Infection and sudden unexpected death in infancy: a systematic retrospective case review


Summary

Background The cause and mechanism of most cases of sudden unexpected death in infancy (SUDI) remain unknown, despite specialist autopsy examination. We reviewed autopsy results to determine whether infection was a cause of SUDI.

Methods We did a systematic retrospective case review of autopsies, done at one specialist centre between 1996 and 2005, of 546 infants (aged 7–365 days) who died suddenly and unexpectedly. Cases of SUDI were categorised as unexplained, explained with histological evidence of bacterial infection, or explained by non-infective causes. Microbial isolates gathered at autopsy were classified as non-pathogens, group 1 pathogens (organisms usually associated with an identifiable focus of infection), or group 2 pathogens (organisms known to cause septicemia without an obvious focus of infection).

Findings Of 546 SUDI cases, 39 autopsies were excluded because of viral or pneumocystis infection or secondary bacterial infection after initial collapse and resuscitation. Bacteriological sampling was done in 470 (93%) of the remaining 507 autopsies. 2079 bacteriological samples were taken, of which 571 (27%) were sterile. Positive cultures yielded 2871 separate isolates, 484 (32%) of which showed pure growth and 1024 (68%) mixed growth. Significantly more isolates from infants whose deaths were explained by bacterial infection (78/322, 24%) and from those whose death was unexplained (440/2306, 19%) contained group 2 pathogens than did those from infants whose death was explained by a non-infective cause (262/1628, 16%) or unexplained. Significantly more cultures from infants whose deaths were unexplained contained Staphylococcus aureus (262/1628, 16%) or Escherichia coli (93/1628; 6%) than did those from infants whose deaths were of non-infective cause (S aureus: 19/211, 9%; difference 7·1%, 95% CI 2·2–10·8, p=0·005; E coli: 3/211, 1%, difference 4·3%, 1·5–5·9, p=0·003).

Interpretation Although many post-mortem bacteriological cultures in SUDI yield organisms, most seem to be unrelated to the cause of death. The high rate of detection of group 2 pathogens, particularly S aureus and E coli, in otherwise unexplained cases of SUDI suggests that these bacteria could be associated with this condition.

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Introduction

Sudden infant death (also known as cot death) remains one of the most common presentations of post-neonatal infant death in the UK, with many theories regarding its pathogenesis.1 Sudden unexpected death in infancy (SUDI) is defined as the sudden and unexpected death of an infant aged less than 1 year. Such deaths are a heterogeneous group, including those in which a careful review of the death scene and a meticulous post-mortem examination will disclose a cause of death, and those that remain unexplained even after such examination. Of the deaths that remain unexplained, death might be classified as sudden infant death syndrome (SIDS) if the infant died during sleep.2 In 2005, there were 268 unexplained infant deaths in England and Wales at a rate of 0·41 deaths per 1000 livebirths, making this one of the leading categories of post-neonatal infant death.3 In England and Wales all such cases are investigated on behalf of Her Majesty’s Coroner (HMC) by a detailed post-mortem examination done by a paediatric pathologist in accordance with a suggested autopsy protocol that incorporates a range of associated ancillary investigations,4 including post-mortem sampling for microbiological examination. However, the current protocol is based mainly on expert opinion and perceived best practice, with a paucity of published objective evidence, and meaningful and appropriate interpretation of such samples could be difficult.5–7

For many decades, underlying infection has been suggested to be a possible important mechanism in SUDI, on the basis of demographic features, autopsy findings, and results of studies reporting detection of organisms.8–10 There is no doubt that infection-related disease represents the underlying cause of death in a subgroup of SUDI,11 although the proportion of cases in which post-mortem microbiological examination provides the cause of death remains uncertain. These data are derived from small case series or regional multicentre studies, with inherent variation in sampling methodology and populations. Additionally, culture results were often used to determine the cause of death,
requiring empirical, but non-evidence based, assumptions to be made regarding their clinical importance. Furthermore, the appropriate interpretation of microbiological findings has major medicolegal importance, as evidenced by recent high court reviews of child death investigations.14,15 Our aim was to examine the role of post-mortem microbiological sampling in determining the possible cause of SUDI using data derived from a large series of SUDI autopsies done at a specialist centre.

Methods
Study population
Great Ormond Street Hospital for Children (London, UK) is a specialist tertiary referral centre for paediatric diagnosis and treatment, including post-mortem examinations done on paediatric deaths from a wide geographical area covering the south of England. We did a systematic retrospective case review of all autopsies done on behalf of HMC for the indication of SUDI at the centre between 1996 and 2005, inclusive. Local research ethics committee approval was obtained before the start of the project.

Autopsy findings were reviewed, and data abstracted and entered into a specially designed database by a paediatric pathologist (MAW) in accordance with strictly defined criteria (available on request). For each organ system, macroscopic and microscopic findings were recorded separately and all microbiological data were incorporated into the database. All autopsies were done by paediatric pathologists in accordance with a common protocol, which included macroscopic examination and dissection, histological sampling of all major organs, and ancillary investigations such as virological and microbiological sampling, similar to the Kennedy protocol.4 Routine microbiological samples were obtained from cardiac blood, cerebrospinal fluid (CSF), lung, and spleen, with additional samples where specifically indicated.

In all cases, the final cause of death was classified into three separate categories with criteria specifically set up for this study. The categories were defined with no reference to the results of post-mortem microbiological sampling to avoid interpretation bias. Cases of SUDI were categorised as: unexplained death (ie, deaths that remained unexplained after post-mortem examination, with no significant abnormal findings at autopsy, excluding the results of microbiological analysis); explained with histological evidence of (bacterial) infection (histological evidence of pneumonia, meningitis, or any other focus of acute neutrophilic inflammation normally associated with a bacterial infective process and deemed sufficient to be responsible for the death); and death explained by non-infective causes (a clear non-infective cause of death—eg, congenital heart disease, head injury, or deaths due to other accidents—without any histological evidence of infection). Deaths potentially due to viral or non-bacterial infections (eg, fungal infections and infection with Pneumocystis jirovecii) were excluded from this analysis, as were all cases of SUDI that were explained by non-infective causes but in which there was evidence of a superimposed, secondary, or incidental bacterial infection, often as a result of a period of post-resuscitation survival. These methods allowed the determination of the proportion of cases of SUDI in which microbiologically detectable infection could be present in the absence of associated histological abnormalities shown by routine sampling. All cases were identified in the database by study number only.

For the purposes of this study, microbial isolates were categorised into one of three groups: non-pathogens (organisms that are usually non-pathogenic, accepting the possibility that in specific clinical settings, especially in immunocompromised patients, these organisms can be pathogenic), group 1 pathogens (organisms which, when isolated in blood in life in the investigation of a bacteraemic or septicaeic illness, would be interpreted as significant or probably significant, but which would usually be associated with an identifiable focus of infection; eg, group C beta-haemolytic streptococcus, Candida spp, coliform, Enterobacter spp, Enterococcus spp, Haemophilus influenzae [not type b], Klebsiella spp, Proteus spp, Pseudomonas aeruginosa, Salmonella spp, Serratia marcescens), and group 2 pathogens (organisms which, when found in blood in life in the investigation of a bacteraemic or septicaeic illness, would be interpreted as significant or possibly significant and which are recognised to cause septicaemia or death without there necessarily being an identifiable focus of infection; eg, group A beta-haemolytic streptococcus, group B beta-haemolytic streptococcus, Escherichia coli, H influenzae type b, Listeria monocytogenes, Neisseria meningitidis, Staphylococcus aureus, Streptococcus pneumoniae). A more complete list of organisms in each category can be found in the webappendix. Only specimens obtained during the post-mortem examination were included, and microbiological findings were restricted to bacteriological culture results.

Statistical analysis
We examined differences in the frequencies of isolated microbiological organisms between groups with the χ² and comparison of proportion tests; differences in distributions of findings between groups were analysed with the Mann-Whitney U test. Analyses were done with StatsDirect version 2.6.2.

Role of the funding source
The study sponsor had no role in the design or conduct of the study, in the collection, analysis, or interpretation of the data, or in the writing of the report. All authors had access to all the data, and had final responsibility for the decision to submit the paper for publication.
Results

1516 paediatric autopsies were done between 1996 and 2005 in accordance with the common protocol, 1502 (99%) of which were done by one of six paediatric pathologists. There were 546 autopsies of infants who had died suddenly and unexpectedly, of which 39 were excluded from further analysis because of probable or identified viral or pneumocystis infection or documented development of a secondary bacterial infection after initial collapse and resuscitation. Of the remaining 507 autopsies, 379 were unexplained, 72 were explained by non-infective cause, and 56 were explained by bacterial infection. Infants who died because of bacterial infection were significantly older than were infants whose death was unexplained (median age 112 [IQR 77·5–222] days vs 67 [36–143] days; p<0·0001), but there were no significant differences in age between infants whose death was unexplained and those who died from non-infective causes (97 [27·5–194] days; p=0·29); likewise, there was no difference in age between those who died from non-infective causes and those who died from bacterial infection (p=0·14). Clinical symptoms of fever or upper respiratory tract infection—including descriptive terms such as snuffles, cold, and cough—were recorded by parents or carers in a significantly higher proportion of infants who died from bacterial infection were significantly older than were infants whose death was unexplained (median age 112 [IQR 77·5–222] days vs 67 [36–143] days; p<0·0001), but there were no significant differences in age between infants whose death was unexplained and those who died from non-infective causes (97 [27·5–194] days; p=0·29); likewise, there was no difference in age between those who died from non-infective causes and those who died from bacterial infection (p=0·14). Clinical symptoms of fever or upper respiratory tract infection—including descriptive terms such as snuffles, cold, and cough—were recorded by parents or carers in a significantly higher proportion of infants who died from bacterial infection (23/56, 41%) than in those whose death was unexplained (7/379, 2%; difference 21·5%, 95% CI 8·9–35·2; p=0·0005) or in infants who died from non-infective causes (9/72, 13%; difference 8·6%, 13·5–43·3; p=0·0002), but there was no significant difference in the reported frequency of such symptoms between those whose death was unexplained and infants who died from a non-infective cause (difference 7·0%, –3·2 to 14·4; p=0·14).

Post-mortem bacteriological investigations were done in 470 (93%) of the 507 autopsies. The sudden deaths that were not tested bacteriologically were mainly from the explained non-infective group in which the cause of death was apparent on macroscopic examination and thus bacteriological investigations were not warranted. Overall, 2079 individual samples were collected, including 452 cultures from blood, 440 from CSF, 435 from the lungs, and 410 from the spleen. Most cases had between four (264/470, 56%) and five (100/470, 21%) samples taken per autopsy, and there was no significant difference between the groups in the proportion of cultures from blood, CSF, lung, or spleen (χ² 4·53, p=0·81). The median interval between death and post mortem was 3 [IQR 2–4] days, with no significant differences between the three cause-of-death groups (data not shown).

Of the 2079 bacteriological samples, 571 (27%) were sterile; 1508 (73%) cultures were positive for organisms.
There were no significant differences in the proportion of sterile samples between the three groups (437/1628 [27%] for unexplained SUDI, 67/240 [28%] for infective SUDI, and 67/211 [32%] for non-infective SUDI; \( \chi^2 2.29, p=0.10 \)). 484 (32%) of the positive cultures grew single isolates (a pure growth of one species), whereas 1024 (68%) samples were polymicrobial (showing a mixed growth of organisms, with a total of 2387 polymicrobial isolates from all sites; webfigure 1). 1241 (43%) of the 2871 microbial isolates were non-pathogenic and 1630 (57%) were potentially pathogenic, of which 1085 were group 1 pathogens and 545 were group 2 pathogens.

There was no significant difference in the proportion of non-pathogenic isolates from infants whose death was unexplained and from infants who had died of non-infective causes (table; difference 2.4%, 95% CI –4.2 to 8.8; \( p=0.46 \)); likewise, there was no significant difference in the proportion of group 1 pathogens isolated from infants who died of non-infective causes and from those whose death was explained by bacterial infection (table; difference 6.9%, –1.4 to 15.1; \( p=0.10 \)). However, there were significantly more isolates of group 2 pathogens from infants whose death was explained by bacterial infection than from those whose death was unexplained (difference 5.1%, 0.5–10.3; \( p=0.03 \)) and from those whose death was explained by non-infective causes (difference 13.1%, 6.9–19.2; \( p<0.0001 \)).

Furthermore, there were significantly more isolates of group 2 pathogens from infants whose death was unexplained than from those whose death was explained by non-infective causes (difference 8.0%, 3.2–11.8; \( p=0.001 \)). Similarly, a significantly greater proportion of infants whose death was unexplained had at least one group 2 pathogen isolated from any site than did those whose death resulted from a non-infective cause (50% [181/365] vs 26% [14/53]; difference 23.2%, 9.1–34.6; \( p=0.001 \)).

There were significantly more samples with pure growths of a group 2 pathogen from infants whose death was explained by bacterial infection (23 of 240 samples, 10%) than from those whose death was unexplained (58 of 1628 samples, 4%; difference 6.0%, 95% CI 2.7–10.5; \( p=0.0001 \)) or from those whose death was explained by a non-infective cause (5/211, 2%; difference 7.2%, 95% CI 3.0–11.9; \( p=0.0009 \)), but there was no difference in the proportion of pure growths of a group 2 pathogen from infants whose death was unexplained (58/1628, 4%) and from those whose death was explained by non-infective causes (5/211, 2%; difference 1.2%, –1.9 to 2.9; \( p=0.43 \)).

However, there were significantly more mixed cultures containing group 2 pathogens isolated from infants whose death was caused by bacterial infection (49/240, 20%) and from infants whose death was unexplained (317/1628, 20%) than there were from infants whose death was of a non-infective origin (20/211, 10%; difference 10.9%, 4.4–17.5; \( p=0.001 \)) vs bacterial cause; and difference 10.0%, 5.0–13.9; \( p=0.0002 \) vs unexplained; wefigure 2). Specific group 2 pathogens isolated included \textit{S aureus}, \textit{E coli}, groups A and B beta-haemolytic streptococcus, \textit{S pneumoniae}, and \textit{N meningitidis}, with the greatest proportion of samples yielding \textit{S aureus} and \textit{E coli} from infants whose death was unexplained (figure 2), especially from the lung and spleen (figure 3).

**Discussion**

Although more than 70% of the post-mortem bacteriological samples that were analysed grew microbial organisms when cultured, we found that significantly more organisms that were potentially pathogenic were isolated from infants whose sudden, unexpected death could not be explained than from infants whose death was of non-infective cause. Although the reasons for this difference are unclear, our findings suggest that microbes...
or microbial products could be related to the pathogenesis of a proportion of unexplained SUDI. We must now investigate the pathophysiological mechanism involved in these cases.

Infection has been identified previously as the probable cause of death in a subgroup of SUDI, usually on the basis of a combination of microbiological and histopathological findings. However, if potentially pathogenic organisms are identified by culture in the absence of a tissue reaction recognisable with standard histological examination techniques, definitive determination that infection is the cause of SUDI is at present impossible. Hence, the role of infectious agents as a causative or potential contributing factor in the pathogenesis of SUDI has remained controversial. One reason for this emerged during the course of this study. Despite more than 70% of SUDI post-mortem bacteriological samples showing the presence of microbial organisms, there were no differences in the overall rates of sterile cultures between the various cause-of-death groups. This finding would indicate that the mere detection of organisms on post-mortem culture is not in itself diagnostic of the cause of death at autopsy. In this study, over 40% of positive cultures showed the presence of probable non-pathogenic organisms, while around 60% contained potential pathogens. Although histological findings suggestive of infection were associated with increased rates of isolation of group 2 pathogens, these data also indicate that such pathogens could be present in microbiological cultures in around a quarter of deaths with a known non-infective cause—eg, accidents or congenital heart disease—confirming that the mere detection of pathogens does not necessarily indicate the cause of death, possibly explaining why the interpretation of investigations into the potential role of infectious causes of SUDI has been so problematic. Nevertheless, the prevalence of group 2 pathogens was significantly greater in both the bacterial infection and unexplained SUDI groups than in the non-infective group, suggesting that infection or colonisation with a group 2 pathogen could be of pathophysiological importance in a subset of otherwise unexplained SUDI.

At present, there is no unifying theory for the pathogenesis of unexplained SUDI, but data derived from epidemiological, pathological, genetic, and animal studies suggest that the cause, although probably multifactorial, could be infection-related. This hypothesis is based on the natural variation in the incidence and age distribution of unexplained SUDI, the more frequent isolation of potentially pathogenic organisms from the upper respiratory tract of such infants, and the possible synergistic effects of common risk factors—eg, prone sleeping (associated with a reported increased prevalence of upper respiratory tract bacterial colonisation) and smoking (associated with a possible increased effect of bacterial toxins). Furthermore, certain genetic polymorphisms in immunoregulatory genes related to infection might increase the infant’s vulnerability to SUDI. Here, the occurrence of *S aureus* and *E coli* was greater in the group of infants whose death was otherwise unexplained, especially in the lungs and spleen, than in those whose death was explained; both organisms have been implicated previously in the potential pathogenesis of SUDI, possibly via a toxigenic pathway.

Advantages of the methodology of our study include the large sample size and the fact that all the autopsies were done by specialist paediatric pathologists at one tertiary centre according to a common post-mortem protocol. Most importantly in relation to the validity and strength of our findings, no a-priori interpretation of the significance of any microbiological finding was made before analysis, thus removing potential subjective bias in the interpretation of the clinical relevance of post-mortem microbiological results. The current challenge, however, remains to determine the optimum methods for differentiating those cases that are truly infection related from those in which the isolates might represent simple contaminants, post-mortem translocation, or incidental colonisation. Contamination during sampling at autopsy could be minimised with good technique using appropriate antiseptic preparations and sterile instruments. Empirical evidence suggests that contamination is more likely to result in a mixed growth, but there is little published evidence to confirm this at autopsy, and our results do not support this notion. Agonal spread is essentially a hypothetical concept, in which the mucosal integrity is postulated to be compromised by peri-mortem hypoxia or ischaemia, thereby allowing bacteria to enter the tissues or blood around the time of death. Post-mortem translocation or transmigration occurs as part of the normal putrefaction process, in which bacteria that have colonised the mucosal surfaces in life start to invade the body after death; however, such spread is unlikely to be a significant problem if bodies are stored in appropriately refrigerated units.

Many of the isolates in our study could represent incidental colonisation before death or might represent organisms that have colonised the respiratory tract or other sites around the time of death or during resuscitation. However, although one or more of the mechanisms described above could theoretically be responsible for the high detection rate of bacteria at post-mortem examination, it is difficult for such mechanisms to explain the finding of a significantly higher prevalence of pathogenic bacteria in the unexplained SUDI group compared with the non-infective group, especially since there were no obvious differences between the groups in terms of post-mortem interval or sampling sites, for example, to account for these findings. The absence of histological evidence of infection in these cases makes a diagnosis of classical infection unlikely, but it is possible that microbial products, rather than...
direct microbial invasion, could in some way lead to SUDI. Colonisation with organisms such as *S aureus* and *E coli* could produce toxins which might affect the infant.  

However, many of the known toxins cause syndromes with characteristic clinical and histological features, which were, by definition, not present in these cases. The detection of these bacteria could be an epiphenomenon indicative of another underlying mechanism of death, such as overheating or impaired infant arousal responses, which are epidemiologically associated with SUDI and which might also predispose to secondary abnormal bacterial growth. This inter-relation is further supported by the association between epidemiological risk factors for SUDI such as prone sleeping and increased nasopharyngeal isolates of bacterial organisms.

**Contributors**

NJS and JCH designed the study. NJS, PEL, and MAW collected the data. MAW, NJK, JCH, and NJS analysed the data. MAW, NJK, JCH, MM, and NJS wrote the manuscript. All authors saw and approved the final version of the manuscript.

**Conflict of interest statement**

We declare that we have no conflict of interest.

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**References**


