirculation

One trial with neonate participants ([Liu 1994](#)), the only trial with paediatric participants ([Dhabangi 2013](#)) and four trials with adult participants reported data for this outcome ([Bennett-Guerrero 2009 \[1\]](#); [Neuman 2013](#); [Walsh 2004](#); [Yuruk 2013](#)).

We did not undertake a meta-analysis of adult participant trials because of differences in the way that this outcome was measured across trials (see [Table 4](#)). Nor did we statistically analyse any of these outcome data because the measures used could not be compared; instead we have reported individual study data in [Table 4](#). No trial measured the same physiological markers of oxygen consumption or alterations in microcirculation. Two trials measured one marker each ([Dhabangi 2013](#); [Yuruk 2013](#)); two trials measured two markers ([Liu 1994](#); [Bennett-Guerrero 2009 \[1\]](#)); [Neuman 2013](#) measured three markers; and [Walsh 2004](#) measured eight markers.

Assessment of economic or blood stock inventory outcomes

Trials with neonate participants

[Fernandes 2005](#) and [Liu 1994](#) reported data on these outcomes. The outcome was measured as mean number of donor exposures per infant. The mean number of donor exposures per infant is of particular interest in this population of very low birth weight, premature neonates. In practice, red blood cells from dedicated donors may be reserved for use by a particular neonate, hence becoming a blood stock management activity. Investigators noted a difference in the mean number of donor exposures per infant favouring the 'older' intervention arm in both trials (MD 2.80, 95% CI 1.46 to 4.14; 52 participants in [Fernandes 2005](#); MD 1.70, 95% CI 0.07 to 3.33; 25 participants in [Liu 1994](#)).

Trials with paediatric participants

The only trial in this participant population ([Dhabangi 2013](#)) did not report data for this outcome.

Trials with adult participants

[Yuruk 2013](#) reported data on this outcome measured as the median (and interquartile range) number of bags of red blood cells transfused to adult haematology participants. The median number of bags was the same in both intervention arms: in the 'fresher' arm, a median (interquartile range) of 3 (2 to 3) bags, and in the 'older' arm, a median of 3 (3 to 3) bags.

Fresher red blood cell transfusion versus current standard practice

Eight studies (with 1585 participants) reported data for this comparison ([Strauss 1996](#); [Strauss 2000](#); [Hebert 2005](#); [Bennett-Guerrero 2009 \[2\]](#); [Aubron 2012](#); [Fergusson 2012](#); [Heddle 2012](#); [Kor 2012](#)). Three studies included neonate participants ([Strauss 1996](#); [Strauss 2000](#); [Fergusson 2012](#)), and five studies included adult participants ([Hebert 2005](#); [Bennett-Guerrero 2009 \[2\]](#); [Aubron 2012](#); [Heddle 2012](#); [Kor 2012](#)).

We used the terms 'fresher' when referring to the fresher red blood cell transfusion intervention arm and 'standard practice' when referring to the current standard practice intervention arm. Diversity in the clinical setting, age of participants (see [Characteristics of included studies](#)), variability in what constituted 'fresher' and 'current standard practice' red blood cells within interventions of each of the trials (see Additional tables) and differences in how outcomes were measured have meant that we had no opportunity to pool data for any of our primary and secondary outcomes. Therefore we have reported data per trial throughout this section.

We calculated the number of participants included in the analysis of mortality at 30-day and 90-day follow-up points in both intervention arms in [Hebert 2005](#) from data provided in study authors' Table 2 (page 1437), as this value was not reported. In Table 2 (page 1437), [Hebert 2005](#) reported the overall number of participants included in these analyses and events and percentage event rates. From these figures, it was possible to calculate the actual number of participants included in these outcome analyses per intervention.

Primary outcome

Mortality: immediate (occurring within seven days in hospital) and short term (up to 30 days)

Trials with neonate participants

[Strauss 1996](#) (40 participants) reported data on mortality up to 30 days. They observed no differences in the numbers of reported deaths at up to 30 days between 'fresher' and 'standard practice' intervention arms (RR 0.30, 95% CI 0.01 to 7.02; 40 participants).

Trials with adult participants

[Aubron 2012](#), [Hebert 2005](#) and [Heddle 2012](#) (1018 participants) reported data on mortality within seven days in hospital, and [Bennett-Guerrero 2009 \[2\]](#), [Hebert 2005](#) and [Kor 2012](#) (172 participants) reported data on mortality up to 30 days. We did not undertake a meta-analysis for either measure because of diversity in clinical diagnoses between randomly assigned participants in these trials (see [Characteristics of included studies](#) table).

Investigators reported no differences in the numbers of reported deaths within seven days in hospital between 'fresher' and 'standard practice' intervention arms in the three trials (RR 2.60, 95% CI 0.55 to 12.29; 51 participants in [Aubron 2012](#); RR 1.99, 95% CI 0.52 to 7.54; 57 participants in [Hebert 2005](#); RR 1.12, 95% CI 0.75 to 1.65; 910 participants in [Heddle 2012](#)).

They observed no differences in the numbers of reported deaths at up to 30 days between 'fresher' and 'standard practice' intervention arms in [Hebert 2005](#) (RR 0.21, 95% CI 0.54 to 7.58; 53 participants) and in [Kor 2012](#) (RR 0.79, 95% CI 0.48 to 1.29; 99 participants). In [Bennett-Guerrero 2009 \[2\]](#), no deaths were reported among 20 participants.

Secondary outcomes

Long-term mortality

Trials with neonate participants

[Fergusson 2012](#) reported data on mortality at 90 days' follow-up with no differences in the numbers of deaths reported between 'fresher' and 'standard practice' intervention arms (RR 0.97, 95% CI 0.61 to 1.54; 377 participants).

Trials with adult participants

[Hebert 2005](#) reported data on mortality at 90 days' follow-up with no differences in the numbers of deaths reported between 'fresher' and 'standard practice' intervention arms (RR 1.81, 95% CI 0.55 to 5.93; 49 participants).

Clinically accepted measures of multiple organ dysfunction

Trials with neonate participants

[Fergusson 2012](#) reported data on this outcome with no differences between 'fresher' and 'standard practice' arms in the numbers of premature neonates with intraventricular haemorrhage (RR 1.65, 95% CI 0.80 to 3.39; 377 participants), retinopathy of prematurity (RR 0.89, 95% CI 0.53 to 1.50; 377 participants) and bronchopulmonary dysplasia (RR 0.96, 95% CI 0.72 to 1.28; 377 participants).

Trials with adult participants

[Kor 2012](#) reported data on this outcome for 99 participants and reported changes in sequential organ failure assessment (SOFA) scores from before until after transfusion (a mean (SD) of 1.8 (0.46) hours in the 'fresher' intervention arm and 1.9 (0.63) hours in the 'standard practice' intervention arm). The mean change in SOFA score in the 'fresher' versus 'standard issue' intervention arm was -0.7 versus -1.0. Researchers provided no further details for this outcome.

Incidence of in-hospital infection

Trials with neonate participants

[Fergusson 2012](#) reported data on this outcome and observed no differences between 'fresher' and 'standard practice' arms in the numbers of premature neonates with necrotising enterocolitis (RR 1.01, 95% CI 0.51 to 2.00; 377 participants), the numbers of clinically suspected infections (RR 1.01, 95% CI 0.90 to 1.12; 377 participants) and the numbers of confirmed infections (RR 1.06, 95% CI 0.91 to 1.22; 377 participants).

Trials with adult participants

Hebert 2005 reported no incidences of bacteraemia, deep incisional infection, endocarditis or mediastinitis in 57 participants. No other trial in this participant population reported data for this outcome.

Duration of respiratory support (invasive and non-invasive ventilation), haemodynamic support (inotropic) and renal support (haemofiltration)

Duration of respiratory support was reported as duration of mechanical ventilation and as the number of participants requiring mechanical ventilation. In terms of reporting of the 'duration of mechanical ventilation', this type of reporting falls under the same consideration as per length of stay, in that it is usually a skewed distribution and hence is better reported as a median value (and interquartile range); none of the included trials did this. When trials reported data on the duration of mechanical ventilation, we did not analyse the data but reported data narratively and tabulated (see Table 2).

Trials with neonate participants

Fergusson 2012 reported data on respiratory support and counted the numbers of participants requiring mechanical ventilation and the numbers of participants requiring high-frequency ventilation, noting no difference between 'fresher' and 'standard practice' arms in the number of people requiring mechanical ventilation (RR 0.99, 95% CI 0.90 to 1.10; 377 participants) or high-frequency ventilation (RR 1.02, 95% CI 0.79 to 1.31; 377 participants). Full study data are reported in Table 2. No trial reported data on haemodynamic or renal support.

Trials with adult participants

Three trials reported data on respiratory support (Hebert 2005; Bennett-Guerrero 2009 [2]; Aubron 2012), and one trial reported data on renal support (Hebert 2005). No trial reported data on haemodynamic support. A variety of methods were used to measure this outcome. We did not undertake a meta-analysis because of clinical diversity (participant clinical setting of intensive care unit in Aubron 2012, and cardiac surgery in Hebert 2005) and because of the ways in which outcomes were measured (see Table 2).

In Aubron 2012 and Hebert 2005, researchers reported no differences in the numbers of people requiring mechanical ventilation (RR 1.26, 95% CI 0.97 to 1.63; 51 participants in Aubron 2012; RR 1.38, 95% CI 1.00 to 1.91; 57 participants in Hebert 2005). In Bennett-Guerrero 2009 [2], investigators noted no differences between 'fresher' and 'standard practice' arms in the numbers of participants taking any vasopressor longer than 48 hours after

surgery (RR 3.67, 95% CI 0.46 to 29.49; 20 participants), and participants receiving 'standard practice' red blood cells required fewer hours of mechanical ventilation when compared with adult participants receiving 'fresher' red blood cells (see Table 2).

In Hebert 2005, researchers reported no differences between 'fresher' and 'standard practice' arms in the numbers of participants requiring prolonged invasive mechanical ventilation for longer than 48 hours (RR 1.67, 95% CI 0.60 to 4.64; 57 participants), or in the numbers of participants requiring prolonged mechanical ventilation for longer than 48 hours (RR 1.34, 95% CI 0.60 to 2.98; 57 participants) or requiring vasoactive drugs, an aortic balloon pump or ventricular assist for longer than 48 hours (RR 1.49, 95% CI 0.45 to 4.98; 57 participants).

Hebert 2005 reported the numbers of participants requiring renal dialysis at 30 days and observed no differences in the numbers of participants requiring renal dialysis at 30 days between 'fresher' and 'standard practice' intervention arms (RR 0.24, 95% CI 0.01 to 4.73; 57 participants).

Length of hospital and ICU stay

Trials with neonate participants

Strauss 1996 reported percentage data for length of hospital stay, and Fergusson 2012 reported median (plus interquartile range) data for the duration of intensive care unit stay. These data are available in Table 3.

Strauss 1996 reported the percentage of neonates discharged up to day 84: 67% in the 'fresher' intervention arm and 53% in the 'standard care' intervention arm. These data are available in Table 3.

Fergusson 2012 presented data as median (plus interquartile range). The 'fresher' intervention arm required a longer stay in the intensive care unit. Median values are reported in Table 3.

Trials with adult participants

Aubron 2012 presented data as median (plus interquartile range). The 'fresher' intervention arm required a longer stay in hospital and a longer stay in the intensive care unit than the 'standard practice' intervention arm. Median values are reported in Table 3. Bennett-Guerrero 2009 [2] reported mean (with standard deviation) length of hospital stay. We did not report these data: Actual data are presented in Table 3.

Adverse transfusion reactions

Trials with neonate participants

Three trials (with 431 neonate participants) reported data for this outcome (Strauss 1996; Strauss 2000; Fergusson 2012). We did not undertake a meta-analysis because event data were lacking. No incidence of adverse transfusion reactions was reported in Strauss 1996 or Strauss 2000, along with one diagnosis of cytomegalovirus (CMV) infection within a 90-day neonatal intensive care unit stay in a neonate randomly assigned to 'standard practice' in Fergusson 2012.

Trials with adult participants

No trial in this participant population reported data for this outcome.

Physiological markers of oxygen consumption or alterations in microcirculation

Three trials with neonate participants (431 participants) (Strauss 1996; Strauss 2000; Fergusson 2012) and two trials with 119 adult participants reported data for this outcome (Bennett-Guerrero 2009 [2]; Kor 2012).

We did not undertake a meta-analysis of either participant population because of differences in the way that this outcome was measured across trials (see Table 4). Nor did we analyse any of these outcome data, as data could not be compared with the measures used; instead individual study data are reported in Table 4. No trial measured the same physiological markers of oxygen consumption or alterations in microcirculation. One trial measured one marker (Kor 2012); two trials measured two markers (Bennett-Guerrero 2009 [2]; Fergusson 2012); Strauss 1996 and Strauss 2000 measured six markers.

Assessment of economic or blood stock inventory outcomes

Trials with neonate participants

Fergusson 2012, Strauss 1996 and Strauss 2000 reported this outcome as the mean number of donor exposures per infant. The mean number of donor exposures per infant is of particular interest in this population of very low birth weight, premature neonates. In practice, red blood cells from dedicated donors may be reserved for use by a particular neonate, thereby becoming a blood stock management activity. Differences in the mean numbers of donor exposures per infant favoured the 'standard practice' intervention arm in both trials (MD 1.62, 95% CI 1.17 to 2.07; 377 premature neonate participants in Fergusson 2012; MD 2.10, 95% CI 0.82 to 3.38; 29 premature neonate participants in Strauss 1996; MD 4.60, 95% CI 2.28 to 6.92; 21 premature neonate participants in Strauss 2000).

Trials with adult participants

No trial in this participant population reported data for this outcome.

DISCUSSION

Summary of main results

This review has identified 16 randomised controlled trials (with 1864 participants). Diversity in the age of included participants (five trials enrolled neonates, one children and 12 adult participants); in the protocol definition; in the actual ages of 'fresher', 'older' and 'standard practice' transfused red blood cells; and in the measure and recording of outcomes precluded meta-analysis. Therefore we tabulated and reported individual study data by outcome. Only four trials evaluated clinical outcome as the main outcome; in 10 included trials, physiological parameters constituted the primary outcome, and two trials did not report the primary outcome.

In all trials, no clear difference was reported between 'fresher' and 'older' red blood cell transfusion arms, or between 'fresher' and 'standard practice' red blood cell transfusion arms, for the outcomes of mortality (immediate, short term and long term), clinically accepted measures of multiple organ dysfunction, incidence of in-hospital infection, duration of respiratory and renal support and adverse transfusion reactions. Assessment of economic or blood stock inventory was reported as the mean number of donor exposures in two neonatal trials; in both trials, participants receiving 'fresher' blood had a significantly greater number of donor exposures than participants receiving the 'older' or 'standard practice' intervention.

No trial reported on all outcomes. No trial measured duration of haemodynamic support. Two outcomes - length of hospital or intensive care unit stay, and physiological markers of oxygen consumption or alterations in microcirculation - were not formally assessed within the review because evidence suggested skewed data for length of stay and considerable heterogeneity was apparent in the reporting of physiological markers of oxygen consumption or alterations in microcirculation (presented in Table 4).

We identified three trials awaiting further assessment and five ongoing randomised controlled trials that evaluated mortality and organ dysfunction as primary outcomes, and that need to be incorporated into future updates of this systematic review.

Overall completeness and applicability of evidence

Applicability of the evidence is complicated by several factors. Identified studies recruited patients across a very broad range of patient settings, including trauma, cancer and cardiac disease. Whilst common pathophysiological mechanisms may apply across all participant groups, how the size of this effect may differ between subpopulations is unclear. Moreover these participant populations will have different baseline risks for outcomes, reflecting the case mix and degree of severity of the underlying illness, and therefore might be differentially affected by transfusion of older storage age red cells. This diversity was also apparent in the data for total volumes of red blood cells or numbers of transfusions across trials. This clinical diversity is particularly pertinent when randomised controlled trials are considered that recruited the neonatal population, whose physiology is fundamentally different from that of an adult.

Many of the identified trials were smaller studies, and six were feasibility or pilot trials. Only [Fergusson 2012](#) and [Heddle 2012](#) recruited more than 100 participants in the outcome analyses. More than half of trials randomly assigned fewer than 50 participants to the trial. A broad diversity of primary outcomes were assessed, many of which were physiological parameters. Reporting of time scales for recording mortality was variable.

Differing characteristics of red blood cell products may also affect applicability of the evidence. Trials were set in different countries, which had different specifications for red blood cell products including use of leucoreduction and the nature of additive solutions. This potentially allows the findings to be applicable to most countries but may not cover all scenarios. In addition, the age of red blood cells from 'standard practice' will vary according to the policies of the issuing blood bank. Many blood banks operate according to a 'first in first out' policy, whereby the oldest available red blood cell unit is selected for transfusion; this was certainly the case with some of the trials included in this review.

It was not clear to what degree investigators considered the effects of multiple transfusions. For example, in the context of significant bleeding, participants (trauma, critical care) may receive more than one unit of red blood cells during a transfusion episode. In these situations, older storage age units are more likely to be transfused. These factors may not be adequately covered by measures reporting the mean age of all red cell units transfused, the maximum age of red cell units transfused, the mean of the oldest units or the proportion of red cell units older than a given number of days ([Lelubre 2009](#)).

A final consideration when applicability of the evidence is considered relates to whether these trials are adequately powered to identify the different possible temporal patterns linking clinical outcomes with red cell storage lesions ([Pereira 2013](#)). This consideration was discussed in a modelling analysis in which different types of temporal patterns were assumed for red blood cell storage age and clinical consequences, and it was unclear whether trials would have the power to detect deleterious consequences of red cell storage if, for example, the effects occur mainly at prolonged

storage ages.

Quality of the evidence

We assessed risk of bias only. As it was not possible to pool data in a meta-analysis because of clinical diversity, and as differences were noted in how outcomes were measured across studies, we did not explore the quality of the evidence using the GRADE (Grades of Recommendation, Assessment, Development and Evaluation) approach ([Schünemann 2011](#)).

Overall, it was difficult to assess risk of bias accurately, as much information was lacking; many elements of the risk of bias assessment in each trial were marked as having unclear risk of bias. Four incidences of high risk of bias in allocation concealment (three trials for non-use of sequentially numbered, sealed and opaque envelopes) and incomplete outcome data (one trial) were found. No dimension was low across all included trials.

Two domains - allocation concealment and blinding of outcome assessors - had a greater proportion of unclear or high risk of bias assessments (in comparison with low risk of bias assessments) than the other domains (randomisation, sequence generation, blinding of participants and study personnel, selective outcome reporting and incomplete outcome assessment). Blinding may be difficult to achieve for study personnel, as much information is required on the label of a unit of red blood cells to ensure external compliance and safety. Blinding would seem to be possible for outcome assessors if those employed were not core members of the clinical treating team. However, as none of the outcomes were patient-reported ones, we did not deem lack of blinding to be an important risk of bias.

As has already been noted, interpretation of trial findings is complicated by various issues, including overlap in the cut points of aged red blood cells transfused. This and other clinical diversity in the trials, coupled with variability in how outcome data were reported and measured, have prevented pooling of outcome data; thus the consistency (or inconsistency) of results across trials is difficult to ascertain. Addressing such variability should be a key goal of future research in this area. For example, employing different forms of analysis, such as individual patient data analysis, will be an important means of addressing some of these limitations when different trials are evaluated; this will allow for the limitation of overlapping ages in treatment arms.

Potential biases in the review process

We identified no biases in the review process. The strengths of this review lie in the robust and comprehensive methods employed to find and assess all relevant trials. We have followed standard Cochrane methods for data extraction and results analyses with reference to the *Cochrane Handbook for Systematic Reviews of Interventions* ([Higgins 2011a](#); [Higgins 2011b](#); [Higgins 2011c](#)), and

we have referred to a statistician when necessary. We have had a clinician and a methodologist working at all stages, independently of each other, to control for bias that ensues as the result of knowledge of clinical or systematic review methods. Although no trial data were obviously missing, additional information on the methods of the trials might have helped in our risk of bias assessments. However we chose not to contact trial authors because, as we were not able to undertake meta-analyses, we believe that additional information would not have enhanced the review at this stage. We will revisit this decision in future updates of the review.

A potential weakness of this systematic review can be seen in our decision not to predefine 'fresher', 'older' and 'standard practice'. We decided against this early in the review process (reasons outlined in the [Types of interventions](#) section) and still believe that this was the correct decision. Allied with this is the grouping of trials comparing 'freshest available' red blood cells with those comparing 'fresher' red blood cells in the presentation of findings. Both have allowed this systematic review to present all available trial data, which show variability and heterogeneity. In future updates, we may choose to revisit these decision.

Agreements and disagreements with other studies or reviews

Findings of this review are in broad agreement with those of other recent systematic reviews. [Lelubre 2013](#) identified eight randomised trials within a wider review of the literature and reported finding no clear, definitive argument to support the superiority of fresh when compared with older red blood cells. [Flegel 2014](#) presented information on completed and ongoing randomised controlled trials alongside a review of the general literature in this area and noted that none of the 11 randomised controlled trials included in their review detected any effect of red blood cell storage time on clinical outcomes. A recent meta-analysis of observational studies and randomised controlled trials by [Wang 2012](#) reported "detrimental effects of red cell storage on clinical outcomes" including mortality, pneumonia and multi-organ dysfunction syndrome.

A meta-analysis by Vamvakas ([Vamvakas 2010](#); [Vamvakas 2011](#)) included only studies reporting adjusted results for mortality but reported that the age of blood was not associated with greater risk of dying. Some of these reviews included observational studies that have other limitations. It should be noted that for reasons that are unclear, some observational studies have reported an increase in the mortality rate after transfusion of fresh red cells compared with older stored red cells ([Middelburg 2013](#)).

Implications for practice

This review has identified a number of larger ongoing trials. It would be premature to describe implications for practice based on the findings of this review, given the many limitations of the identified randomised controlled trial literature.

An understanding of the clinical consequences of different storage ages for transfusion could have significant operational and financial implications for blood transfusion services. The potential importance of a better understanding of this topic is illustrated by the results of preclinical studies with volunteers receiving red cell units of different storage age. [Hod 2011](#) reported significant increases in serum iron and transference saturation after transfusion of older red blood cells in volunteers. In an animal study, differences were reported in a dog model of experimentally induced bacterial pneumonia in which older storage age red blood cells significantly increased rates of mortality ([Solomon 2013](#)).

Implications for research

The results of this review have more implications for research than for current practice. The main implications for research are twofold: findings for ongoing trials need to be appraised and incorporated into updates of this systematic review, and greater consensus is needed regarding the definitions of 'fresher' and 'older' blood to minimise diversity and thereby limit the possibility of comparative research in this area. (This lack of consensus is evident in the identified ongoing trials.)

Future updates to this review will need to consider whether different forms of analysis should be employed to handle current and potential future variability in interventions and outcomes; whether a 'fresher' red blood cell intervention arm should be further subdivided into 'freshest available' and 'fresher' red blood cells and whether investigators should determine if the size of observed effects is dependent on recipient factors such as clinical background, participant age and clinical presentation. Additional trials in this area need to employ robust methods of allocation blinding, so as to manage selection and performance bias and to consider the sample size of trials.

Robust evidence of clinically meaningful effects of different storage ages of red cells would be expected to lead to significant operational and financial consequences for blood transfusion services. Blood transfusion services might already start to investigate different strategies or modelling of inventories and collection practices in the event of clinical trial evidence of benefit or harm for different storage ages of blood cells.

AUTHORS' CONCLUSIONS

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* Indicates the major publication for the study

CHARACTERISTICS OF STUDIES

Characteristics of included studies *[ordered by study ID]*

Aubron 2012

Methods	<p>Parallel-group, randomised controlled trial, multi-centre study</p> <p>Randomly assigned at: point of transfusion</p> <p>Sample size calculation: none reported</p> <p>Duration of follow-up: for the duration of ICU stay</p>
Participants	<p>Inclusion criteria: aged over 18 years; hospitalised in ICU and prescribed at least 1 red cell unit</p> <p>Exclusion criteria: receipt of palliative care or history of organ transplantation or of haematological disease</p> <p>Transfusion indication: support during acute illness</p>
Interventions	<p>Comparison: fresher red blood cell transfusion vs current standard practice</p> <p>Intervention: 'freshest available'</p> <p>Comparator: "compatible, non expired [red cell] units with the longest storage duration at the time of transfusion request", as per 2010/11 Australian transfusion practice</p> <p>Number of participants enrolled: 52</p> <p>Number of participants randomly assigned to the interventions: 25/26</p> <p>Numbers completing assessment: 25/26</p> <p>Compliance with assigned treatments: Nothing is stated in the text about compliance with assigned interventions</p>
Outcomes	<p>Primary: feasibility trial</p> <ul style="list-style-type: none"> Feasibility of study procedures: success of Web randomisation and blinding of ICU staff; correct delivery of study-assigned red blood cell units (measured at study end) <p>Secondary</p> <ul style="list-style-type: none"> Differences in age of red blood cell units between intervention groups (measured as mean storage duration) In-hospital mortality Duration of ICU and hospital stay (days) Duration of mechanical ventilation (days)
Notes	<p>Population nationality: Australian</p> <p>Sources of funding: Australian Red Blood Cross Service and ANZIC Research Centre, Monash University, Australia</p> <p>Declarations of interest (among primary researchers): "None of the authors have any conflict of interest" (page 1201)</p> <p>Dates the study was conducted: September 2010 to January 2011</p> <p>This is a feasibility study for the 'TRANSFUSE (STandaRd Issue TrAnsfusion versuS Fresher red blood cell Use in intenSive carE - a randomised controlled trial)' trial. The TRANSFUSE trial is an ongoing trial, listed in this review under Kaukonen 2013</p>
<i>Risk of bias</i>	

Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Low risk	Randomly assigned 1:1 by intensive care unit (ICU) staff using a Web-based system. System generated a participant-specific identification number, which maintained blinding of clinical and ICU research staff
Allocation concealment (selection bias)	Low risk	Participant-specific identification number was sent (in writing and by telephone) to the transfusion service with a notification of transfusion request. The unique number when applied to a printed randomisation schedule of participant study numbers held only by the transfusion laboratory allowed the unblinded transfusion service scientist to issue trial-specific red blood cell units. The transfusion service computer system ensured that all subsequent units were similarly allocated while the participant was in the ICU
Blinding of participants (performance bias)	Low risk	All participants were treated in an intensive care whilst involved in the trial
Blinding of personnel (performance bias)	Low risk	Manuscript states that all ICU staff involved in the care of participants (research co-ordinator, bedside nurse, treating intensivist, study investigators) were blinded to study arm assignment However 2 ICU nurses were unblinded to treatment assignment, so as to perform the necessary checking of blood products ahead of a transfusion. Trialists took steps to measure whether study arm assignment was revealed by these nurses Investigators in this study measured no subjective outcomes
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	The trial does not state who undertook the outcome assessment, so information is insufficient to permit assessment of risk of detection bias
Incomplete outcome data (attrition bias) All outcomes	Low risk	One enrolled participant declined to consent. All randomly assigned participants were included in the outcome analyses

Selective reporting (reporting bias)	Low risk	All outcomes outlined in the Methods section were reported in the Results section
Other bias	Low risk	We have no concerns about other potential sources of bias

Bennett-Guerrero 2009 [1]

Methods	<p>Parallel-group, randomised controlled trial, single-centre study</p> <p>Randomly assigned at: start of the surgical process</p> <p>Sample size calculation: none reported</p> <p>Duration of follow-up: to postoperative day 30</p>
Participants	<p>Inclusion criteria: aged > 18 years; high morbidity and mortality risk defined as a preoperative Parsonnet risk score > 15 and scheduled for non-emergent CABG and/or cardiac valve repair or replacement surgery using CPB</p> <p>Exclusion criteria: patient refusal of red blood cell products; haemodialysis or peritoneal dialysis within 7 days before surgery; planned significant concomitant procedure; planned avoidance of CPB or a minimally invasive surgical approach; planned use of an alternative to heparinisation; (scheduled) participation in an experimental drug or device trial within 30 days of study entry; history of significant abnormality or red blood cell disorders; positive antibody screen; known allergic reactivity to blood products requiring washed red blood cells; significant immunocompromise and patients with blood type 0 (due to insufficient quantities of middle-aged or old O- units)</p> <p>Transfusion indication: support during acute illness</p>
Interventions	<p>Comparisons: fresher red blood cell transfusion vs an intervention arm with actively transfused older red blood cells</p> <p>Intervention: mean (SD) 7 (4) days</p> <p>Comparator: mean (SD) 21 (4) days</p> <p>Number of participants enrolled: 845</p> <p>Number of participants randomly assigned to the interventions: 12/11</p> <p>Numbers completing assessment: 12/11</p> <p>Compliance with assigned treatments: Nothing is stated in the text about compliance with assigned interventions. All participants in this trial underwent surgery, and all were included in the feasibility analysis. 20 (87%) of participants received at least 1 unit of red blood cells; 20 (87%) of participants were excluded from the organ injury analysis</p>
Outcomes	<p>Primary: feasibility trial</p> <ul style="list-style-type: none"> Primary endpoint was whether more than 90% of participants in a particular arm received all red blood cell units within the specified age window <p>Secondary</p> <ul style="list-style-type: none"> Duration of storage of red blood cells received by each participant Comparison of duration of storage in study arms Mortality (at postoperative day 30) Length of ICU and hospital stay (hours) Duration of mechanical ventilation (hours) Measures of organ dysfunction/injury by PaO₂/FiO₂ ratio (at 2 hours post

	operation) and lactic acid (at 2 hours post operation)	
Notes	Population nationality: American Sources of funding: Duke University internal funds Declarations of interest (among primary researchers): Manuscript reports “None” Dates the study was conducted: conducted over a 15-month period, but no start and end dates reported	
<i>Risk of bias</i>		
Bias	Authors’ judgement	Support for judgement
Random sequence generation (selection bias)	Low risk	“Computerised randomisation schedule that was stratified by the attending surgeon and maintained by the hospital’s transfusion service” - page 1376
Allocation concealment (selection bias)	Low risk	“Computerised randomisation schedule that was stratified by the attending surgeon and maintained by the hospital’s transfusion service” - page 1376
Blinding of participants (performance bias)	Low risk	“Red blood cells units provided by the hospital’s transfusion service in a blinded manner from induction of general anaesthesia to post-operative day 7 (page 1376) such that patients, care providers, study personnel and investigators were blinded to the study group assignment” “To maintain blinding, the transfusion service obscured the expiration date of red blood cell [red blood cell] units with a removable sticker that contained the institutional review board number and study contact information in case a clinician had any questions” (page 1376) Investigators in this study measured no subjective outcomes
Blinding or personnel (performance bias)	Low risk	“Red blood cell units provided by the hospital’s transfusion service in a blinded manner from induction of general anaesthesia to post-operative day 7 (page 1376) such that patients, care providers, study personnel and investigators were blinded to the study group assignment” “To maintain blinding, the transfusion service obscured the expiration date of red blood cell units with a removable sticker

Bennett-Guerrero 2009 [1] (*Continued*)

		that contained the institutional review board number and study contact information in case a clinician had any questions" (page 1376)
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	The trial does not state who undertook the outcome assessment, so information is insufficient to permit assessment of risk of detection bias
Incomplete outcome data (attrition bias) All outcomes	Low risk	All randomly assigned participants were included in all of the outcome analyses for organ injury analysis. Three participants (no details were reported as to which intervention arm) were excluded from the organ injury analysis
Selective reporting (reporting bias)	Low risk	Outcome data of interest to this review were reported in supplemental tables. All assessments detailed in the Methods section have been reported in the Results section and in supplemental tables
Other bias	Low risk	We have no concerns about other potential sources of bias

Bennett-Guerrero 2009 [2]

Methods	Parallel-group, randomised controlled trial, single-centre study Randomly assigned at: start of the surgical process Sample size calculation: none reported Duration of follow-up: to postoperative day 30
Participants	Inclusion criteria: aged > 18 years; high morbidity and mortality risk defined as a preoperative Parsonnet risk score > 15 and scheduled for non-emergent CABG and/or cardiac valve repair or replacement surgery using CPB Exclusion criteria: patient refusal of red blood cell products; haemodialysis or peritoneal dialysis within 7 days before surgery; planned significant concomitant procedure; planned avoidance of CPB or a minimally invasive surgical approach; planned use of an alternative to heparinisation; (scheduled) participation in an experimental drug or device trial within 30 days of study entry; history of significant abnormality or red blood cell disorders; positive antibody screen; known allergic reactivity to blood products requiring washed red blood cells; significant immunocompromise and patients with blood type 0 (due to insufficient quantities of middle-aged or old 0- units) Transfusion indication: support during acute illness
Interventions	Comparisons: fresher red blood cell transfusion vs current standard practice Intervention: less than 21 days Comparator: unit standard of care = oldest red blood cell units out first

	Number of participants enrolled: 845 Number of participants randomly assigned to the interventions: not stated; 22 were randomly assigned but surgery was cancelled in 2 participants Numbers completing assessment: 9/11 Compliance with assigned treatments: Nothing is stated in the text about compliance with assigned interventions. All participants in this trial underwent surgery, and all were included in the feasibility and organ injury analysis. 18 (90%) participants received at least 1 unit of red blood cells	
Outcomes	Primary: feasibility trial <ul style="list-style-type: none">Primary endpoint was the expected difference in the percentage of participants in each arm who received at least 1 unit of red blood cells of at least 31 days' storage duration Secondary <ul style="list-style-type: none">Duration of storage of red blood cells received by each participantComparison of duration of storage in study armsMortality (at postoperative day 30)Length of ICU and hospital stay (hours)Duration of mechanical ventilation (hours)Measures of organ dysfunction/injury by PaO2/FiO2 ratio (at 2 hours post operation) and lactic acid (at 2 hours post operation)	
Notes	Population nationality: American Sources of funding: Duke University (North Carolina) internal funds Declarations of interest (among primary researchers): Manuscript reports "None" Dates the study was conducted: conducted over a 15-month period, but no start and end dates reported	
<i>Risk of bias</i>		
Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Low risk	"Computerised randomisation schedule that was stratified by the attending surgeon and maintained by the hospital's transfusion service" - page 1376
Allocation concealment (selection bias)	Low risk	"Computerised randomisation schedule that was stratified by the attending surgeon and maintained by the hospital's transfusion service" - page 1376
Blinding of participants (performance bias)	Low risk	"Red blood cell units provided by the hospital's transfusion service in a blinded manner from induction of general anaesthesia to post-operative day 7 (page 1376) such that patients, care providers, study personnel and investigators were blinded to the study group assignment"

		<p>“To maintain blinding, the transfusion service obscured the expiration date of red blood cell units with a removable sticker that contained the institutional review board number and study contact information in case a clinician had any questions” (page 1376)</p> <p>Investigators in this study measured no subjective outcomes</p>
Blinding or personnel (performance bias)	Low risk	<p>“Red blood cell units provided by the hospital’s transfusion service in a blinded manner from induction of general anaesthesia to post-operative day 7 (page 1376) such that patients, care providers, study personnel and investigators were blinded to the study group assignment”</p> <p>“To maintain blinding, the transfusion service obscured the expiration date of red blood cells units with a removable sticker that contained the institutional review board number and study contact information in case a clinician had any questions” (page 1376)</p>
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	The trial does not state who undertook the outcome assessment, so information is insufficient to permit assessment of risk of detection bias
Incomplete outcome data (attrition bias) All outcomes	Low risk	22 participants were randomly assigned into the study. Surgery was cancelled for 2 participants (it is unclear what intervention arm they had been randomly assigned to). Only 20 participants received a red blood cell transfusion and were included in the outcome analyses
Selective reporting (reporting bias)	Low risk	Outcome data of interest to this review were reported in supplemental tables. All assessments detailed in the Methods section were reported in the Results section and in supplemental tables
Other bias	Low risk	We have no concerns about other potential sources of bias

Dhabangi 2013

Methods	Parallel-group, randomised controlled trial, single-centre study Randomly assigned at: point of transfusion Sample size calculation: none reported Duration of follow-up: 24 hours
Participants	Inclusion criteria: positive blood smear for malaria; severe anaemia (Hb < 5 g/dL); lactic acidosis (blood lactate > 5 mmol/L); written informed consent from parent or guardian Exclusion criteria: known or concurrent cardiac disease; undergoing transfusion with blood products other than packed red blood cells Transfusion indication: support during acute illness
Interventions	Comparison: fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells Intervention: aged 1 to 10 days Comparator: aged 21 to 35 days Number of participants enrolled: 250 Number of participants randomly assigned to the interventions: 37/37 Numbers completing assessment: 37/37 Compliance with assigned treatments: Nothing is stated in the text about compliance with assigned interventions
Outcomes	Primary: feasibility trial <ul style="list-style-type: none"> Proportion of transfused children who resolved lactic acidosis by 4 hours from the start of transfusion Secondary <ul style="list-style-type: none"> None declared, but in-hospital mortality was reported (3 hours after transfusion)
Notes	Population nationality: Ugandan Source of funding: Canadian Institute for Health Research (MCT 75527) Declarations of interest (among primary researchers): "The authors declare that they have no competing interests" Dates the study was conducted: December 2010 to August 2011 This is a feasibility study for 'TOTAL' (RBC (red blood cell) Transfusion in Severe Anemia With Lactic Acidosis). The TOTAL trial is an ongoing trial listed in this review under NCT01586923

Risk of bias

Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Unclear risk	Information about the sequence generation process was insufficient to permit assessment of risk of selection bias. The study reports that "eligible children were randomised using sealed envelopes"
Allocation concealment (selection bias)	Unclear risk	Information was insufficient to permit assessment of risk of selection bias The only information reported was in a

Dhabangi 2013 (Continued)

		'sealed envelope'. No further details such as whether the envelopes were sequentially numbered and opaque are reported
Blinding of participants (performance bias)	Unclear risk	Information was insufficient to permit assessment of risk of performance bias
Blinding or personnel (performance bias)	Unclear risk	Information was insufficient to permit assessment of risk of performance bias
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	Information was insufficient to permit assessment of risk of detection bias
Incomplete outcome data (attrition bias) All outcomes	Low risk	No outcome data were missing
Selective reporting (reporting bias)	Low risk	All outcomes outlined in the Methods section were reported in the Results section
Other bias	Low risk	We have no concerns about other potential sources of bias

Fergusson 2012

Methods	Parallel-group, randomised controlled trial, multi-centre study Randomly assigned at: point of transfusion Sample size calculation: yes Duration of follow-up: 90 days
Participants	Inclusion criteria: admitted to neonatal ICU and consent form signed by parents; birth weight < 1250 g and required 1 or more red blood cell transfusions for anaemia Exclusion criteria: Premature infants who had already received red blood cells and were scheduled to receive an exchange transfusion or a directed donation; had a rare blood type; were moribund on admission to neonatal ICU; were not expected to survive because of a severe congenital abnormality; whose attending clinician team specifically requested fresh red blood cells and whose families were experiencing mitigating circumstances Transfusion indication: support during acute illness
Interventions	Comparison: fresher red blood cell transfusion vs current standard practice Intervention: aged ≤ 7 days Comparator: range 2 to 42 days. In this group, units were divided into aliquots to increase usage and reduce waste. Each aliquot was designated for use in a single infant up to its expiry date Number of participants enrolled: 1752 Number of participants randomly assigned to the interventions: 188/189 Numbers completing assessment: 188/189 Compliance with assigned treatments: Manuscript reports a blood bank non-compli-

	ance rate less than 4%, identified upon data safety monitoring board first interim analysis (page 1446/7)	
Outcomes	<p>Primary</p> <ul style="list-style-type: none">• Necrotising enterocolitis (Bell criteria stage ≥ 2)• Intraventricular haemorrhage (Papile criteria grade ≥ 3)• Retinopathy of prematurity (stage ≥ 3)• Bronchopulmonary dysplasia (defined as oxygen dependency for at least 28 days at 36 weeks' postmenstrual age) <p>• Death</p> <p>• Composite of any of the above</p> <p>These complications had to occur after receipt of the initial transfusion to up to 90 days' neonatal ICU stay to be recorded as a primary outcome. Data were presented for each individual outcome as well as for composite outcomes. For this review, we reported and used just individual outcome data</p> <p>Secondary</p> <ul style="list-style-type: none">• Rates of individual complications• Rates of nosocomial infections (clinically suspected and positively cultured)• Length of mechanical ventilation and supplemental oxygen use• Need for vasopressors, other blood products and invasive vascular access• Length of stay in neonatal ICU (days)• Rates of minor (laser therapy, tracheostomy and endoscopic procedures) and major (including all major surgical procedures) interventions <p>These complications had to occur after receipt of the initial transfusion to up to 90 days' neonatal ICU stay to be measured as a secondary outcome. Data were presented for each individual outcome and for composite outcomes. For this review, we reported and used just individual outcome data</p>	
Notes		
<i>Risk of bias</i>		
Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Low risk	"Interactive voice response system whereby following eligibility screening by the research co-ordinator, the system generated a unique number, the research co-ordinator then telephoned the hospital blood bank staff and reported the number. In turn the blood bank staff referred to a manual of unique numbers generated by an independent statistician prior to study activation to determine the study intervention allocated to the randomised patient" (page 1445) Schedule stratified by site in variable blocks of 6 and 8

Allocation concealment (selection bias)	Low risk	This was undertaken and managed outside the control of the treating clinician Allocation occurred only after an order to transfuse was received, and only if a supply of red blood cells stored for 7 days or less was available at the time of allocation
Blinding of participants (performance bias)	Low risk	Participants were neonates, and the manuscript reports that "Infants' families were blinded to treatment allocation" (page 1445)
Blinding of personnel (performance bias)	Low risk	Manuscript reports that "study investigators, research co-ordinators (and) attending care teams were blinded to treatment allocation" (page 1445) Study protocol states that each blood bag was issued with a label that could not be altered but did bear details of the expiry date of the unit. Therefore it is possible that the clinician checking and transfusing the unit would be unblinded to treatment allocation. But once the unit was transfused, all paperwork in the notes did not bear expiry date details. Study authors believe that information bias was limited by the methods above
Blinding of outcome assessment (detection bias) All outcomes	Low risk	Manuscript reports that "individual elements of the composite outcome were adjudicated independently by 2 neonatologists blinded to the study group allocation" (page 1445)
Incomplete outcome data (attrition bias) All outcomes	Low risk	No outcome data were missing
Selective reporting (reporting bias)	Low risk	Study protocol is available, and all of the study's prespecified outcomes that are of interest in this systematic review have been reported in the prespecified way
Other bias	Low risk	We have no concerns about other potential sources of bias

Fernandes 2005

Methods	Parallel-group, randomised controlled trial, single-centre study Randomly assigned at: point of transfusion Sample size calculation: yes Duration of follow-up: not stated	
Participants	Inclusion criteria: gestational age < 37 weeks; birth weight < 1500 g; received at least 1 red blood cell transfusion during hospital stay and parents signed consent form Exclusion criteria: major congenital malformations; haemolytic anaemia of any aetiology; total or partial exchange transfusion during hospitalisation; requiring rapid infusion of packed red blood cells for haemorrhage and requiring more than 1 red blood cell transfusion within 24 hours Transfusion indication: support during acute illness	
Interventions	Comparison: fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells Intervention: red blood cells < 3 days old Comparator: red blood cells up to 28 days old Number of participants enrolled: 108 Number of participants randomly assigned to the interventions: 26/26 Numbers completing assessment: 26/26 Compliance with assigned treatments: Nothing is stated in the text about compliance with assigned interventions	
Outcomes	Primary: number of donor exposures. Secondary: none specifically stated, but the following outcomes were reported <ul style="list-style-type: none">• Duration of mechanical ventilation (days)• Length of hospital stay (days)• Death (up to 40 weeks' postconceptual age)• Incidence of (in-hospital) infection: clinical sepsis and necrotising enterocolitis	
Notes	Population nationality: Brazilian Sources of funding: Fundo de Auxilio aos Docentes e Alunos da Universidade Federal de Sao Paulo, Brazil Declarations of interest (among primary researchers): none reported Dates the study was conducted: 15 May 2002 to 15 December 2003	
<i>Risk of bias</i>		
Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Unclear risk	Information about the sequence generation process was insufficient to permit assessment of risk of selection bias
Allocation concealment (selection bias)	Unclear risk	Information about the sequence generation process was insufficient to permit assessment of risk of selection bias

Blinding of participants (performance bias)	Unclear risk	Information was insufficient to permit assessment of risk of performance bias
Blinding of personnel (performance bias)	Unclear risk	Information was insufficient to permit assessment of risk of performance bias
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	Information was insufficient to permit assessment of risk of detection bias
Incomplete outcome data (attrition bias) All outcomes	Low risk	No outcome data were missing
Selective reporting (reporting bias)	Low risk	All outcomes defined in the Methods section were in the Results section
Other bias	Low risk	We have no concerns about other potential sources of bias

Hebert 2005

Methods	<p>Parallel-group, randomised controlled trial, multi-centre study</p> <p>Randomly assigned at: point of transfusion</p> <p>Sample size calculation: yes</p> <p>Duration of follow-up: duration of hospital stay (up to 30 days)</p>
Participants	<p>Inclusion criteria: will require at least 1 unit of allogeneic red blood cells; accepted invitation to participate in the study and to undergo elective or urgent cardiac surgical procedures or admitted to an ICU</p> <p>Exclusion criteria: aged < 16 years; enrolment in another interventional study; previous enrolment in this study; have a terminal illness with a life expectancy < 3 months; not expected to survive longer than 12 hours post ICU admission; not provided with all required therapeutic interventions to sustain life; unable to receive blood products because of difficulties with cross-matching or a history of unexplained severe transfusion reaction; receipt of a blood transfusion in the 6 months before enrolment; had undergone a bone marrow transplant; had a malignant haematological disease within a year of study enrolment or was a heart transplant or artificial heart recipient</p> <p>Transfusion indication: not stated</p>
Interventions	<p>Comparison: fresher red blood cell transfusion vs current standard practice</p> <p>Intervention: aged < 8 days old</p> <p>Comparator: This group received red blood cells issued from the hospital blood bank with the longest storage time in accordance with standard blood bank procedure. To ensure maximum age separation between groups, participants were allocated only on days when the average age of red blood cells in the blood bank exceeded 15 days</p> <p>Number of participants enrolled: not stated</p> <p>Number of participants randomly assigned to the interventions: 33/33</p> <p>Numbers completing assessment: 26/31</p>

	Compliance with assigned treatments: some non-compliance: 2 participants in the 'fresher' intervention arm were non-compliant with randomisation as they did not receive red blood cells less than 8 days old. These participants were excluded from outcome analyses Another 7 participants (5 in the 'fresher' intervention arm and 2 in the 'standard practice' intervention arm) did not receive any transfusions Also reported was compliance with allocated red blood cell storage times. Compliance target was 90%. This was attained in 91% of participants randomly assigned to the 'fresher' intervention arm and in 59% of participants randomly assigned to the 'standard practice' intervention arm	
Outcomes	Primary: feasibility trial <ul style="list-style-type: none">Composite outcome of hospital mortality, serious nosocomial infections (pneumonia, surgical wound infections including mediastinitis and osteomyelitis and bacteraemia from organisms not considered normal skin flora) and thrombotic events including myocardial infarction and acute ischaemic stroke Secondary <ul style="list-style-type: none">Composite outcome of number of organ failures combined with death (assessed using score by Hebert 1993)Intensity of interventions measured by (1) respiratory support (number of days on mechanical ventilation); (2) intensity of cardiovascular support (number of days requiring vasoactive drugs, an aortic balloon pump or a ventricular assist device); (3) Intensity of renal support (number of days of renal replacement therapy)Vital organ support measured by (1) prolonged invasive mechanical ventilation (requirement for invasive mechanical ventilation once randomly assigned for longer than 48 hours); (2) prolonged mechanical ventilation (requirement for invasive and non-invasive mechanical ventilation once randomly assigned for longer than 48 hours); (3) prolonged low cardiac output (requirement for vasoactive drugs, left ventricular assist device or aortic balloon pump for longer than 48 hours)Mortality at 30-day and 90-day follow-up Investigators measured outcomes over the duration of the hospital stay	
Notes	Population nationality: Canadian Sources of funding: not stated Declarations of interest (among primary researchers): none reported Dates the study was conducted: 28 October 1999 to 31 May 2001 This is a feasibility study for the 'Age of Blood Evaluation [ABLE]' trial. The ABLE trial is a trial awaiting assessment, listed in this review as Lacroix 2011	
Risk of bias		
Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Low risk	The randomisation process was started by a telephone request from the attending team for a red blood cell transfusion. The randomisation schedule consisted of a computer-generated random listing of the 2 treatment allocations blocked into groups

		of 4 and 6 and stratified by centre and categories of cardiac surgery or critical care
Allocation concealment (selection bias)	Low risk	As above, on receipt of the phone call, a blood bank technologist not involved in clinical care opened a sequentially numbered sealed opaque envelope
Blinding of participants (performance bias)	Low risk	Few details were given, but as participants were adults and the trial did not measure subjective outcomes, we have judged blinding of participants to be at low risk of bias
Blinding of personnel (performance bias)	Unclear risk	No further details were given on blinding than were reported for participants; therefore risk of performance bias is unclear because information was insufficient for assessment
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	No further details were given on blinding than were reported for participants; therefore risk of detection bias is unclear because information was insufficient for assessment
Incomplete outcome data (attrition bias) All outcomes	High risk	Outcome data were incomplete for participants in the analysis of 30-day and 90-day follow-up mortality outcomes All participants who received red blood cells of correct age based on the intervention group they were randomly assigned to were included in the analysis of mortality rates in the intensive care unit and in hospital ('fresher', n = 26; 'standard practice, n = 31). At 30-day and 90-day follow-up assessment, the numbers included in the intervention arms were (at 30 days: 'fresher', n = 24; 'standard practice, n = 29; at 90 days; 'fresher', n = 20; 'standard practice, n = 29). No details were reported as to what happened to the missing participants at 30-day and 90-day follow-up
Selective reporting (reporting bias)	Low risk	All outcomes defined in the Methods section were reported in the Results section
Other bias	Low risk	We have no concerns about other potential sources of bias

Heddle 2012

Methods	Parallel-group, randomised controlled trial, single-centre study Randomly assigned at: point of transfusion Sample size calculation: no Duration of follow-up: “until discharge or 31 January 2011 at which time they were censored”	
Participants	Inclusion criteria: requirement for a blood transfusion Exclusion criteria: medical indication for fresh blood (e.g. sickle cell disease); scheduled to receive directed or autologous donations; expected to receive massive blood transfusion (defined as > 10 units of blood transfused within 24 hours); received a transfusion as an outpatient Transfusion indication: support during an acute illness	
Interventions	Comparison: fresher red blood cell transfusion vs current standard practice Intervention: “freshest available” Comparator: “oldest in the inventory” Number of participants enrolled: 1129 Number of participants randomly assigned to the interventions: 309/601 Numbers completing assessment: 309/601 Compliance with assigned treatments: 5% (n= 46) of participants did not receive red blood cell transfusion quite as intended: 19 received a massive transfusion; 19 received transfusions before the study was implemented and 8 were transfused in a day surgery clinic with same-day discharge. All were included in the outcome analyses	
Outcomes	Primary: feasibility trial <ul style="list-style-type: none">• Enrolment of at least 80% of eligible hospitalised participants over the study period• Minimum of 10 days’ contrast in the mean maximum age of blood between treatment arms• 100% compliance with generating weekly reports on accrual, blood bank inventory and age distribution of transfused blood per treatment arm• Blood outdate rate below the current rate of 25% Secondary <ul style="list-style-type: none">• In-hospital mortality• Quantity of blood products transfused	
Notes	Population nationality: Canadian Sources of funding: Canadian Institutes for Health Research, Canadian Blood Services and Health Canada Declarations of interest (among primary researchers): “The authors have no conflicts of interest to disclose” Dates the study was conducted: 14 June 2010 to 19 December 2010 This is a feasibility study for the ‘Informing Fresh versus Old red cell Management [INFORM]’ trial	
Risk of bias		
Bias	Authors’ judgement	Support for judgement

Random sequence generation (selection bias)	Low risk	<p>“Computer generated random allocation schedule was used to assign [participants] 1:2 to freshest available or standard issue blood” (page 1204)</p> <p>(The unbalanced randomisation was to maintain a contrast of at least 10 days in the mean maximum age of transfused red blood cells between the two treatment groups and to avoid outdating of red blood cells)</p>
Allocation concealment (selection bias)	High risk	<p>Manuscript reports that for the first 24 weeks of the study, “the randomisation was performed using an unconcealed paper-based sequence held in the blood bank” (page 1204). Manuscript further reports that in the (additional) last 3 weeks of the trial, allocation was concealed through a secure website (thus low risk of selection bias). However as allocation was unconcealed over most of the trial, this dimension has been judged to be at high risk of bias overall</p>
Blinding of participants (performance bias)	Low risk	<p>Participants were not blinded, but as outcomes were objective, we believe that such knowledge would not have had an effect on outcome assessment, hence the judgement of low risk of bias</p>
Blinding of personnel (performance bias)	Low risk	<p>Study personnel were not blinded, but as outcomes were objective, we believe that such knowledge would not have had an effect on outcome assessment, hence the judgement of low risk of bias</p>
Blinding of outcome assessment (detection bias) All outcomes	Low risk	<p>Outcome assessors were not blinded, but as outcomes were objective, we believe that such knowledge would not have had an effect on outcome assessment, hence the judgement of low risk of bias</p>
Incomplete outcome data (attrition bias) All outcomes	Low risk	<p>No outcome data were missing</p>
Selective reporting (reporting bias)	Low risk	<p>All outcomes outlined in the Methods section were reported in the Results section</p>

Other bias	Low risk	We have no concerns about other potential sources of bias
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Kor 2012

Methods	<p>Parallel-group, randomised controlled trial, single-centre study</p> <p>Randomly assigned at: point of transfusion</p> <p>Sample size calculation: yes</p> <p>Duration of follow-up: outcome dependent over a 48-hour interval post red blood cell transfusion or for mortality at hospital discharge</p>
Participants	<p>Inclusion criteria: endotracheally intubated; mechanically ventilated ICU participants; arterial access in situ; haemoglobin concentration below 9.5 g/dL</p> <p>Exclusion criteria: concurrent transfusion with another blood product; emergency transfusion and/or haemodynamic instability as defined by upward titration of vasoactive medications in the 2-hour interval between red blood cell transfusions</p> <p>Transfusion indication: support during acute illness</p>
Interventions	<p>Comparison: fresher red blood cell transfusion vs current standard practice</p> <p>Intervention: single unit of fresh red blood cells < 5 days old. Subsequent transfusions (after first study transfusion) were standard issue</p> <p>Comparator: single unit of standard issue red blood cells (median = 21 days)</p> <p>Number of participants enrolled: 665</p> <p>Number of participants randomly assigned to the interventions: 50/50</p> <p>Numbers completing assessment: 49/50</p> <p>Compliance with assigned treatments: 1 participant randomly assigned to the study was excluded before administration of a red blood cell transfusion because an appropriately cross-matched fresh unit of red blood cells could not be found</p>
Outcomes	<p>Primary</p> <ul style="list-style-type: none"> Change in pulmonary gas exchange as assessed by ratio of partial pressure of arterial oxygen to fraction of inspired oxygen concentration* <p>Secondary</p> <ul style="list-style-type: none"> Changes in pulmonary function (post-transfusion changes in peak and plateau airway pressures, static and dynamic respiratory system compliance and fraction of dead space ventilation (Vd/V): both of the latter measured with the NICO cardiopulmonary management system)* Changes in markers of immune status (including tumour necrosis factor-α, IL-8 and C-reactive protein)* Coagulation status (platelet count, fibrinogen concentration and antithrombin consumption)* New or worsening acute lung injury over a 48-hour period after red blood cell transfusion Changes in organ failure over a 48-hour period after red blood cell transfusion Mortality (at hospital discharge) <p>* Changes measured between baseline (measurements were obtained in the 2-hour interval before transfusion) and post transfusion (on completion of the intervention of red blood cell transfusion and within 2 hours of initiation of the intervention of red blood</p>

	cell transfusion); mean (SD) in the fresh intervention arm was 1.8 (0.46) hours and in the comparator arm was 1.9 (0.63) hours	
Notes	Population nationality: American Sources of funding: National Institutes of Health grant and Department of Critical Care Medicine, Mayo Clinic College of Medicine Declarations of interest (among primary researchers): All authors declared received NIH grant award monies Dates the study was conducted: June 2008 to May 2010	
<i>Risk of bias</i>		
Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Low risk	“simple, computer-generated list” (page 844)
Allocation concealment (selection bias)	High risk	Manuscript reports that treatment allocation was determined by personnel within the blood bank at the time of red blood cell issue by opening ‘sealed, opaque envelopes’ (page 844). No details are given as to whether the envelopes were sequentially numbered, hence assessment of high risk of bias for allocation concealment
Blinding of participants (performance bias)	Low risk	Manuscript reports that participants were blinded to treatment allocation. Moreover participants were mechanically ventilated when transfused whilst in the trial, so knowledge of treatment allocation does not seem important, hence low risk of bias
Blinding or personnel (performance bias)	Low risk	Trial states that all investigators were blinded to treatment allocation status for the duration of the study procedures, as the clinical service responsible for ordering the blood product was not involved in administration of the red blood cell transfusion. Manuscript further states that the product expiration date was not concealed during the transfusion process, so there would have been the potential to work out a participant’s allocation using the expiration date. However we believe that working out a participant’s allocation by red blood cell unit expiration date is unlikely, and that as all outcomes are objective and most were measured while a participant was ventilated, the

		risk of performance bias is low
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	Trial does not state who undertook the outcome assessment, so information is insufficient to permit assessment of risk of detection bias
Incomplete outcome data (attrition bias) All outcomes	Low risk	All participants who received the study intervention were included in the outcome analyses
Selective reporting (reporting bias)	Low risk	All outcomes outlined in the Methods section were reported in the Results section
Other bias	Low risk	We have no concerns about other potential sources of bias

Liu 1994

Methods	Parallel-group, randomised controlled trial, multi-centre study Randomly assigned at: point of transfusion Sample size calculation: yes Duration of follow-up: 24 ± 6 hours post transfusion
Participants	Inclusion criteria: infants weighing < 1.5 kg at the time of first or second blood transfusion Exclusion criteria: An exchange transfusion was anticipated more than 5 days after the first transfusion; active bleeding or coagulopathy; renal failure; very premature sick neonates not expected to survive longer than 48 hours; not expected to need more than 1 transfusion Transfusion indication: support during acute illness
Interventions	Comparison: fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells Intervention: quint packs < 5 days old. When the need for multiple transfusions was anticipated, attempts were made to limit donor exposures by reserving multiple quint packs from the same donor for a participant Comparator: assigned a single adult unit of packed red blood cells, which was reserved for that infant up to the recommended storage time of 35 days All transfusions were delivered by syringe pump during a 4-hour period Number of participants enrolled: not stated Number of participants randomly assigned to the interventions: 12/13 Numbers completing assessment: 12/13 Compliance with assigned treatments: Nothing is stated in the text about compliance with assigned interventions
Outcomes	Primary ● Not stated

	Secondary <ul style="list-style-type: none">• Change in haematocrit (before and 24 (+/-6) hours after the transfusion)• Change in whole blood potassium (K+) concentration pH and base excess (immediately before and after transfusion)• Volume of red blood cell transfusion	
Notes	Population nationality: American Sources of funding: none stated Declarations of interest (among primary researchers): none reported Dates the study was conducted: not stated	
<i>Risk of bias</i>		
Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Low risk	"Random numbers table"
Allocation concealment (selection bias)	Unclear risk	Information about the sequence generation process was insufficient to permit assessment of risk of selection bias
Blinding of participants (performance bias)	Unclear risk	Information was insufficient to permit assessment of risk of performance bias
Blinding or personnel (performance bias)	Unclear risk	Information was insufficient to permit assessment of risk of performance bias
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	Information was insufficient to permit assessment of risk of detection bias
Incomplete outcome data (attrition bias) All outcomes	Low risk	No outcome data were missing
Selective reporting (reporting bias)	Low risk	All outcomes outlined in the Methods section were reported in the Results section
Other bias	Low risk	We have no concerns about other potential sources of bias

Neuman 2013

Methods	Parallel-group, randomised controlled trial Randomly assigned at: point of transfusion Sample size calculation: none reported Duration of follow-up: 24 hours
Participants	Inclusion criteria: hospital in-patients receiving cross-matched packed red blood cells for clinical indications Exclusion criteria: not stated Transfusion indication: support during acute illness
Interventions	Comparison: fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells Intervention: aged < 10 days Comparator: aged > 21 days Number of participants enrolled: 44 Number of participants randomly assigned to the interventions: 20/24 Numbers completing assessment: 20/24 Compliance with assigned treatments: Nothing is stated in the text about compliance with assigned interventions
Outcomes	Primary <ul style="list-style-type: none">• Not stated Secondary <ul style="list-style-type: none">• Plasma nitrite (NO₂) levels measured via high-performance liquid chromatography• Vascular endothelial function measured using flow-mediated dilation of the brachial artery before (1 hour) and after (24 hours) red blood cell transfusion
Notes	Population nationality: American Source of funding: not stated Declarations of interest (among primary researchers): "The authors declare that they have nothing to disclose" Dates the study was conducted: not stated

Risk of bias

Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Unclear risk	Information was insufficient to assess whether an important risk of selection bias exists
Allocation concealment (selection bias)	Unclear risk	Information was insufficient to assess whether an important risk of selection bias exists
Blinding of participants (performance bias)	Unclear risk	Information was insufficient to assess whether an important risk of performance bias exists

Neuman 2013 (Continued)

Blinding or personnel (performance bias)	Unclear risk	Information was insufficient to assess whether an important risk of performance bias exists
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	Information was insufficient to assess whether an important risk of detection bias exists
Incomplete outcome data (attrition bias) All outcomes	Unclear risk	No outcome data were missing
Selective reporting (reporting bias)	Unclear risk	All outcomes defined in the Methods section were reported in the Results section
Other bias	Low risk	We have no concerns about other potential sources of bias

Schulman 2002

Methods	Parallel-group, randomised controlled trial, single-centre study Randomly assigned at: point of transfusion but only in the blood bank was at least 15 units of young and old blood available Sample size calculation: no Duration of follow-up: not stated
Participants	Inclusion criteria: admission to level 1 trauma centre; patients with blood type A only Exclusion criteria: not stated Transfusion indication: support during acute illness Patients were considered for analysis only if 2 or more units of type-specific blood was transfused
Interventions	Comparison: fresher red blood cell transfusion vs current standard practice Intervention: aged < 11 days Comparator: aged > 20 days Number of participants enrolled: not stated Number of participants randomly assigned to the interventions: 8/9 Numbers completing assessment: 8/9 Compliance with assigned treatments: Nothing is stated in the text about compliance with assigned interventions
Outcomes	Mortality
Notes	This is noted as a pilot trial and was published in a letter only: no full publication for the trial can be found Population nationality: American Sources of funding: none stated Declarations of interest (among primary researchers): none reported Dates the study was conducted: August 2000 to July 2001

<i>Risk of bias</i>		
Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Unclear risk	Information was insufficient to assess whether an important risk of selection bias exists
Allocation concealment (selection bias)	Unclear risk	Information was insufficient to assess whether an important risk of selection bias exists
Blinding of participants (performance bias)	Unclear risk	Information was insufficient to assess whether an important risk of performance bias exists
Blinding of personnel (performance bias)	Unclear risk	Information was insufficient to assess whether an important risk of performance bias exists
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	Information was insufficient to assess whether an important risk of detection bias exists
Incomplete outcome data (attrition bias) All outcomes	Unclear risk	Information was insufficient to assess whether an important risk of attrition bias exists
Selective reporting (reporting bias)	Unclear risk	Information was insufficient to assess whether an important risk of reporting bias exists
Other bias	Low risk	We have no concerns about other potential sources of bias

Strauss 1996

Methods	Parallel-group, randomised controlled trial, single-centre study Randomly assigned at: point of transfusion Sample size calculation: yes Duration of follow-up: discharge before age 84 days, hospital day 84 or death before age 84 days
Participants	Inclusion criteria: very low birth weight infants (0.6 to 1.3 kg); admission to neonatal ICU Exclusion criteria: if the infant's survival or need for prolonged support seemed unlikely Transfusion indication: support during acute illness

Interventions	Comparison: fresher red blood cell transfusion vs current standard practice Intervention: aged < 7 days Comparator: aged < 42 days. From 1 dedicated donor who consented to donate a second unit when needed by the infant Number of participants enrolled: not stated Number of participants randomly assigned to the interventions: 21/19 Numbers completing assessment: for mortality, 21/19; for physiological markers of oxygen consumption or alterations in microcirculation and assessment of economic or blood stock inventory outcomes, 15/14 Compliance with assigned treatments: 29% (n = 6) of participants in the 'fresher' intervention arm and 26% (n = 5) of participants in the 'standard practice arm' did not receive any transfusions All participants randomly assigned to the 'fresher' arm received the allocated intervention: red blood cells < 7 days old	
Outcomes	Primary <ul style="list-style-type: none">Whether AS-1 red blood cells could safely satisfy all red blood cell needs of an individual very low birth weight infant Secondary <ul style="list-style-type: none">Assessment of potential risks, both clinical reactions and laboratory abnormalities (glucose, sodium, potassium and calcium) of transfusing stored AS-1 red blood cells (measured as change from pre to post (not further defined) transfusion)Mortality (to day 30)Hospital discharge < 84 daysNumber of neonates in hospital for 84 days	
Notes	Population nationality: American Sources of funding: Program Project Grant, a Clinical Research Centre Grant, National Institutes of Health Declarations of interest (among primary researchers): none reported Dates the study was conducted: not stated	
<i>Risk of bias</i>		
Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Unclear risk	Information was insufficient to assess whether an important risk of selection bias exists Manuscript reports that "envelopes arranged according to a table of random numbers" but provides no further details
Allocation concealment (selection bias)	Unclear risk	Information was insufficient to assess whether an important risk of selection bias exists Manuscript reports that "envelopes arranged according to a table of random

Strauss 1996 (Continued)

		numbers" but provides no further details, such as if the envelopes were sequentially numbered and opaque
Blinding of participants (performance bias)	Low risk	Participants were neonates and their parents were not informed as to the results of randomisation
Blinding of personnel (performance bias)	Low risk	Manuscript reports that the neonatal intensive care unit staff were not informed as to the results of random arm assignment
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	Information was insufficient to assess whether an important risk of detection bias exists
Incomplete outcome data (attrition bias) All outcomes	Low risk	All participants who received a red blood cell transfusion were included in the outcome analysis for all outcomes
Selective reporting (reporting bias)	Unclear risk	It is unclear what outcomes trialists wanted to measure; therefore information was insufficient to permit assessment of risk of selection bias
Other bias	Low risk	We have no concerns about other potential sources of bias

Strauss 2000

Methods	Parallel-group, randomised controlled trial, single-centre study Randomly assigned at: not stated Sample size calculation: yes Duration of follow-up: discharge before age 84 days, hospital day 84 or death before age 84 days
Participants	Inclusion criteria: premature infants weighing between 0.6 and 1.3 kg Exclusion criteria: not stated Transfusion indication: support during acute illness
Interventions	Comparison: fresher red blood cell transfusion vs current standard practice Intervention: aged < 7 days Comparator: aged < 42 days (red blood cell units from dedicated donors only) Number of participants enrolled: not stated Number of participants randomly assigned to the interventions: 10/11 Numbers completing assessment: 6/8 Compliance with assigned treatments: All participants randomly assigned to the 'fresher' arm received the allocated intervention, red blood cells < 7 days old

Outcomes	Primary: whether AS-3 red blood cells could safely satisfy all red blood cells needs of an individual very low birth weight infant Secondary: assessment of potential risks, both clinical reactions and laboratory abnormalities (glucose, sodium, potassium and calcium) of transfusing stored AS-1 red blood cells (measured as change from pre to post (not further defined) transfusion)	
Notes	Population nationality: American Sources of funding: Program Project Grant, a Clinical Research Centre Grant and a National Institutes of Health and Research Grant Declarations of interest (among primary researchers): none reported Dates the study was conducted: 1992 to 1994 Other: The trial was stopped before completion because of the Liu 1994 trial, which demonstrated the feasibility and safety of transfusing stored rather than fresh red blood cells to limit donor exposure of preterm infants who received transfusions	
<i>Risk of bias</i>		
Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Unclear risk	Information was insufficient to assess whether an important risk of selection bias exists Manuscript reports that “envelopes arranged according to a table of random numbers” but provides no further details
Allocation concealment (selection bias)	Unclear risk	Information was insufficient to assess whether an important risk of selection bias exists Manuscript reports that ”envelopes arranged according to a table of random numbers” but provides no further details, such as if the envelopes were sequentially numbered and opaque
Blinding of participants (performance bias)	Low risk	Participants were neonates, and their parents were not informed as to the results of randomisation
Blinding or personnel (performance bias)	Low risk	Manuscript reports that the neonatal intensive care unit staff were not informed as to the results of random arm assignment
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	Information was insufficient to assess whether an important risk of detection bias exists

Incomplete outcome data (attrition bias) All outcomes	Low risk	All participants who received a red blood transfusion were included in each outcome analysis Of note, the trial was stopped early “before completion because of a report (Liu 1994) that demonstrated the feasibility and safety of transfusing stored rather than fresh red blood cells to limit donor exposure of preterm infants who received transfusion” (page 216) Therefore the calculated sample size of 40 infants (20 per arm) was never reached. Instead the trial randomly assigned 22 participants (10 in the ‘fresher’ intervention arm and 11 in the ‘standard practice’ arm), 14 participants (6 participants from the ‘fresher’ arm and 8 from the ‘standard practice’ arm) who received a red blood cell transfusion and were included in the analysis of outcomes
Selective reporting (reporting bias)	Unclear risk	It is unclear what outcomes trialists wanted to measure; therefore information was insufficient to permit assessment of risk of selection bias
Other bias	Low risk	We have no concerns about other potential sources of bias

Walsh 2004

Methods	Parallel-group, randomised controlled trial, single-centre study Randomly assigned at: point of transfusion Sample size calculation: yes Duration of follow-up: a total of 10.5 hours (baseline = 2.5 hours; transfusion period = 3 hours; post-transfusion period = 5 hours)
Participants	Inclusion criteria: ICU physician caring for the patient had decided to transfuse the patient 2 units of red blood cells to increase haemoglobin concentration in the absence of clinical bleeding; transfusion could be deferred 12 to 18 hours to enable relatives’ assent to be sought when necessary; haemoglobin concentration at the time of screening was < 9 g/dL; patient had not received a red blood cell transfusion for at least 48 hours before baseline measurements were to start Exclusion criteria: presence of clinically apparent bleeding; contradiction to placement of a nasogastric tube; patient required frequent changes in respiratory or cardiovascular support because of physiological instability; patient not expected to survive > 48 hours; previous gastric surgery; postoperative liver transplant patient; age < 16 years; pregnancy Transfusion indication: support during acute illness

Interventions	Comparison: fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells Intervention: red blood cell units ≤ 5 days old Comparator: red blood cell units ≥ 20 days old Number of participants enrolled: 50 Number of participants randomly assigned to the interventions: 10/12 Numbers completing assessment: 10/12 Compliance with assigned treatments: Nothing is stated in the text about compliance with assigned interventions	
Outcomes	Primary: intragastric-arterial difference in PCO ₂ during and after red blood cell transfusion using air tonometry Secondary Change in indexes of tissue hypoxia and organ perfusion between pre and post transfusion (pretransfusion measures taken in the 2.5 hours before transfusion, and post-transfusion measures taken in the 5 hours post red blood cell transfusion. All of the following indexes were measured <ul style="list-style-type: none">• pHi• Arterial lactate concentration• PaCO₂• Arterial pH• Arterial base excess• Arterial haemoglobin concentration	
Notes	Population nationality: Scottish Sources of funding: Mason Medical Research Foundation, Effective Use of Blood Group of the Scottish National Blood Transfusion Service and the Royal Infirmary of Edinburgh Intensive Care Unit Research Fund Declarations of interest (among primary researchers): none reported Dates the study was conducted: November 1999 to December 2000	
<i>Risk of bias</i>		
Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Low risk	Participants were randomly assigned only if red blood cells of both ages were available Random length block randomisation was carried out by the Health Services Research Unit, University of Aberdeen
Allocation concealment (selection bias)	High risk	Information was insufficient to assess whether an important risk of bias exists Manuscript reports "Nonresealable envelopes were opened for individual patients by blood bank staff only after they had established that compatible blood of each age range was available" but provides no fur-

Walsh 2004 (Continued)

		ther details, such as if the envelopes were sequentially numbered and opaque
Blinding of participants (performance bias)	Low risk	All participants were treated in an intensive care unit whilst involved in the trial
Blinding of personnel (performance bias)	Low risk	“To ensure that all individuals in the intensive care unit were blinded to the age of the transfused units, special blood pack labels and forms were printed for the study. These obscured the collection and expiry dates but stated a time within which the blood must be transfused, which allowed full checking before administration” (page 365). Additionally, trial outcomes were objective as physiological markers of oxygenation and mortality
Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	Trial does not state who undertook the outcome assessment, so information is insufficient to permit assessment of risk of detection bias
Incomplete outcome data (attrition bias) All outcomes	Low risk	All participants who received treatment were included in the data analysis It is not clear how many were randomly assigned to each arm, but the number not included at each stage through enrolment to receipt of study treatment was reported
Selective reporting (reporting bias)	Low risk	All outcomes outlined in the Methods section of the trial have been reported in the Results section
Other bias	Low risk	We have no concerns about other potential sources of bias

Yuruk 2013

Methods	Parallel-group, randomised controlled trial, single-centre study Randomly assigned at: point of transfusion Sample size calculation: no Duration of follow-up: Measurements were performed before and 30 minutes after transfusion of the last red blood cell unit
Participants	Inclusion criteria: anaemic hematological outpatients requiring monthly red blood cell transfusions Exclusion criteria: age < 18 years; chemotherapy < 4 months previously; use of vasopressor-inotropic, sedative or analgesic and anticoagulant pharmacological drugs; indication

	to receive irradiated and/or washed blood products Transfusion indication: haemoglobin level < 9.6 g/dL. Transfusion was stopped after this trigger succeeded or after infusion of 4 red blood cell units	
Interventions	Comparison: fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells Intervention: red blood cell units < 1 week old Comparator: red blood cell units stored for 3 to 4 weeks Number of participants enrolled: 20 Number of participants randomly assigned to the interventions: 10/10 Numbers completing assessment: 10/10 Compliance with assigned treatments: Nothing is stated in the text about compliance with assigned interventions	
Outcomes	Primary: not stated Secondary <ul style="list-style-type: none">• Whole blood viscosity (measured using Couette low-shear viscometry)• Red blood cell deformability and aggregability (measured using laser-assisted optical rotational cell analysis)• Microcirculatory density and perfusion (measured using sidestream dark field imaging)	
Notes	Population nationality: The Netherlands Sources of funding: Landsteiner Foundation for Blood Research (Grant 20056-0621) Declarations of interest (among primary researchers): none reported Dates the study was conducted: not stated	
<i>Risk of bias</i>		
Bias	Authors' judgement	Support for judgement
Random sequence generation (selection bias)	Unclear risk	Information was insufficient to assess whether an important risk of selection bias exists
Allocation concealment (selection bias)	Unclear risk	Information was insufficient to assess whether an important risk of selection bias exists
Blinding of participants (performance bias)	Unclear risk	Information was insufficient to assess whether an important risk of performance bias exists
Blinding or personnel (performance bias)	Unclear risk	Information was insufficient to assess whether an important risk of performance bias exists

Blinding of outcome assessment (detection bias) All outcomes	Unclear risk	Information was insufficient to assess whether an important risk of detection bias exists
Incomplete outcome data (attrition bias) All outcomes	Low risk	All participants who received a red blood cell transfusion were included in the outcome analysis
Selective reporting (reporting bias)	Low risk	All outcomes outlined in the Methods section were reported in the Results
Other bias	Low risk	We have no concerns about other potential sources of bias

HCO₃ is a blood gas measure of the bicarbonate content of serum or plasma and is a significant indicator of electrolyte dispersion and anion deficit.

ICU = intensive care unit.

PaO₂/FiO₂ = ratio of partial pressure arterial oxygen and fraction of inspired oxygen; comparison between the oxygen level in the blood and the oxygen concentration that is breathed.

Characteristics of excluded studies [ordered by study ID]

Study	Reason for exclusion
Eshleman 1994	Unit of randomisation: In this study (reported as a conference abstract), the intervention was the unit of randomisation, not the participant. Minimal intervention, participant and outcome data were reported, making interpretation of findings difficult
Lebiedz 2012	This is an observational, non-randomised study (but it is mentioned in some reviews as an RCT)
Seitelbach 2011	Unit of randomisation: Participants in this abstract report were not randomly assigned between intervention arms, rather they received both interventions. No details on study methodology were given, and no outcome data were reported
Strauss 1994	Outcomes: This conference abstract seems to be a composite of the data subsequently presented in 2 full-text papers: Strauss 1996 ; Strauss 2000 In Strauss 1996 , the 'current standard practice' red blood cell transfusion was stored in AS-1 (Adsol: a commercial additive solution containing saline, adenine, dextrose and mannitol), and in Strauss 2000 , the 'current standard practice' red blood cell transfusion was stored in AS-1 (Adsol-3, a commercial additive solution containing sodium chloride, dextrose, adenine, tri-sodium citrate, citric acid and sodium phosphate) Data in Strauss 1994 are not presented separately for the AS-1 and AS-3 'older red blood cell' transfusion arms, and so we are unable to use the details and findings presented in this conference abstract

(Continued)

Wasser 1989	Ineligible intervention: This trial compares 2 units of fresh whole blood, where 'fresh' means transfused within 12 hours of collection with stored blood (i.e. donated 2 to 5 days before transfusion and stored at 4°C). This comparison does not fit into any of the comparison arms of interest for this review
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Characteristics of studies awaiting assessment *[ordered by study ID]*

Lacroix 2011

Methods	Double-blind, multi-centre, parallel randomised controlled clinical trial
Participants	Adults in intensive care units (ICUs) who <ul style="list-style-type: none"> • have had a request for a first red blood cell unit transfusion during the first 7 days of ICU admission and • have an anticipated requirement for ongoing invasive and non-invasive mechanical ventilation exceeding 48 hours
Interventions	Enrolled participants were randomly assigned at the time of transfusion to receive <ul style="list-style-type: none"> • prestorage leucoreduced red blood cells stored for 7 days or less (fresh arm) or • standard-issue red blood cells stored on average 15 to 20 days (current standard practice)
Outcomes	Primary outcome: 90-day all-cause mortality Secondary outcomes: ICU and hospital mortality, organ failure and serious nosocomial infections
Notes	ISRCTN register number 44878718 Trial opened to recruitment in January 2008 and closed to recruitment in April 2013 Target number of participants: 2510 This is the 'Age of Blood Evaluation (ABLE)' trial

NCT00458783

Methods	Parallel-arm, randomised, controlled trial
Participants	Inclusion criteria: all primary and reoperative adult (aged 18 years and older) cardiac surgical patients undergoing cardiopulmonary bypass for coronary artery bypass grafting, coronary artery bypass grafting with a valve procedure, isolated valve procedures, ascending aortic aneurysm or dissection repair alone or combined with CABG and valve procedures Exclusion criteria: descending thoracic aortic aneurysm repair; left or right ventricular assist devices; those unable to receive blood for religious reasons
Interventions	Participants undergoing cardiac surgery will be randomly assigned into 1 of 2 groups <ul style="list-style-type: none"> • Red blood cell transfusion with blood cell storage duration less than 14 days • Red blood cell transfusion with blood cell storage duration longer than 20 days
Outcomes	Primary outcome: to determine whether length of storage of red blood cells is related to postoperative morbid outcomes up to 30 days post surgery Secondary outcome: none reported

NCT00458783 (Continued)

Notes	ClinicalTrials.gov identifier: NCT00458783 Study start date: April 2007 Study completion date: February 2014 Principal Investigator: Collen Koch, The Cleveland Clinic, Ohio, USA Target number of participants: not stated
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Steiner 2010

Methods	Multi-centre, randomised, controlled trial
Participants	Patients undergoing scheduled complex cardiac surgery with planned use of median sternotomy who are likely to require a red blood cell transfusion (in patients aged older than 18 years, this will be assessed by a Transfusion Risk Understanding Scoring Tool (TRUST) probability score ≥ 3)
Interventions	Prestorage leucoreduced red blood cell units stored for 10 or fewer days or 21 or more days Randomisation will only occur if the blood bank has enough units of red blood cells of both storage times to meet the cross-match request; hence, participants randomly assigned to the 21-day arm will receive red blood cells of the same storage time as they would have received according to the standard inventory practice of "oldest units out first"
Outcomes	Primary outcome: change in Multiple Organ Dysfunction Score (MODS), a composite measure of multi-organ dysfunction, by day 7 Secondary outcomes: change in MODS by day 28, all-cause mortality and several composite and single measures of specific organ system function
Notes	ClinicalTrials.gov identifier: NCT00991341 Study start date: January 2010 Study completion date: March 2014 Trial has enrolled 1481 participants This is the 'RECESS' (Red Cell Storage Duration Study) trial

CABG = coronary artery bypass graft

CPB = cardiopulmonary bypass

Hb = haemoglobin

ICU = intensive care unit

Characteristics of ongoing studies [ordered by study ID]

Kaukonen 2013

Trial name or title	Age of blood and mortality in critically ill patients: the TRANSFUSE trial
Methods	Multi-centre (n = 60), randomised, controlled trial
Participants	Intensive care unit patients requiring red blood cell transfusion

Interventions	Intervention: the freshest compatible available red blood cells Comparator: standard practice (oldest compatible available red blood cells) Participants will receive randomly assigned treatment for all transfusion episodes during the hospital stay
Outcomes	Primary outcome <ul style="list-style-type: none"> 90 days' mortality Secondary outcomes <ul style="list-style-type: none"> Measures of organ dysfunction Sepsis Length of hospital and intensive care unit stays Quality of life
Starting date	November 2012
Contact information	Jamie Cooper, Monash University
Notes	Estimated primary completion date: end 2015 Estimated enrolment: 5000 Sponsors: NHMRC in Australia and HRC in New Zealand

NCT01586923

Trial name or title	RBC (red blood cell) Transfusion in Severe Anemia With Lactic Acidosis (TOTAL)
Methods	Randomised controlled trial
Participants	Inclusion criteria <ul style="list-style-type: none"> Aged 6 months to 5 years Hg < 5 g/dL and lactate > 5 mM Exclusion criteria <ul style="list-style-type: none"> Receiving transfusion other than red blood cells Children with known or suspected cardiac disease
Interventions	Intervention: red blood cell transfusion with packed red blood cells stored for 10 days or less Comparator: red blood cell transfusion with packed red blood cells stored for 25 days or longer
Outcomes	Primary outcome <ul style="list-style-type: none"> Reduction in elevated levels of blood lactate during the first 24 hours following a red blood cell transfusion Secondary outcomes <ul style="list-style-type: none"> Normalisation of vital signs (within 24 hours) Correction of acidosis (within 24 hours)
Starting date	February 2013
Contact information	Walter Dzik; sdzik@partners.org

NCT01586923 (Continued)

Notes	<p>Estimated primary completion date: April 2015</p> <p>Estimated enrolment: 250</p> <p>ClinicalTrials.gov identifier: NCT01586923</p> <p>Sponsor: Massachusetts General Hospital</p> <p>Study taking place in Mulago Hospital, Kampala, Uganda</p>
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NCT01976234

Trial name or title	Stored red blood cells: transfusion and immunomodulation
Methods	Randomised controlled trial
Participants	<p>Inclusion criteria</p> <ul style="list-style-type: none"> • Patients transfused during or within 24 hours after surgical operation • 18 to 85 years of age <p>Exclusion criteria</p> <ul style="list-style-type: none"> • Patients transfused during the 30 days before the operation • Immunodepressed patients • Infection during the 30 days before the operation • Emergency surgery
Interventions	<p>Intervention: fresh group: received red blood cells stored for less than 14 days</p> <p>Comparator: old group: received red blood cells stored for 14 days or longer</p>
Outcomes	Primary outcome: incidence of infection at 28 days
Starting date	September 2013
Contact information	Savino Spadaro; savinospadaro@gmail.com
Notes	<p>Estimated primary completion date: September 2014</p> <p>Estimated enrolment: 2000</p> <p>ClinicalTrials.gov identifier: NCT01976234</p> <p>Sponsor: Università degli Studi di Ferrara, Italy</p>

NCT01977547

Trial name or title	Age of Blood in Children in Pediatric Intensive Care Units (ABC PICU)
Methods	Randomised controlled trial
Participants	<p>Patients are considered eligible to participate in the trial if 1 of the following occurs</p> <ul style="list-style-type: none"> • First red blood cell transfusion is requested within the first 7 days (168 hours) of intensive care unit admission OR • First red blood cell transfusion is requested for a patient in the emergency room, the paediatric intensive care unit team is involved with clinical care of the patient, and the patient will definitively be transferred to the intensive care unit OR

	<ul style="list-style-type: none"> • Patient is assessed preoperatively and intensive care unit admission is planned postoperatively, and patient is determined to definitively require a first red blood cell transfusion during surgery <p>Inclusion criteria</p> <ul style="list-style-type: none"> • Eligible critically ill paediatric patients who have an expected length of stay after transfusion in the intensive care unit > 24 hours based on the best judgement of the attending intensive care unit staff <p>Exclusion criteria</p> <ul style="list-style-type: none"> • Age at time of enrolment < 3 days from birth or has reached 16th birthday • Postconception age < 36 weeks at time of enrolment • Documented red blood cell transfusion within 28 days before fulfilling eligibility criteria • Previously randomly assigned in this study • Weight < 3.0 kg on intensive care unit admission • Pregnant • Conscious objection or unwillingness to receive blood products • Not expected to survive beyond 24 hours, brain death or suspected brain death • Limitation or withdrawal of care decisions • Enrolment in another randomised clinical trial that has not been approved for co-enrolment • Patients for whom autologous and/or directed donation red blood cells will be provided • Patients for whom the treating physician routinely and systematically requests red blood cells ≤ 14 days of storage • Patients for whom red blood cell aliquoting policies systematically exist that mandate initial use of units stored ≤ 14 days (e.g. Pedi-Pack) • On extracorporeal membrane oxygenation (ECMO) or plan to be immediately placed on ECMO at time of enrolment • Patient predicted or presumed to require a massive transfusion (> 40 mL/kg of all blood components in a 24-hour period) according to treating physician judgement • Refusal by physician • Inability to obtain consent • Blood bank personnel experience difficulties in securing blood products (difficult cross-matches, rare blood groups and diseases like IgA deficiency) • Insufficient number of ABO-type-compatible red blood cell units available in the blood bank at randomisation with a storage time ≤ 7 days (minimum 1 unit regardless of patient age) • All red blood cell units available for the patient are not leucocyte-reduced before storage
Interventions	<p>Intervention: short storage: red blood cell storage duration equal to or less than 7 days</p> <p>Comparator: standard issue: red blood cell storage duration of 2 to 42 days with expected average length of storage of about 17 to 21 days</p>
Outcomes	<p>Primary outcome</p> <ul style="list-style-type: none"> • New or Progressive Multiple Organ Dysfunction Syndrome (NPMODS), 28 days after randomisation <p>NPMODS is defined as the proportion of patients who die during the 28 days after randomisation, or who develop NPMODS. For patients with no organ dysfunction at randomisation, New MODS is the development of ≥ 2 concurrent organ dysfunctions during the 28 days after randomisation. For patients with 1 organ dysfunction at randomisation, New MODS is the development of at least 1 other concurrent organ dysfunction after randomisation. Patients with MODS (i.e. concurrent dysfunction of ≥ 2 organ systems) at randomisation can develop progressive MODS, defined as development of at least 1 additional concurrent organ dysfunction at or during the 28 days after randomisation. All deaths will be considered progressive MODS. NPMODS will be monitored up to 28 days or until ICU discharge because it is almost never observed beyond this time in children</p> <p>Secondary outcomes</p>

	<ul style="list-style-type: none"> • Organ dysfunction, up to 28 days after randomisation • PELOD-2 (Pediatric Logistic Organ Dysfunction) score, up to 28 days after randomisation • Difference in nosocomial infection rate, up to 28 days after randomisation • Difference in rate of sepsis, severe sepsis or septic shock, up to 28 days after randomisation • Difference in rate of acute respiratory distress syndrome, up to 28 days after randomisation • Difference in duration of mechanical ventilation and ventilation-free days, up to 28 days after randomisation • Difference in intensive care unit-free days, up to 28 days after randomisation • Difference in 90-day mortality, up to 28 days after randomisation
Starting date	January 2014
Contact information	Rachel Jacobs; jacobs_r@kids.wustl.edu Lucy Clayton; lucy.clayton@recherche-ste-justine.qc.ca
Notes	Estimated primary completion date: March 2018 Estimated enrolment: 1538 ClinicalTrials.gov identifier: NCT01977547 Sponsor: National Heart, Lung, and Blood Institute (NHLBI)

NCT02050230

Trial name or title	Hemodynamic Effects of Stored Blood Transfusion in Intensive Care Patients
Methods	Randomised controlled trial
Participants	<p>Inclusion criteria</p> <ul style="list-style-type: none"> • Patient has a pulmonary artery catheter in place • Transfusion of 1 unit of packed red blood cells indicated for standard intensive care therapy • Patient has an arterial catheter • 18 years of age and older <p>Exclusion criteria</p> <ul style="list-style-type: none"> • Age < 18 years • Clinical prediction that patient will not survive at least 48 hours • Acute bleeding: > 2 units of packed red blood cells/h • Vasopressor use: noradrenalin > 0.2 µg/kg/min or any use of adrenalin within 6 hours of inclusion into the study • Therapy with inhaled nitric oxide, inhaled prostacyclin or phosphodiesterase-5-inhibitors • Sepsis
Interventions	<p>Intervention: fresh blood transfusion: 1 unit of blood stored for less than 14 days</p> <p>Comparator: standard issue blood transfusion: 1 unit of blood stored under standard conditions</p>
Outcomes	<p>Primary outcomes</p> <ul style="list-style-type: none"> • Pulmonary arterial pressure: change from baseline to immediately after transfusion (time frame of 20 minutes) • Pulmonary vascular resistance: change from baseline to immediately after transfusion (time frame of 20 minutes)

NCT02050230 (Continued)

	Secondary outcomes <ul style="list-style-type: none"> • Systemic pressure: change from baseline to immediately after transfusion (time frame of 20 minutes) • Systemic vascular resistance: change from baseline to immediately after transfusion (time frame of 20 minutes)
Starting date	January 2014
Contact information	David M Baron, MD; david.baron@meduniwien.ac.at
Notes	Estimated primary completion date: December 2015 Estimated enrolment: 64 ClinicalTrials.gov identifier: NCT02050230 Sponsor: Medical University of Vienna

NCT02087514

Trial name or title	Duration of Red Blood Cell Storage
Methods	Randomised controlled trial
Participants	Inclusion criteria <ul style="list-style-type: none"> • 18 to 65 years old • Body weight > 110 lb • Haemoglobin > 12.5 g/dL Exclusion criteria <ul style="list-style-type: none"> • Ineligible for donation based on the New York Blood Center Blood Donor Questionnaire • Systolic blood pressure > 180 or < 90 mm Hg, diastolic blood pressure > 100 or < 50 mm Hg • Heart rate < 50 or > 100 • Temperature > 99.5°F before donation • Temperature > 100.4°F or subjective feeling of illness before transfusion (this is to avoid having a concurrent illness affect post-transfusion measurements) • Positive results on standard blood donor infectious disease testing • Positive urine pregnancy test
Interventions	Intervention: transfusion of 1 week stored red cells Transfusion of blood randomly assigned to 1 week of storage Comparator: transfusion of 2 week stored red cells Comparator: transfusion of 3 week stored red cells Comparator: transfusion of 4 week stored red cells Comparator: transfusion of 5 week stored red cells Comparator: transfusion of 6 week stored red cells
Outcomes	Primary outcome <ul style="list-style-type: none"> • Non-transferrin-bound iron level: up to 24 hours post transfusion
Starting date	January 2014
Contact information	Steven L Spitalnik, MD, Columbia University, USA

NCT02087514 (Continued)

Notes	This study is ongoing but is not recruiting participants Estimated primary completion date: April 2017 Estimated enrolment: 60 ClinicalTrials.gov identifier: NCT02087514 Sponsor: National Heart, Lung, and Blood Institute (NHLBI)
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DATA AND ANALYSES

Comparison 1. Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells

Outcome or subgroup title	No. of studies	No. of participants	Statistical method	Effect size
1 Mortality	5		Risk Ratio (M-H, Fixed, 95% CI)	Totals not selected
1.1 Within 7 days in hospital (paediatric participant population)	1		Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
1.2 At up to 30 days (adult participant population)	3		Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
1.3 'Long-term' mortality (neonate participant population)	1		Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
2 Clinically accepted measures of multiple organ dysfunction	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
3 Incidence of in-hospital infection in a neonate population	1		Risk Ratio (M-H, Fixed, 95% CI)	Totals not selected
3.1 Clinical sepsis	1		Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
3.2 Necrotising enterocolitis	1		Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
4 Number of participants requiring organ support	1		Risk Ratio (M-H, Fixed, 95% CI)	Totals not selected
4.1 Number of adult participants on any vasopressor > 48 hours after surgery	1		Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
5 Economic or blood stock inventory outcomes	2		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
5.1 Mean donor exposure (neonate participant population)	2		Mean Difference (IV, Fixed, 95% CI)	0.0 [0.0, 0.0]

Comparison 2. Fresher red blood cell transfusion vs current standard practice

Outcome or subgroup title	No. of studies	No. of participants	Statistical method	Effect size
1 Mortality	7		Risk Ratio (M-H, Fixed, 95% CI)	Totals not selected
1.1 Within 7 days in an adult participant population	3		Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
1.2 At up to 30 days in a neonate participant population	1		Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
1.3 At up to 30 days in an adult participant population	3		Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]

1.4 'Long-term' mortality (up to 90 days) in a neonate participant population	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
1.5 'Long-term' mortality (at 90 days) in an adult participant population	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
2 Measures of multiple organ dysfunction (neonate participant population)	1	Risk Ratio (M-H, Fixed, 95% CI)	Totals not selected
2.1 Intraventricular haemorrhage (Papite criteria >3)	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
2.2 Retinopathy of prematurity (stage ≥ 3)	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
2.3 Bronchopulmonary dysplasia (oxygen dependency for at least 28 days at 36 weeks' gestation)	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
3 Incidence of in-hospital infection (neonate participant population)	1	Risk Ratio (M-H, Fixed, 95% CI)	Totals not selected
3.1 Necrotising enterocolitis (Bell criteria ≥ 2)	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
3.2 Clinically suspected infection	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
3.3 Confirmed infection	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
4 Number of participants requiring organ support	4	Risk Ratio (M-H, Fixed, 95% CI)	Totals not selected
4.1 Number of neonate participants requiring mechanical ventilation	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
4.2 Number of neonate participants requiring high-frequency ventilation	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
4.3 Number of adult participants requiring mechanical ventilation	2	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
4.4 Number of adult participants requiring mechanical ventilation for > 48 hours	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
4.5 Number of adult participants requiring prolonged invasive ventilation for > 48 hours	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
4.6 Number of adult participants requiring vasoreactive drugs or an aortic balloon pump or a ventricular assist device for > 48 hours	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]

4.7 Number of adult participants on any vasopressor > 48 hours after surgery	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
4.8 Number of adult participants requiring renal support	1	Risk Ratio (M-H, Fixed, 95% CI)	0.0 [0.0, 0.0]
5 Economic or blood stock inventory outcomes	3	Mean Difference (IV, Fixed, 95% CI)	Totals not selected
5.1 Mean donor exposure (neonate participant population)	3	Mean Difference (IV, Fixed, 95% CI)	0.0 [0.0, 0.0]

Analysis 1.1. Comparison 1 Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells, Outcome 1 Mortality.

Review: Transfusion of fresher versus older red blood cells for all conditions

Comparison: 1 Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells

Outcome: 1 Mortality

Analysis 1.2. Comparison 1 Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells, Outcome 2 Clinically accepted measures of multiple organ dysfunction.

Review: Transfusion of fresher versus older red blood cells for all conditions

Comparison: 1 Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells

Outcome: 2 Clinically accepted measures of multiple organ dysfunction

Analysis 1.3. Comparison 1 Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells, Outcome 3 Incidence of in-hospital infection in a neonate population.

Review: Transfusion of fresher versus older red blood cells for all conditions

Comparison: 1 Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells

Outcome: 3 Incidence of in-hospital infection in a neonate population

Analysis 1.4. Comparison 1 Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells, Outcome 4 Number of participants requiring organ support.

Review: Transfusion of fresher versus older red blood cells for all conditions

Comparison: 1 Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells

Outcome: 4 Number of participants requiring organ support

Analysis 1.5. Comparison 1 Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells, Outcome 5 Economic or blood stock inventory outcomes.

Review: Transfusion of fresher versus older red blood cells for all conditions

Comparison: 1 Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells

Outcome: 5 Economic or blood stock inventory outcomes

Analysis 2.1. Comparison 2 Fresher red blood cell transfusion vs current standard practice, Outcome 1 Mortality.

Review: Transfusion of fresher versus older red blood cells for all conditions

Comparison: 2 Fresher red blood cell transfusion vs current standard practice

Outcome: 1 Mortality

Analysis 2.2. Comparison 2 Fresher red blood cell transfusion vs current standard practice, Outcome 2 Measures of multiple organ dysfunction (neonate participant population).

Review: Transfusion of fresher versus older red blood cells for all conditions

Comparison: 2 Fresher red blood cell transfusion vs current standard practice

Outcome: 2 Measures of multiple organ dysfunction (neonate participant population)

Analysis 2.3. Comparison 2 Fresher red blood cell transfusion vs current standard practice, Outcome 3 Incidence of in-hospital infection (neonate participant population).

Review: Transfusion of fresher versus older red blood cells for all conditions

Comparison: 2 Fresher red blood cell transfusion vs current standard practice

Outcome: 3 Incidence of in-hospital infection (neonate participant population)

Analysis 2.4. Comparison 2 Fresher red blood cell transfusion vs current standard practice, Outcome 4 Number of participants requiring organ support.

Review: Transfusion of fresher versus older red blood cells for all conditions

Comparison: 2 Fresher red blood cell transfusion vs current standard practice

Outcome: 4 Number of participants requiring organ support

Analysis 2.5. Comparison 2 Fresher red blood cell transfusion vs current standard practice, Outcome 5 Economic or blood stock inventory outcomes.

Review: Transfusion of fresher versus older red blood cells for all conditions

Comparison: 2 Fresher red blood cell transfusion vs current standard practice

Outcome: 5 Economic or blood stock inventory outcomes

ADDITIONAL TABLES

Table 1. Characteristics of the interventions

		Quantity of red blood cells transfused		Age of red blood cells as defined in the Methods		Actual age of red blood cells received			
Trial	Types of RBCs (RBC additive solutions) ^a	'Fresh'	'Older/ Standard issue'	'Fresh'	'Older/ Standard issue'	'Fresh'	'Older/ Standard issue'	Fur-ther infor-mation: 'Fresh'	Fur-ther infor-mation 'Older/ Standard issue'
Fresher red blood cell transfusion versus an intervention arm actively transfused older red blood cells									
Neonate participant population									
Fernandes 2005	Irridiated and leu-coreduced (CPDA-1)	Number of trans-fusions per infant transfused: mean (SD) 4.2 (3.1); range 1 to	Number of trans-fusions per infant transfused: mean (SD) 4.4 (4.0);	RBCs < 3 days old	RBCs up to 28 days old	Mean (SD) : 1.6 (0.6) days	Mean (SD) : 9.0 (8.9) days	(nothing)	(nothing)

Table 1. Characteristics of the interventions (Continued)

		13 transfusions Mean (SD) red cell volume trans- fused: 62.9 (45.1) mL kg ⁻¹	range 1 to 20 transfu- sions Mean (SD) red cell volume trans- fused: 65.3 (58.3) mL kg ⁻¹						
Liu 1994	Irradiated (CPDA-1)	Infants re- ceived 55 RBC trans- fusions during the study	Infants re- ceived 73 RBC trans- fusions during the study	Quint packs < 5 days old When the need for multi- ple trans- fusions was antic- ipated, at- tempts were made to limit donor ex- posures by reserving mul- tiple quint packs from the same donor for a patient	Assigned a single adult unit of PRBC, which was reserved for that in- fant up to the recom- mended stor- age time of 35 days	Mean (SD) : 3.5 (1.2) days	Mean (SD) : 12.9 (10. 9) days	(nothing)	(nothing)
Paediatric participant population									
Dhabangi 2013	Not stated (SAGM)	Volume of RBCs transfused, mean (SD) : 12.7 (2.6) mL/kg	Volume of RBCs transfused, mean (SD) : 12.7 (2.2) mL/kg	Aged 1 to 10 days	Aged 21 to 35 days	Mean (SD) : 7.8 (1.8) days	Mean (SD) : 27.2 (3.9) days	(nothing)	(nothing)
Adult participant population									
Bennett- Guerrero 2009 [1],	Leucore- duced (AS-1 or AS-3)	Number of units transfused per partic-	Number of units transfused per partic-	Mean (SD) : 7(4) days	Mean (SD) : 21 (4) days Transfu-	Mean (SD) : 6 (2) days	Mean (SD) : 18 (7) days	Age of youngest red cell	Age of youngest red cell

Table 1. Characteristics of the interventions (Continued)

		ipant, median (IQR) : 4 (2.5-6.5)	ipant, median (IQR) : 5 (4-10)		sions in this arm were within the target window for only 5 of 10 transfused participants			unit, mean (SD): 5 (1) days. Maximum age, mean (SD) : 7 (2) days	unit, mean (SD): 17 (7) days. Maximum age, mean (SD) : 20 (7) days
Neuman 2013	Not stated	Not stated	Not stated	< 10 days old	> 21 days old	Mean 9.1 (3.1) days	Mean 29.8 (5.7) days	(nothing)	(nothing)
Schulman 2002	Not stated	Number of transfusions per infant transfused: mean (SD) 9.3 (1.9)	Number of transfusions per infant transfused: mean (SD) 10.6 (3.35)	< 7 days old	> 20 days old	Not stated	Not stated	(nothing)	(nothing)
Walsh 2004	Leucoreduced (SAGM)	Mean (SD) red cell volume transfused: 307 (25) mL	Mean (SD) red cell volume transfused: 344 (26) mL	Red blood cell units ≤ 5 days old	Red blood cell units ≥ 20 days old	Median (IQR) : 2 (2-2.25) days	Median (IQR): 28 (26.75-31) days	Range = 2 to 3	Range = 22 to 32
Yuruk 2013	Leucoreduced (SAGM)	Number of red cell bags: median (IQR) : 3 (2-3)	Number of red cell bags: median (IQR) : 3 (3-3)	Storage of less than 1 week	Storage of 3 to 4 weeks	Median (IQR) : 7 (5-7) days	Median (IQR) : 23 (22-28) days	(nothing)	(nothing)
Fresher red blood cell transfusion vs current standard practice									
Neonate participant population									
Fergusson 2012	Irrigated and leucoreduced (not stated)	Mean (SD) number of transfusion episodes: 5.01 (4.00). Range 1 to 10 transfu-	Mean (SD) number of transfusion episodes: 4.94 (3.88) Range 1 to 10 transfu-	Aged ≤ 7 days	Range 2 to 42 days In this group, units were divided into aliquots to	Mean (SD) : 5.10 (2.05) days	Mean (SD) : 14.58 (8.26) days	Median (IQR) = 5.00 (4.00-6.00) days	Median (IQR) = 13.00 (8.00-19.00) days

Table 1. Characteristics of the interventions (Continued)

		sion episodes	sion episodes		increase usage and reduce waste. Each aliquot was des- ignated for use in a sin- gle infant up to its ex- piry date				
Strauss 1996	Irridiated (CPDA-1 or AS-1)	Mean (SD) number of trans- fusions per infant: 3.9 (2.6)	Mean (SD) number of trans- fusions per infant: 4.7 (2.8)	Aged < 7 days	Aged < 42 days From 1 dedicated donor who consented to donate a second unit when needed by the infant	Aged < 7 days = 100% of RBC transfu- sions (58 units transfused)	Aged < 7 days = 17% of RBC transfu- sions (n = 11 units) Aged 7 to 12 days = 36% of RBC transfu- sions (n = 24 units)	(nothing)	The follo- ing consti- tuted 47% of transfu- sions given 15 to 21 days = 16 units 22 to 28 days = 7 units 29 to 35 days = 3 units 36 to 42 days = 5 units
Strauss 2000	Irridi- ated, leu- coreduced (CPDA-1 or AS-3)	Mean (SD) number of trans- fusions per infant: 6.7 (3.9) To- tal number of transfu- sions given = 40	Mean (SD) number of trans- fusions per infant: 3.5 (2.1) To- tal number of transfu- sions given = 28	Aged < 7 days	Aged < 42 days (RBC units from dedicated donors only)	All units < 7 days	28 units transfused in this arm: aged < 7 days old = 5 units; aged 7 to 14 days = 9 units; aged 15 to 42 days = 14 units	(nothing)	(nothing)
Adult participant population									
Aubron 2012	Leucore- duced (SAGM)	Number of units	Number of units	"Freshest"	"Compati- ble, non	Mean (SD) : 12.1 (3.8)	Mean (SD) : 23 (8.4)	Minimum age,	Minimum age,

Table 1. Characteristics of the interventions (Continued)

		transfused per patient, mean (SD) : 3.2 (2.6)	transfused per patient, mean (SD) : 3.8 (3.6)		expired units with the longest storage du- ration at the time of transfu- sion request”	days Mean age range of red blood cells = 3 to 19 days	days Mean age range of red blood cells = 7 to 41.5 days	mean (SD) : 9.5 (4.5) days Maximum age, mean (SD) : 15 (6.5) days	mean (SD) : 20.5 (8.9) days Maximum age, mean (SD) : 26 (9.2) days
Bennett- Guerrero 2009 [2]	Leucore- duced (AS-1 or AS-3)	Number of units transfused per pa- tient, me- dian (IQR) : 5 (2-7)	Number of units transfused per pa- tient, me- dian (IQR) : 4 (2-5)	Less than 21 days	Unit standard of care = old- est RBC unit out first	Mean (SD) : 11 (4) days	Mean (SD) : 16 (5) days	Age of youngest red cell unit, mean (SD): 13 10 (5) days Maximum age, mean (SD) : 12 (4) days	Age of youngest red cell unit, mean (SD): 13 (4) days Maximum age, mean (SD) : 18 (7) days No red cell unit was > 31 days old
Heddle 2012	Leucore- duced (SAGM)	Total num- ber of RBC units transfused: 1157 Me- dian (IQR) : 2 (2-4) Range 1 to 102	Total num- ber of RBC units transfused: 2369 Me- dian (IQR) : 2 (2-5) Range 1 to 38	“Freshest available”	“Oldest in the inven- tory”	Max- imum age, mean (SD) : 12.0 (6.8) days	Max- imum age, mean (SD) : 26.6 (7.8) days	Range 2 to 27 days and 33 to 37 days	Range 3 to 42 days
Hebert 2005	Leucore- duced (CPD-2 & AS-3)	Me- dian (IQR) number of RBC units transfused: 3 (2-5)	Me- dian (IQR) number of RBC units transfused: 2 (2-4)	Aged < 8 days old	This group received RBCs issued from the hospital blood bank with the longest storage time in accordance	Median = 4 days	Median = 19 days	Overall 73% of partic- ipants received RBCs with storage times that corre- spond to treatment allocation	Overall 73% of partic- ipants received RBCs with storage times that corre- spond to treatment allocation

Table 1. Characteristics of the interventions (Continued)

					with standard blood bank procedure. To ensure the maximum age separation between groups, participants were allocated only on days when average age of RBCs in the blood bank exceeded 15 days			more than 90% of the time. Compliance target of 90% was attained by 91% of patients allocated to fresh arm compared with 59% in standard arm	more than 90% of the time. Compliance target of 90% was attained by 91% of participants allocated to fresh arm compared with 59% in standard arm
Kor 2012	Leucoreduced (not stated)	1 unit	1 unit	Single unit of fresh red blood cells < 5 days old Subsequent transfusions (after the first study transfusion) were standard issue	Single unit of standard issue red blood cells (median = 21 days)	Median (IQR) : 4.0 (3.0-5.0) days	Median (IQR): 26.5 (21.0-36.0) days	(nothing)	4 participants received an RBC unit that had been stored for ≤ 14 days; 9 participants received an RBC unit that had been stored between 15 and 21 days, and 37 participants received an RBC unit that had been stored for > 21 days

^aType of RBCs relates to details of whether trials report that red blood cells were leucoreduced or irradiated, or whether whole blood rather than red blood cells were transfused. The nature of the blood that was transfused is recorded here.

AS-1 = Adsol, a commercial additive solution containing sodium chloride, dextrose, adenine, mannitol; AS-3 = Adsol-3, a commercial additive solution containing sodium chloride, dextrose, adenine, tri-sodium citrate, citric acid, sodium phosphate; CPD-1 = citrate phosphate dextrose 1; CPDA-1 = citrate, phosphate, dextrose, adenine 1; PRBC = packed red blood cells; RBCs = red blood cells; SAGM = saline adenine glucose mannitol.

Table 2. Duration of organ support

Trial	'Fresh'	'Older/Standard issue'
Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells		
Respiratory support (invasive and non-invasive ventilation)		
Neonatal participant population		
Fernandes 2005	Duration of mechanical ventilation, mean (SD) 16.5 (14.5) days	Duration of mechanical ventilation, mean (SD) 22.9 (28.9) days
Adult participant population		
Bennett-Guerrero 2009 [1]	Duration of mechanical ventilation, mean (SD) = 31 (33) hours Percentage [number]* of participants on any vasopressor > 48 hours after surgery = 17% [2]	Duration of mechanical ventilation, mean (SD) = 16 (6) hours Percentage [number]* of participants on any vasopressor > 48 hours after surgery = 25% [3]
Fresher red blood cell transfusion vs current standard practice		
Respiratory support (invasive and non-invasive ventilation)		
Neonatal participant population		
Fergusson 2012	Percentage [number] of participants requiring: mechanical ventilation = 80.3% [151] high-frequency ventilation = 39.9% [75]	Percentage [number] of participants requiring: mechanical ventilation = 80.9% [153] high-frequency ventilation = 39.2% [74]
Adult participant population		
Aubron 2012	Percentage [number] of participants requiring: mechanical ventilation = 92% [23] Duration of mechanical ventilation, median (IQR) 156 (6.1-253) days	Percentage [number] of participants requiring: mechanical ventilation = 73% [19] Duration of mechanical ventilation, median (IQR) 9.85 (0-198) days

Table 2. Duration of organ support (Continued)

Bennett-Guerrero 2009 [2]	Duration of mechanical ventilation, mean (SD) = 22 (20) hours Percentage [number]* of participants on any vasopressor > 48 hours after surgery = 33% [3]	Duration of mechanical ventilation, mean (SD) = 18 (10) hours Percentage [number]* of participants on any vasopressor > 48 hours after surgery = 9% [1]
Hebert 2005	Percentage [number] of participants requiring: mechanical ventilation = 85% [22] Prolonged invasive mechanical ventilation for > 48 hours = 27% [7] Prolonged mechanical ventilation for > 48 hours = 35% [9] Requirement for vasoactive drugs or an aortic balloon pump or ventricular assist device for > 48 hours = 19% [5]	Percentage [number] of participants requiring: mechanical ventilation = 61% [19] Prolonged invasive mechanical ventilation for > 48 hours = 16% [5] Prolonged mechanical ventilation for > 48 hours = 26% [8] Requirement for vasoactive drugs or an aortic balloon pump or ventricular assist device for > 48 hours = 13% [4]
Duration of renal support (haemofiltration)		
Hebert 2005 Number of participants on dialysis over the 30 days of the study	0% [n = 0]	6% [n = 2]

* As the number of participants included in the analysis for this outcome was known, the number reported here was calculated for the purposes of this review.

Table 3. Length of stay: hospital and intensive care unit

Trial	'Fresh'	'Older/Standard issue'
Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells		
Neonate participant population		
Fernandes 2005 <i>Length of hospital stay: mean (standard deviation)</i>	60.8 (37.3) days	62.6 (48.3) days
Adult participant population		
Bennett-Guerrero 2009 [1] <i>Length of hospital stay: mean (standard deviation)</i>	10 (9) days	8.6 (4) days
Bennett-Guerrero 2009 [1] <i>Length of intensive care unit stay: mean (standard deviation)</i>	51 (67) hours	51 (56) hours

Table 3. Length of stay: hospital and intensive care unit (Continued)

Fresher red blood cell transfusion vs current standard practice		
Neonate participant population		
Fergusson 2012 <i>Length of intensive care unit stay: median (interquartile range)</i>	84 (50, 104) days*	77 (50, 104) days*
Strauss 1996 <i>Percentage discharge from hospital before 84 days</i>	14%	10%
Adult participant population		
Aubron 2012 <i>Length of hospital stay: median (interquartile range)</i>	21 (12, 38) days*	17 (8, 27) days*
Bennett-Guerrero 2009 [2] <i>Length of hospital stay: mean (standard deviation)</i>	10 (7) days	13 (14) days
Aubron 2012 <i>Length of intensive care unit stay: median (interquartile range)</i>	11 (5, 15) days*	7 (3, 17) days*
Bennett-Guerrero 2009 [2] <i>Length of intensive care unit stay: mean (standard deviation)</i>	69 (136) hours	47 (51) hours

* This outcome is reported as median values (with 25 [and] 75 percentiles).

Table 4. Physiological markers of oxygen consumption or alterations in microcirculation

Trial	'Fresher'	'Older/Standard issue'
Fresher red blood cell transfusion vs an intervention arm actively transfused with older red blood cells		
Neonate participant population		
Liu 1994	Mean (SD) change (before and after transfusion) in: potassium concentration (mmol/L) = 0.10 (1.22) pH level = -0.02 (0.08) base excess = -0.82 (1.99)	Mean (SD) change (before and after transfusion) in: potassium concentration (mmol/L) = 0.10 (1.22) pH level = -0.02 (0.08) base excess = -0.82 (1.99)

Table 4. Physiological markers of oxygen consumption or alterations in microcirculation (Continued)

Paediatric participant population		
Dhabangi 2013	Resolution of lactic acidosis within 4 hours of red blood cell transfusion = 92% [n = 34]	Resolution of lactic acidosis within 4 hours of red blood cell transfusion = 81% [n = 30]
Adult participant population		
Bennett-Guerrero 2009 [1]	PaO ₂ /FiO ₂ ratio, mean (SD), at baseline = 420 (158) 2 hours post operation = 256 (73) Lactic acid, concentration 2 hours post operation, mean (SD) = 1.9 (1.6)	PaO ₂ /FiO ₂ ratio, mean (SD), at baseline = 386 (128) 2 hours post-operation = 360 (64) Lactic acid, concentration 2 hours post operation, mean (SD) = 2.5 (1.5)
Neuman 2013	“patients had a [mean (SD)] 0.191 (0.438) and 0.069 (0.242) μM increase in N ₂ at 1 and 24 hours after transfusion respectively” “Patients receiving fresh pRBC [packed red blood cells] had no significant change in FMD” [flow-mediated dilation, a measure of vascular endothelial function] No significant change in FHB [plasma free haemoglobin] was noted from before to after a red blood cell transfusion	“patients had a [mean (SD)] 0.055 (0.227) and 0.079 (0.226) μM decrease in N ₂ at 1 and 24 hours after transfusion respectively” “FMD [flow-mediated dilation, a measure of vascular endothelial function] significantly decreased by 2.4% 24 hour after transfusion of aged pRBC” [packed red blood cells] No significant change in FHB [plasma free haemoglobin] was noted from before to after a red blood cell transfusion
Walsh 2004	Median (plus interquartile range) from baseline to post-transfusion period* Intragastric arterial difference in PCO ₂ (Pg-PaCO ₂ gap) = -0.15 (-0.31 to -0.01) PgCO ₂ , kPa = -0.46 (-1.36 to -0.18) PgCO ₂ -PaCO ₂ , kPa = -0.41 (-0.82 to 0.10) pHi = 0.02 (-0.01 to 0.05) Lactate = 0.10 (-0.16 to 0.21) mmol/L arterial pH = 0.00 (-0.001 to 0.001) HCO ₃ ⁻ (actual) = 0.85 (-1.44 to -0.53) mmol/L Arterial base excess = -0.70 (-1.42 to -0.45)	Median (plus interquartile range) from baseline to post-transfusion period* Intragastric arterial difference in PCO ₂ (Pg-PaCO ₂ gap) = 0.02 (-0.29 to 0.11) PgCO ₂ , kPa = 0.16 (0.36-1.19) PgCO ₂ -PaCO ₂ , kPa = 0.37 (-0.17 to 1.66) pHi = -0.02 (-0.06 to 0.01) Lactate = 0.03 (-0.10 to 0.23) mmol/L arterial pH = 0.00 (-0.001 to 0.001) HCO ₃ ⁻ (actual) = -0.29 (-0.90 to 0.07) mmol/L Arterial base excess = -0.71 (-1.5 to -0.42)
Yuruk 2013	Mean (SD) changes in microcirculatory density (measured before and 30 minutes after transfusion) from 17 (3) mm/mm ² to 19 (3) mm/mm ² .	Mean (SD) changes in microcirculatory density (measured before and 30 minutes after transfusion) from 17 (3) mm/mm ² to 19 (2) mm/mm ² .
Fresher red blood cell transfusion vs current standard practice.		
Neonate participant population		
Fergusson 2012	Percentage [number] of patients requiring: supplemental oxygen = 96.8% [182] nasal continuous positive airway pressure = 79.3%	Percentage [number] of patients requiring: supplemental oxygen = 94.7% [179]

Table 4. Physiological markers of oxygen consumption or alterations in microcirculation (Continued)

	[149]	nasal continuous positive airway pressure = 80.4% [152]
Strauss 1996	Mean (SD) change from before to after transfusion pH = 0.00 (0.05) Sodium = 0.5 (4.4) mEq/L Potassium = 0.3 (0.9) mEq/L Calcium = -0.1 (0.7) mg/dL Lactate = -0.3 (0.4) mmol/L Glucose = -9 (32) mg/dL	Mean (SD) change from before to after transfusion pH = 0.00 (0.13) Sodium = 0.6 (5.1) mEq/L Potassium = -0.2 (1.5) mEq/L Calcium = -0.1 (0.5) mg/dL Lactate = -0.1 (0.6) mmol/L Glucose = -7 (25) mg/dL
Strauss 2000	Mean (SD) change from before to after transfusion pH = -0.01 (0.9) Sodium = -0.4 (5.5) mEq/L Potassium = 0.26 (1.16) mEq/L Calcium = -0.13 (1.04) mg/dL Lactate = -0.16 (0.30) mmol/L Glucose = -17.5 (40.3) mg/dL	Mean (SD) change from before to after transfusion pH = -0.03 (0.6) Sodium = 1.9 (4.7) mEq/L Potassium = 0.38 (0.91) mEq/L Calcium = 0.13 (0.64) mg/dL Lactate = -0.53 (1.46) mmol/L Glucose = -16.7 (24.4) mg/dL
Adult participant population		
Bennett-Guerrero 2009 [2]	PaO ₂ /FiO ₂ ratio, mean (SD), at baseline = 414 (97) 2 hours post operation = 267 (81) Lactic acid, concentration 2 hours post operation, mean (SD) = 2.2 (1.1)	PaO ₂ /FiO ₂ ratio, mean (SD), at baseline = 284 (122) 2 hours post operation = 212 (87) Lactic acid, concentration 2 hours post operation, mean (SD) = 2.4 (1.0)
Kor 2012	Mean change from before to after transfusion Pa _{o2} /Fi _{o2} = 2.5 (49.3) mm Hg	Mean change from before to after transfusion Pa _{o2} /Fi _{o2} = -9.0 (69.8) mm Hg

* Baseline period (2.5 hours: mean of 5 measurements) to post-transfusion period (5 hours: mean of 5 measurements).

PaO₂/FiO₂ = ratio of partial pressure of arterial oxygen to fraction of inspired oxygen.

APPENDICES

Appendix I. Search strategies

CENTRAL

- #1 MeSH descriptor: [Erythrocyte Transfusion] explode all trees
- #2 (red cell* or red blood cell* or erythrocyte* or RBC* or blood) near/1 (transfus* or infus* or hypertransfus* or retransfus*)
- #3 MeSH descriptor: [Blood Transfusion] this term only
- #4 MeSH descriptor: [Blood Component Transfusion] this term only
- #5 #3 or #4
- #6 MeSH descriptor: [Erythrocytes] this term only
- #7 red cell* or red blood cell* or erythrocyte* or RBC* or whole blood
- #8 #6 or #7
- #9 #5 and #8
- #10 (red cell* or red blood cell* or erythrocyte* or RBC* or whole blood):ti
- #11 #1 or #2 or #9 or #10
- #12 MeSH descriptor: [Blood Preservation] explode all trees
- #13 (age or aged or aging or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest or store* or storage or storing or preserv*):ti
- #14 #12 or #13
- #15 #11 and #14
- #16 ((red cell* or red blood cell* or erythrocyte* or RBC* or blood or transfus*) near/5 (age or aged or aging or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest or store* or storage or storing or preserv*)):ti
- #17 #15 or #16

MEDLINE (Ovid SP)

- 1. ERYTHROCYTE TRANSFUSION/
- 2. ((blood or erythrocyte* or red cell* or red blood cell* or RBC*) adj1 (transfus* or infus* or retransfus*)):ti,ab.
- 3. BLOOD TRANSFUSION/ or BLOOD COMPONENT TRANSFUSION/
- 4. ERYTHROCYTES/
- 5. (red cell* or red blood cell* or erythrocyte* or RBC* or whole blood).tw.
- 6. 4 or 5
- 7. 3 and 6
- 8. (RBC* or red cell* or red blood cell* or erythrocyte* or whole blood).ti.
- 9. 1 or 2 or 7 or 8
- 10. exp Blood Preservation/
- 11. *Time Factors/
- 12. (age or aged or aging or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest or store* or storage or storing or preserv*):ti.
- 13. 10 or 11 or 12
- 14. 9 and 13
- 15. ((red cell* or red blood cell* or erythrocyte* or RBC* or blood or transfus*) adj5 (age or aged or aging or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest or store* or storage or storing or preserv*)):ti.
- 16. ((red cell* or red blood cell* or erythrocyte* or RBC* or whole blood) adj3 (store* or storage or storing or preserv* or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest)):ab.
- 17. 14 or 15 or 16

EMBASE (Ovid SP)

- 1. ERYTHROCYTE TRANSFUSION/
- 2. ((blood or erythrocyte* or red cell* or red blood cell* or RBC*) adj1 (transfus* or infus* or retransfus*)):ti,ab.
- 3. BLOOD TRANSFUSION/ or BLOOD COMPONENT THERAPY/
- 4. ERYTHROCYTE/ or ERYTHROCYTE CONCENTRATE/
- 5. (red cell* or red blood cell* or erythrocyte* or RBC* or whole blood).tw.
- 6. 4 or 5
- 7. 3 and 6

8. (RBC* or red cell* or red blood cell* or erythrocyte* or whole blood).ti.
9. 1 or 2 or 7 or 8
10. BLOOD STORAGE/
11. ERYTHROCYTE PRESERVATION/
12. STORAGE TIME/
13. *TIME/
14. (age or aged or aging or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest or store* or storage or storing or preserv*).ti.
15. 10 or 11 or 12 or 13 or 14
16. 9 and 15
17. ((red cell* or red blood cell* or erythrocyte* or RBC* or blood or transfus*) adj5 (age or aged or aging or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest or store* or storage or storing or preserv*).ti.
18. ((red cell* or red blood cell* or erythrocyte* or RBC* or whole blood) adj3 (store* or storage or storing or preserv* or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest)).ab.
19. 16 or 17 or 18

CINAHL (EBSCOhost)

1. (MH "Erythrocyte Transfusion")
2. TI ((red cell* or red blood cell* or erythrocyte* or RBC* or blood) N1 (transfus* or infus* or retransfus*)) OR AB ((red cell* or red blood cell* or erythrocyte* or RBC* or blood) N1 (transfus* or infus* or retransfus*))
3. (MH "Blood Transfusion") OR (MH "Blood Component Transfusion")
4. (MH "Erythrocytes")
5. TI (red cell* or red blood cell* or erythrocyte* or RBC* or whole blood) OR AB (red cell* or red blood cell* or erythrocyte* or RBC* or whole blood)
6. S4 OR S5
7. S3 AND S6
8. TI (red cell* or red blood cell* or erythrocyte* or RBC* or whole blood)
9. S1 OR S2 OR S7 OR S8
10. (MH "Blood Preservation")
11. (MM "Time Factors")
12. TI (age or aged or aging or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest or store* or storage or storing or preserv*)
13. S10 OR S11 OR S12
14. S9 AND S13
15. TI ((red cell* or red blood cell* or erythrocyte* or RBC* or blood or transfus*) N5 (age or aged or aging or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest or store* or storage or storing or preserv*))
16. AB ((red cell* or red blood cell* or erythrocyte* or RBC* or whole blood) N3 (store* or storage or storing or preserv* or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest))
17. S14 OR S15 OR S16

PubMed (e-publications only)

(red cell*[TI] OR blood[TI] OR erythrocyte*[TI] OR transfus*[TI] or RBC*[TI]) AND (random* or trial* OR controlled OR control group OR blind* OR systematic*) AND (age[TI] or aged[TI] or aging[TI] or fresh*[TI] or old[TI] or older[TI] or oldest[TI] or new[TI] or newer[TI] or newest[TI] or young[TI] or younger[TI] or youngest[TI] or store*[TI] or storage[TI] or storing[TI] or preserv*[TI]) AND (publisher[sb] NOT pubstatusnihms)

Transfusion Evidence Library (www.transfusionevidencelibrary.com)

(red cell* or red blood cell* or erythrocyte* or RBC* or blood or transfus*) [in Record Title] AND (age or aged or aging or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest or store* or storage or storing or preserv*) [in Record Title]

LILACS

db:("LILACS") AND type of study:("clinical trials" OR "systematic reviews" OR "guideline" OR "evidence synthesis") AND (tw: ("red cells" OR "red blood cells" OR "age of blood" OR "blood transfusion" OR transfus*) OR ti:(blood)) AND limit:("humans")

Web of Science (CPCI-S)

Title: (red cell* or red blood cell* or erythrocyte* or RBC* or blood or transfus*) AND Title: (age or aged or aging or fresh* or old or older or oldest or new or newer or newest or young or younger or youngest or store* or storage or storing or preserv*) AND RCT Filter/Topic: (random* or blind* or trial* or control*)

ISRCTN and EU Clinical Trials Register

(red cells OR red blood cells OR erythrocytes OR RBCs OR blood OR transfusion) AND (age OR aged OR aging OR fresh OR fresher OR freshest OR old OR older OR oldest OR new OR newer OR newest OR young OR younger OR youngest OR stored OR storage OR storing OR preserved)

ClinicalTrials.gov

Search Terms: (age OR aging OR fresh OR fresher OR freshest OR old OR older OR oldest OR new OR newer OR newest OR young OR younger OR youngest OR stored OR storage) AND transfusion AND red blood cells

WHO ICTRP

Title: (age OR aged OR aging OR fresh OR fresher OR freshest OR old OR older OR oldest OR new OR newer OR newest OR young OR younger OR youngest OR stored OR storage OR storing)

Intervention: transfusion

UMN-CTR Japanese Clinical Trials Registry and the Hong Kong Clinical Trials Registry

Title: (age OR aged OR aging OR fresh OR fresher OR freshest OR old OR older OR oldest OR new OR newer OR newest OR young OR younger OR youngest OR stored OR storage OR storing)

Intervention: transfusion

CONTRIBUTIONS OF AUTHORS

All review authors contributed to development of the protocol and preparation of the final review manuscript. SB, MT and SS prepared the first drafts of the manuscript and thereafter incorporated comments from all other review authors through preparation of the final manuscript.

Susan Brunskill undertook screening of references, data extraction and risk of bias assessments, analysed the data and entered the data into Review Manager 5. She assisted Richard Gregg in the design of data extraction forms, and she led the review.

Kirstin Wilkinson undertook data extraction and risk of bias assessments. She provided cardiac anaesthesia content expertise.

Carolyn Dorée developed the search strategies, advised on resources to be searched for the review, ran the searches and performed a first sift of the references, removing duplicates and clearly irrelevant references.

Marialena Trivella provided statistical and methodological expertise for this review as well as support with decisions about the diversity of the interventions and appropriate data analysis.

Simon Stanworth undertook screening of the references, piloted the data extraction form and undertook data extraction and risk of bias assessments. He provided transfusion medicine content expertise.

DECLARATIONS OF INTEREST

Marialena Trivella was a Training Co-ordinator at the UK Cochrane Centre and is a statistical editor/referee for five Cochrane groups (Anaesthesia, Wounds, Injuries, Breast Cancer and Sexually Transmitted Infections). This work with the Cochrane groups is independent of her involvement in this review. Marialena Trivella declares that her involvement here as a review author does not involve financial relationships.

All other authors: none known.

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- NHS Blood and Transplant, Research and Development, UK, Other.

External sources

- No sources of support supplied

DIFFERENCES BETWEEN PROTOCOL AND REVIEW

In this review we have separated our reporting of data on 'Effects of Interventions' between trials of neonates and trials of adult participants (a predefined subgrouping for this review). The decision to present the data in this way was due to innate clinical differences between these two groups of participants: The physiology of the neonate is fundamentally different from that of the adult, and this particularly applies to extremely premature neonates.

INDEX TERMS

Medical Subject Headings (MeSH)

*Blood Preservation [adverse effects; mortality]; *Erythrocyte Transfusion [adverse effects; mortality]; Guidelines as Topic; Infant, Very Low Birth Weight; Randomized Controlled Trials as Topic; Time Factors

MeSH check words

Adult; Child; Humans; Infant, Newborn