Outbreaks of extended spectrum beta-lactamase-producing Enterobacteriaceae in neonatal intensive care units: a systematic review

Patrick JM Stapleton,1 Madeleine Murphy,2 Naomi McCallion,2,3 Marion Brennan,4 Robert Cunney,1,5 Richard J Drew6,7

ABSTRACT
Objective To establish the number of outbreaks of extended spectrum beta-lactamase (ESBL) producing organisms in neonatal intensive care units (NICUs), to determine causes, mortality rates, proportions of infants colonised and infected and the interventions that terminated outbreaks.

Methods A systematic review of the literature in English, Spanish and French was undertaken with searches in four databases. The review conformed to the PRISMA guidelines, and the data extraction was modelled on the ORION criteria for studies of nosocomial infection.

Results 75 studies fulfilled the inclusion criteria. There were 1185 cases of colonisation, 860 infections and 139 deaths. The median outbreak duration was 6.2 months (IQR 2.0–7.5 months). Klebsiella pneumoniae was the most frequently implicated pathogen. Understaffing was the most frequent risk factor for outbreaks. The most commonly identified source was admission of an ESBL-colonised infant with subsequent horizontal dissemination. The main interventions described were improved infection-control procedures and screening of staff and the environment. 26 studies were included in the quantitative analysis. Random effects meta-analysis indicated high mortality rates in infants who developed infection (31%, 95% CI 20% to 43%).

Conclusion ESBL outbreaks in NICUs are associated with significant mortality and prolonged disruption. Understaffing is a major risk factor, but is infrequently addressed by interventions. Poor infection-control procedures are frequently implicated as contributing to ESBL spread. Better reporting of outbreaks may help clarify the role for routine ESBL screening in NICUs.

INTRODUCTION
Neonates, especially premature neonates, are at risk of significant mortality and morbidity from sepsis, but the clinical presentation is often non-specific, necessitating frequent use of empiric antibiotic therapy in unstable infants. Attempts to limit duration of antibiotic therapy must be balanced against mortality rates of up to 15% for neonatal sepsis.1 Enterobacteriaceae are recognised as serious pathogens in the neonatal intensive care unit (NICU). Rates of Escherichia coli early onset sepsis have increased along with a reduction in group B Streptococcus infection in very low birthweight (VLBW, <1500 g) infants.2,3 The emergence of extended spectrum beta-lactamase (ESBL) producing Enterobacteriaceae presents major challenges in managing neonatal sepsis. ESBL enzymes, most commonly encoded by the TEM, SHV and CTX-M families of beta-lactamase genes, hydrolyse many front-line antibiotics such as penicillin and third-generation cephalosporins. ESBLs are encountered in a variety of Enterobacteriaceae, most commonly Klebsiella (K. pneumonia in particular) and E. coli.

ESBL colonisation rates in paediatric intensive care units (PICUs) range up to 12% for E. coli and 39% for Klebsiella spp.4 ESBL rates as high as 60% for Klebsiella spp and 75% for E. coli have been reported from blood cultures from infected infants in some NICUs.5 In a UK-wide survey, 26% of NICUs reported ESBL in their unit, but less than one-third had reported an outbreak.6 There was considerable variability both in routine screening (11% of units) and isolation practices following identification of ESBL.

The purpose of the review was to establish the number of outbreaks of ESBL-producing organisms in NICUs, to determine the causes, mortality rates, proportions of infants colonised and infected and the interventions that terminated the outbreaks. To our knowledge, no such review has been carried out before. The results can be used by clinicians to estimate the impact of an ESBL outbreak in their own NICU, to take measures to mitigate risk factors and to help choose containment measures in the event of an outbreak.

METHODS
Protocol, registration and eligibility criteria The review methods and eligibility criteria were specified in advance, and the review protocol was registered with PROSPERO, the Centre for Reviews and Dissemination, University of York (registration number CRD42014010744). The review was designed in accordance with the PRISMA guidelines.7 We reviewed published work on outbreaks of ESBL-producing organisms in NICUs. Target languages were English, French and Spanish. There was no publication period restriction. Studies had to relate to outbreaks of ESBL-producing organisms among neonates in an NICU setting. Studies from units that also cared for older infants (<1 year of age) could be included, provided that the unit involved was an NICU, and not a lower acuity unit. Studies involving non-ESBL-producing organisms, or infection or colonisation outside the context of an outbreak, were excluded. Types of studies eligible for inclusion were outbreak reports, observational studies and interventional studies relating to infection...
control and case series. Patient-level outcomes of interest were rates of colonisation versus infection and rates of morbidity and mortality. Study-level outcomes were source and duration of outbreak, culture results, cure rates, adverse events and interventions. An outbreak was defined as cases of ESBL colonisation or infection in excess of what would normally be expected for the NICU concerned. Colonisation was classified as acquisition of an ESBL-producing organism not requiring antimicrobial treatment, and infection as acquisition of ESBL-producing organism requiring antimicrobial treatment. Mortality was classified as attributable or non-attributable to ESBL infection.

Information sources
Searches were conducted in the PubMed, MEDLINE, SCOPUS and Cochrane databases. The last search was performed on 17 July 2014. The search strategy in full was registered with the PROSPERO database, and is available at http://www.crd.york.ac.uk/PROSPEROFILES/10744_STRATEGY_20140618.pdf. The first search was for ‘neonatal’ and ‘outbreak’ and ‘extended spectrum beta lactamase’. The second search was for ‘neonatal’ and ‘outbreak’ and ‘ESBL’. The third search was for ‘NICU’ and ‘outbreak’ and ‘extended spectrum beta lactamase’. The fourth search was for ‘NICU’ and ‘outbreak’ and ‘ESBL’. All databases were interrogated with the same search strategy, and no filters were applied. The references of included studies were reviewed for additional relevant articles.

Study selection
The studies identified by the search strategy were collated and duplicates were removed. One investigator (RJD) screened the titles and abstracts. Studies meeting the eligibility criteria or which did not provide sufficient information in the title and abstract were retrieved for further review. Retrieved studies were divided into two groups and each group of papers was reviewed by a team of two investigators (RJD and MB or PJMS and MM) to determine eligibility using a standardised assessment tool based on the study protocol. The full text of each article was reviewed by only one investigator, but decisions on eligibility were reviewed by a second investigator. Any disagreements on eligibility were discussed with all study investigators and resolved by consensus. Papers that reported data from multiple outbreaks of ESBL organisms as if they were one large outbreak were excluded.

Data extraction
Data extraction was performed using a standardised assessment tool adapted from the ORION guidelines on reporting criteria for outbreaks of nosocomial organisms. Key outcome data were extracted by one investigator and checked by a second investigator for completeness and accuracy. None of the authors of the included articles were contacted for further information or results. For binary and count outcomes (colonisation, infection, mortality), we extracted only numerical data as we could not extract accurate denominator data (eg, total number of neonates admitted to unit during outbreak period) from most studies. Continuous outcomes such as duration of outbreaks were extracted as simple numeric data and means and IQR were then calculated.

Risk of bias assessment
For each study, a risk of bias assessment was performed by one investigator using a tool based on ‘The Cochrane Collaboration’s tool for assessing risk of bias in randomised studies’, which we adapted to assess bias in outbreak reports. Risk of bias across all studies was discussed at a meeting of all study authors prior to preparation of the manuscript to determine if there was a consistent bias across the studies, which could impact on the systematic review.

Data analysis
Data analysis was planned on pooled data for all ESBL producers and then in subgroups by organism type.

RESULTS
Study selection
The search of databases yielded 480 studies, and review of references yielded a further 26 studies. Following removal of duplicates, 136 articles remained for screening. After screening, 103 underwent full review; 27 of these were excluded, most commonly due to a non-ESBL-producing organism. The remaining 75 studies were eligible for inclusion in the qualitative analysis (see online supplementary table S1). Only 26 of these studies reported sufficient data to be included in the quantitative analysis (figure 1).

Study characteristics
There were 71 studies published in English and 2 each in Spanish and French. Europe accounted for the highest number of publications with 36% (n=27) followed by Africa 15% (n=11), North America and Asia with 13% each (n=10), South America 12% (n=9), the Middle East 8% (n=6) and Australia 3% (n=2). Seventy-two of the studies were retrospective and consisted mainly of outbreak reports or molecular typing studies. Five of the retrospective studies performed analytical epidemiological investigations using case–controls. Two were prospective molecular epidemiological typing studies, and one was a prospective observational cohort study. Forty-nine of the studies (65%) were published in the past decade.

Risk of bias within studies
Only 26 studies were included in the quantitative analysis as the remainder did not provide complete data on key outcomes such as rates of colonisation, infection and death. Studies that focused on ESBL typing and molecular epidemiology frequently provided limited clinical data (see online supplementary table S2).

Morbidity data were reported infrequently and primarily concerned the site of infection (meningitis, bacteraemia, etc) as opposed to infant outcomes or adverse events, and could not be analysed further. An unexpectedly high reporting rate for environmental and staff screening results may reflect publication bias. Significant heterogeneity in infection and mortality rates was observed with clusters of studies reporting either large total numbers with no deaths or low total numbers with high mortality, which may be partly accounted for by differences in screening policies.

Study level outcomes
The median duration of an outbreak was 6.2 months (IQR 2.0–7.5 months). Five studies attempted to define the duration of carriage of ESBL organisms: two reported carriage of an unspecified duration; one reported duration of up to 1 month and another up to 1 year. The remaining study found no evidence of chronic carriage.

Predisposing factors were reported in 25 studies and included understaffing (22%), high antibiotic consumption (19%), a previous outbreak in the unit (15%), poor infection-control procedures (15%) and overcrowding (11%). The source of the
outbreak was identified in only 32 studies, and was most frequently due to admission of an identified index case with subsequent horizontal spread, transfer from contaminated equipment or environmental surfaces, transmission by healthcare workers or a combination of these factors (figure 2).

Sixty studies reported at least one intervention used to control the outbreak, with a mean of four different interventions per outbreak. Improvement of hand hygiene compliance, environmental screening and staff screening were the most commonly reported interventions. Infrequently reported interventions (single reports) included breast milk pump decontamination, breast milk pump restriction, introduction of a central venous catheter care bundle, adoption of single use ointments, restricted use of immersion baths and visitor restrictions (figure 3).

**Patient-level outcomes**

Outbreak size (total numbers colonised and infected) ranged from 4 to 252. The 75 studies involved in total 1185 colonised children and 860 infected children. In the 860 children with infections, there were 139 deaths, giving an all-cause mortality rate of 16%. Two of the deaths were reported as due to causes other than infection. The organisms responsible were *Klebsiella* spp in 69% (n=52), *Enterobacter* spp in 13% (n=10), *E. coli* in 11% (n=8), *Salmonella* spp in 4% (n=3) and *Serratia marcescens* in 3% (n=2). Single studies reported simultaneous outbreaks of *Klebsiella* spp/*E. coli* and *Klebsiella* spp/*S. marcescens*. In studies where molecular typing was performed (n=36), 18 reported identification of one ESBL enzyme family, 12 studies reported two enzyme types and 6 studies reported three enzyme types. The most frequently reported ESBL types were SHV for *Klebsiella* spp, CTX-M for *Enterobacter* spp and TEM for *E. coli*. There were single reports of *E. coli* and *Klebsiella pneumoniae* carrying OXA-1 and of *Enterobacter* spp carrying CTX-M.

**Figure 1** PRISMA flow diagram. ESBL, extended spectrum beta-lactamase; NICU, neonatal intensive care unit.

**Figure 2** Frequency of reported source of outbreaks (n=75). IV, intravenous. HCW, health care workers.
and SHV, respectively. The third study reporting *Salmonella* spp did not perform molecular typing and neither did the two studies reporting *S. marcescens* (table 1).

There was no significant pattern observed in variation in ESBL enzyme type by geographical location.

**Data synthesis**

Quantitative analysis was limited to the 26 studies that reported complete data on colonisation, infection and mortality rates as the other 49 studies failed to report at least one of these key variables. Meta-analysis of the rates of infection versus colonisation was performed using MedCalc software (V15.4). We also performed meta-analysis on the numbers of infants with infections who died; one of the 26 studies was excluded as there was no case of infection. We did not perform subgroup analysis as subgroup sizes for organisms would be small, and statistical analysis would be potentially misleading. Results of the pooled mortality analysis indicate substantial heterogeneity (I²=77%, df=24, p<0.0001); so, the random effects meta-analysis is a more precise measure of the average mortality of infected than the fixed effects model. The severe heterogeneity encountered in analysis of rates of infection versus colonisation (I²=93.7%, df=25, p<0.0001) means caution should be used while interpreting the results, which may reflect differences in screening policies or capabilities in different units rather than a true measure of attack rates in colonised infants (figure 4).

**DISCUSSION**

Meta-analysis indicates ESBL-producing *K. pneumoniae* is the most frequently implicated pathogen in outbreaks, and a diverse array of ESBL enzyme types were identified regardless of geographical location. Random effects meta-analysis of pooled data from 25 studies indicates a high mortality rate in infected infants (31%, 95% CI 20% to 43%). Rates of infection in colonised infants are more difficult to interpret, given the variations in screening policies and thus detection of colonised cases.

The origin of the outbreak was not identified in most studies, although suboptimal hand hygiene practices with horizontal dissemination of organisms from an infant gut reservoir was frequently postulated as the likely cause. Where the source was identified, poor hygienic practices were often contributory. Enhanced infection-control procedures, including better hand hygiene compliance, were the most common interventions implemented to terminate outbreaks. Screening of staff or the NICU environment was performed frequently and the implication of transfer of organisms from staff or contaminated equipment to neonates was correspondingly high.

Factors related to suboptimal hygiene standards, reported as a risk factor predisposing to outbreaks, included understaffing, overcrowding and previous outbreaks of multidrug-resistant organisms. One unifying factor of the mentioned risks is understaffing in the NICU. Overcrowding, relative understaffing and deficiencies in hand hygiene practices all facilitate the spread of nosocomial infection. The Framework of Practice published by the British Association of Perinatal Medicine in 2014 recommended nurse:patient ratios of 1:1 for NICU patients, 1:2 for high dependency and 1:4 for special care infants, with clear criteria for each group. In recent times, widespread shortages of nursing staff and financial constraints have left many NICUs understaffed and under-resourced. Although understaffing was the single most frequently reported predisposing factor contributing to ESBL outbreaks in this review, an increase in staffing rates was one of the least frequent interventions (5%, n=3).

Maternal colonisation with ESBL organisms has been previously identified as the most important risk factor for VLBW infant colonisation. We identified that admission of a single colonised infant to an NICU was the most commonly reported source of an ESBL outbreak in an NICU.

Maternal and infant screening for ESBL carriage has the potential to quickly detect infant colonisation and allow for enhanced infection-control precautions. At present, UK guidelines do not recommend routine screening for ESBL colonisation in neonates. Better reporting of NICU ESBL outbreaks could help address the question of whether screening would prevent or limit future outbreaks. Regulatory authorities could...
consider imposing mandatory confidential reporting of ESBL outbreaks in NICUs. Mandatory reporting, using standardised ESBL outbreak report forms modelled on the ORION criteria, would help reduce the incidence of missing data encountered in this review, and could help inform policies on screening and outbreak control interventions.

Strengths of this review included a comprehensive literature search that retrieved 76 publications, total 2104 affected infants, and reporting of microbiological data such as ESBL enzyme types was robust. Limitations included the language limits employed in the search and infrequently or inconsistently reported key clinical data such as screening policies and evidence for the effectiveness of interventions. The review highlights the need to evaluate the effectiveness of addressing staffing shortages in preventing or terminating outbreaks and for comprehensive reporting of clinical data to allow evaluation of the merits of screening for ESBLs in NICUs.

Competing interests None declared.

Provenance and peer review Not commissioned; externally peer reviewed.

Data sharing statement The study protocol in full is available on the website of PROSPERO, the international prospective register of systematic reviews. Unpublished data, that is, the dataset extracted from the papers included in the review will be made available on reasonable request to the corresponding author.

Figure 4  (A) Meta-analysis of proportion of infants colonised who developed infection. (B) Meta-analysis of proportion of infants with infections who died.
REFERENCES


